

1 **Blood Lactate concentrations during rest and exercise in people with Multiple**  
2 **Sclerosis: A systematic review and meta-analysis**

3

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22 statistical analysis

23 Abstract

24

25 Background: Multiple Sclerosis (MS) is a chronic disorder which irreversibly damages axons within  
26 brain matter. Blood lactate concentration could be a biomarker of MS onset and progression, but no  
27 systematic review has yet sought to confirm or dispute the elevation and biomarker potential of blood  
28 lactate in people with MS (PwMS) or to consolidate understanding of lactate production during  
29 exercise in PwMS.

30 Objective: To perform a systematic review and meta-analysis on blood lactate in PwMS during rest  
31 and exertion compared to Healthy Controls (HC) and following chronic exercise intervention.

32 Methods: A systematic search of six electronic databases (PubMed, CINAHL, Science Direct, Cochrane  
33 Library, SPORTDiscus and PEDro) was performed on 10th April 2020. Mean, standard deviation and  
34 sample size for lactate measures at rest and during exercise were pooled to determine overall effect  
35 size using a random effects model. The 20-point Appraisal tool for Cross-Sectional Studies was utilised  
36 to assess study quality and inherent risk of bias. To qualify for inclusion, studies had to include human  
37 adults (>18 years) with a confirmed clinical diagnosis of MS, be published in English, have undergone  
38 peer review, report absolute blood lactate values for data extraction, and if involving testing  
39 during/after exercise, to do so during bilateral exercise methods.

40 Results: 18 studies were qualitatively analysed and 15 studies quantitatively analysed. Outcome data  
41 was available for 1986 participants ( $n_{MS} = 1129$ ). A total of 7 papers tested blood lactate during rest  
42 ( $Lactate_{REST}$ ), 7 papers tested during sub-maximal intensity exercise ( $Lactate_{SUB-MAX}$ ), and 8 papers  
43 tested during maximal intensity exercise ( $Lactate_{MAX}$ ). Meta analyses showed elevated  $Lactate_{REST}$  and  
44 reduced  $Lactate_{MAX}$  in PwMS compared to HC, higher  $Lactate_{MAX}$  in lower EDSS-scoring PwMS  
45 compared to higher EDSS-scoring PwMS, and that  $Lactate_{SUB-MAX}$  decreases and  $Lactate_{MAX}$  increases in

46 PwMS following a chronic exercise intervention. Qualitative analysis reported Lactate<sub>REST</sub> to be  
47 reduced in PwMS following a chronic exercise intervention.

48 Conclusions: Lactate<sub>REST</sub> is elevated in PwMS compared to HC. Lactate<sub>MAX</sub> is lower in PwMS compared  
49 to HC and lower still in higher compared to lower EDSS-scoring groups of PwMS. Chronic exercise  
50 interventions have the potential to reduce Lactate<sub>SUB-MAX</sub> for a given power output and increase  
51 Lactate<sub>MAX</sub> in PwMS compared to baseline values. Lactate<sub>REST</sub> may be reduced in PwMS following a  
52 chronic exercise intervention but more research is required for confirmation. The results of this review  
53 were limited by small sample sizes and number of studies available for each testing condition, limited  
54 data available for potentially confounding/correlating factors (eg. VO<sub>2</sub> and power output) as well as  
55 heterogeneity of methodology adopted across studies, often due to lactate testing being a secondary  
56 outcome measure.

57

58 PLS:

59 Lactate levels in the blood are different during rest and at intense exercise levels in people with  
60 Multiple Sclerosis (MS) compared to healthy counterparts, with people with MS showing a smaller  
61 jump in lactate during intense exercise from a higher resting level. After exercising for at least 3  
62 months, blood lactate levels during exercise may become more similar to the levels seen in people  
63 without Multiple Sclerosis, but more research is required to give a clearer picture of this. We can  
64 hopefully use blood lactate in future to measure the progression of MS in an individual as well as the  
65 effectiveness of their exercise programme.

66

67 1.0 Introduction

68

69 Multiple Sclerosis (MS) is a chronic disorder characterised by perivenular inflammatory lesions which  
70 ultimately result in demyelinating plaques, oligodendrocyte damage, and irreversible damage of axons  
71 within the grey and white brain matter [1], resulting in a progressive loss of function across the  
72 spectrum of neurological control. Once diagnosed, the phenotype of MS will be established through  
73 the pattern of progression into one of 3 types: relapsing remitting (RR), primary progressive (PP), and  
74 secondary progressive (SP). Since revision of MS classifications in 2013, the term chronic progressive  
75 (CP) was dropped for the more specific PP and SP, and progressive relapsing (PR) was dropped for PP-  
76 active [2].The extent to which loss of function has occurred is measured by clinicians using the  
77 Extended Disability Status Scale (EDSS); an incremental rating from 0 (no observable symptoms) to 10  
78 (death due to MS), with the total score consisting of a combination of the number and the severity of  
79 affected functional systems [3].

80

81 Lactate is the base form of lactic acid and is produced within the human body from the reversable  
82 reduction of pyruvate during anaerobic glycolysis within the cytoplasm of the cell [4]. Increases in  
83 lactate concentration occur during conditions of increased energy demand or reduced oxygen  
84 availability. Elevated lactate has been observed using MRI in acute plaques within brain tissue in  
85 patients with MS and is hypothesized to be a sign of affected mitochondrial function in MS or  
86 increased glycolysis during the inflammatory processes of demyelination [5], though precise causes  
87 are as yet unknown. There has also been evidence presented suggesting that PwMS present higher  
88 levels of lactate in blood plasma and cerebrospinal fluid at rest compared to healthy controls (HC) and  
89 in correlation with EDSS [6,7], cementing further the idea that elevated lactate results from  
90 progression of MS. There are a number of established and newly emerging biomarkers of MS in  
91 current research aiming to fulfil at least one of three functions: predicting an individual's risk of  
92 developing MS, monitoring progression of disease course, and monitoring responsiveness to specific

93 treatments in research or routine care [8]. Measurement of blood lactate can be performed via  
94 catheterisation of arteries within the forearm but sampling at superficial capillarisation sites at the  
95 fingertip and earlobe are the most common due to minimal invasiveness and close similarities with  
96 arterial measurement [9]. With this minimal invasiveness and the relatively low cost of measurement  
97 compared to other biomarkers, the above findings could suggest there is potential that blood lactate  
98 could harbour potential as a simple and accessible biomarker for MS progression.

99

100 Lactate is typically present in healthy individuals at concentrations between 1-2mmol/L in the blood  
101 during rest [10] and rises with increasing demand of rapid ATP production, resulting in a sharp increase  
102 in concentration during higher exercise intensities [11]. When measured as part of an exercise-based  
103 study, the moment during increasing exercise intensity at which the blood lactate concentration  
104 undergoes an inflection point from a gradual to sharp increase is known as the lactate threshold [12],  
105 while the work rate at which blood lactate concentration is 4mmol/L is defined as the onset of blood  
106 lactate accumulation (OBLA) [13]. Lactate levels are often used as a measure of exercise intensity and  
107 training performance due to relatively cheap and easy measurement and reasonable correlation with  
108 anaerobic exercise intensity [14] and energy metabolism [15,16]. It has also been used to assess  
109 exercise intensity and training response in PwMS [17–19]. However, such testing has generally been  
110 performed in studies with relatively small sample sizes and limited statistical power. Understanding  
111 the broader picture of how MS impacts lactate concentrations at sub-maximal and maximal intensity  
112 exercises may help provide an overview of lactate dynamics in pwMS in daily living and during  
113 strenuous activities. It could also indicate methodological suitability of lactate analysis for measuring  
114 intervention and rehabilitation intensity and effectiveness for pwMS. This is particularly the case for  
115 when PwMS are exercising within facilities with limited availability of ergometers for power-output  
116 control and data recording, lacking clinicians for functional assessment, or should PwMS be  
117 participating in home-based interventions or remote research during times of highly restricted ethical  
118 research like those imposed during the onset of COVID-19 [20]. As such, a systematic review and meta-

119 analysis of full lactate response dynamics in PwMS is both warranted and overdue, given the potential  
120 for both simple and cost-effective blood lactate testing and potential use as a biomarker for MS  
121 progression. With this rationale, the objectives of this paper are to provide a systematic review and  
122 meta-analysis of blood lactate concentrations during resting conditions and following acute maximal  
123 intensity exercise bouts in PwMS compared to HC. Additionally, the paper will provide a systematic  
124 review and meta-analyses of the changes in blood lactate responses in PwMS during submaximal and  
125 maximal intensity exercise following chronic exercise interventions.

126

## 127 2.0 Methodology

### 128 2.1 Search Strategy

129

130 This systematic review was conducted was in accordance with the Preferred Reporting Items for  
131 Systematic Reviews and Meta-Analyses (PRISMA) statement [21]. A comprehensive search was  
132 performed to find literature examining blood lactate in people with MS, either as a primary or a  
133 secondary outcome. Six electronic databases (PubMed, CINAHL, Science Direct, Cochrane Library,  
134 SPORTDiscus and PEDro) were searched on 10th April 2020. Search terms used were as follows:  
135 “Multiple Sclerosis” AND (Lactate OR Lactic Acid OR OBLA OR glycolysis). Filters were applied where  
136 possible so that only research articles and articles that were peer-reviewed would be retrieved.

137

### 138 2.2 Inclusion and Exclusion criteria

139

140 To be included into the review, the study had to include human adults (>18 years) with a confirmed  
141 clinical diagnosis of MS; there were no limitations with respect to EDSS score or MS-type. Studies had  
142 to have undergone peer review, be reported in English, report absolute blood lactate values for data  
143 extraction, and, if involving exercise during lactate testing, had to be taken during bilateral exercise

144 methods (eg. Recumbent cycling, stepper or treadmill). In order to maximise the number of relevant  
145 studies, study types included in this review were randomized and non-randomized control trials, pre-  
146 post studies without controls, cross sectional studies, repeatability studies and retrospective analyses.

147

148 Excluded studies included retrospective analyses of original data already accessible by the authors,  
149 case study reports, review articles, and studies with methodology or data presentation which was  
150 incomparable with the general body of literature, e.g. studies focusing on single leg exercise in PwMS  
151 or studies which failed to provide absolute values for blood lactate concentration.

152

### 153 2.3 Data extraction and quality assessment

154

155 Results from database searches were scanned for appropriate headings and abstracts, with eligible  
156 papers subsequently imported to a bibliographic database. Duplicates were then removed and whole  
157 articles were reviewed in full for inclusion eligibility. All papers which fulfilled criteria for exclusion  
158 were removed. The data extracted from the papers included in this review included participant data,  
159 intervention protocols and outcomes. Data was extracted by author LC and verified by author NS.  
160 Study quality was assessed using the 20-point Appraisal tool for Cross-Sectional Studies (AXIS tool)  
161 [22]. The AXIS tool is designed to enable reviewers to incorporate assessment of both bias and study  
162 quality into a single score [22], whereby higher scores indicated higher quality of studies with a lower  
163 risk of bias and lower scores indicated lower quality of studies with a higher risk of bias.

164

### 165 2.4 Statistical Analysis

166

167 Meta-analyses were executed using Comprehensive Meta-Analysis (Biostat, V 2.2.064, Englewood, NJ,  
168 USA). A meta-analysis using a random-effects model was used to pool data and compare data between  
169 HC and PwMS, and the differences between lactate values before and after training interventions.

170 Mean, standard deviation and sample size for PwMS and HC for each variable of interest were used  
171 to determine overall effect size using a random effects model. Standardised mean differences were  
172 utilised in data analysis to accommodate for the variable nature between studies of the timing for  
173 lactate testing during exercise bouts and the differences in devices/processes used to measure overall  
174 blood lactate concentrations. Due to an outlier with a large sample size in participant number for  
175 Lactate<sub>REST</sub> in PwMS compared to HC [7], a one-study-removed analysis was also performed for this  
176 variable.

177

### 178 3.0 Results

#### 179 3.1. Study selection

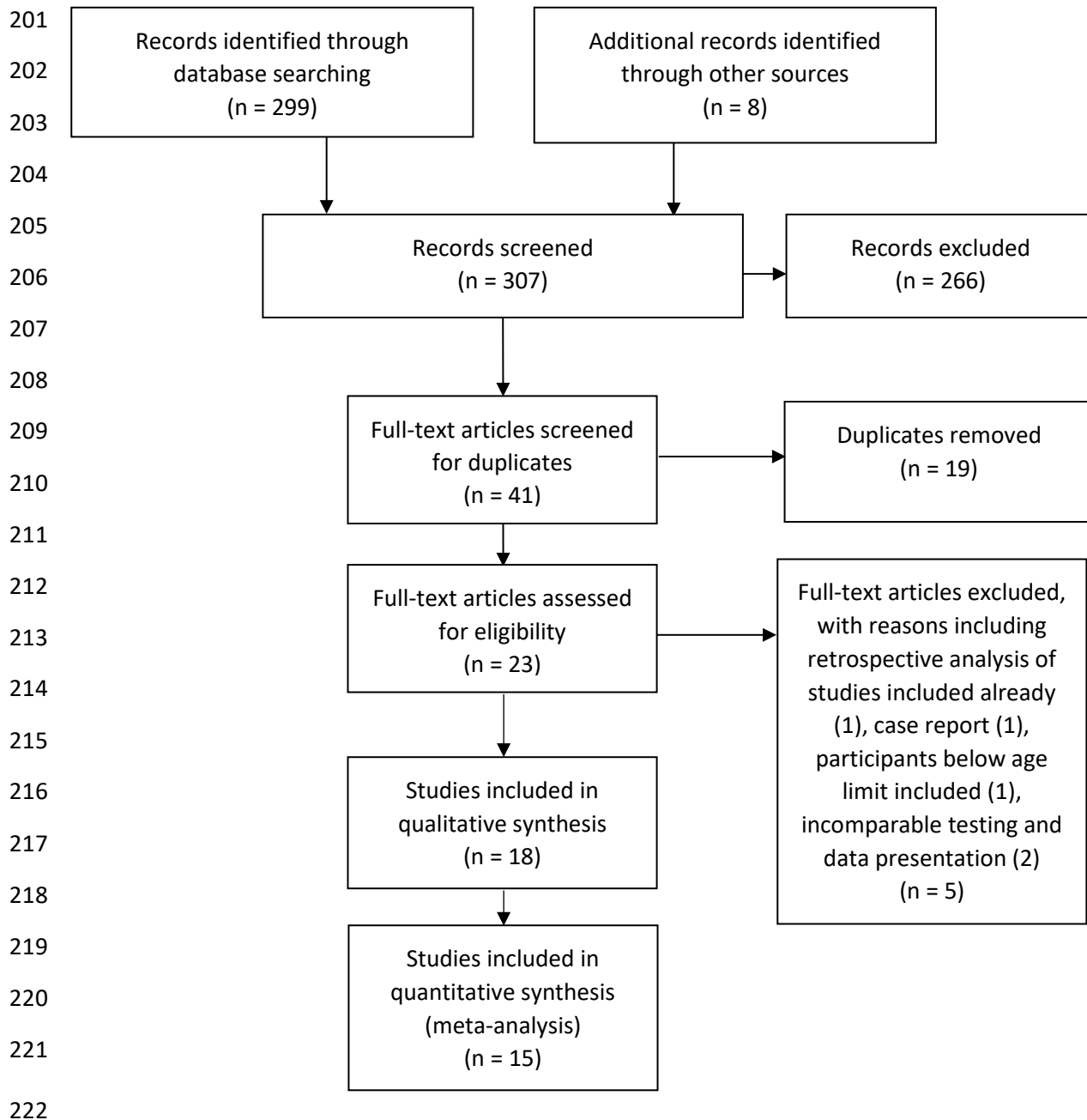
180

181 The search across the databases previous listed yielded 299 results (Figure 1). 8 papers were  
182 additionally identified from hand searching of other sources. In total, 307 papers were screened for  
183 exclusion: first by title, then by abstract and finally by full-paper screening before removing duplicates.  
184 23 eligible publications remained, from which 5 were removed: one for being a retrospective analysis  
185 of another study already included in systematic review, one for being a case report, one for including  
186 ages ranging from 10-50 years without discerning between ages above and below 18 years of age, and  
187 two for being incomparable with other studies (Figure 1). The incomparable methods reported in  
188 Figure 1 included presentation of only *change* in blood lactate without absolute values, and the use  
189 of a unilateral exercise method which measured blood lactate in lower limbs following single-leg  
190 exercise. It was for these reasons the studies were deemed incomparable to the other studies involved  
191 in the review. For the 18 papers which remained, qualitative analyses were performed with a focus on  
192 study quality, MS type EDSS, symptom exacerbations, lactate testing conditions and equipment, and  
193 lactate and power output data during rest and sub-maximal and maximal intensity exercise. 15 papers  
194 were eligible to be included in at least one meta-analysis of Lactate<sub>REST</sub> in PwMS compared to HC,  
195 Lactate<sub>SUB-MAX</sub> in PwMS before and after exercise intervention, and Lactate<sub>MAX</sub> in PwMS compared to



196 HC, before and after exercise intervention and between higher and lower scoring EDSS groups. The 3  
197 studies which were excluded from meta-analysis, 2 provided data only for Lactate<sub>REST</sub> in PwMS before  
198 and after training intervention (and as such were not numerous enough for meta-analysis) and 1  
199 provided incomplete data for the meta-analysis which would otherwise have been appropriate.

200



223 **Fig 1:** PRISMA flow diagram of literature search and review process

Table. 1 – Summary of studies included in systematic review with descriptive statistics and AXIS Score

Study	Study design	Final number of participants	Sex f/m	Age (years) ± SD	BMI (kg/m <sup>2</sup> ) ± SD	EDSS score (mean) ± SD	EDSS score (range)	Maximum EDSS score for study inclusion	MS phenotype (nType)	AXIS Score (x / 20)
Keytsman et al. (2017) [23]	pre-/post no control	n <sub>MS</sub> = 16	9/7	52.8 ± 7.2	23.5 ± 3.3	2.6 ± 1.5	NR	6	NR	14
Zaenker et al. (2018) [24]	pre-/post no control	n <sub>MS</sub> = 26 (n <sub>EDSS ≤ 3</sub> = 18 n <sub>EDSS 3.5-5</sub> = 8)	19/7 (14/4 5/3)	44.6 ± 7.9 (45.6 ± 7.5 42.6 ± 9.6)	24.22 ± 4.7 (24.33 ± 3.61 23.8 ± 6.65)	2.5 ± 1.5 (1.61 ± 0.9 4.38 ± 0.5)	0 – 5 (0 – 3 3.5 – 5)	5	22RR, 3SP, 1PP (17RR, 1PP 5RR, 3SP)	13
Wens et al. (2015) [19]	Randomised control trial	n <sub>INT</sub> = 29 n <sub>CTL</sub> = 15	17/12 8/7	48 ± 2 49 ± 2	22.6 ± 4.84 22.9 ± 5.03	3.25 ± 1.08 3.36 ± 1.55	NR	NR	17RR, 12CP 11RR, 4CP	16
Kerling et al. (2015) [18]	Non-controlled	n <sub>CMBD</sub> = 19 (30) n <sub>END</sub> = 18 (30)	24/6 20/10	42.3 ± 9 45.6 ± 11.4	24.5 ± 3.6 24.7 ± 4	2.6 ± 1.1 3.1 ± 1.3	NR	6	NR	16

Blood Lactate in Multiple Sclerosis: Syst. Review & meta-analysis

(basic group data provided for pre-drop-out, only)	randomised trial	(pre-drop-out total: $n_{MS} = 60$ )								
Hansen et al. (2015a) [25]	Randomised control trial	$n_{INT} = 14$ $n_{CTL} = 9$	10/4 5/4	$50 \pm 7$ $46 \pm 10$	$24.8 \pm 5.3$ $24.2 \pm 2.6$	$3.2 \pm 1.5$ $3.1 \pm 1.4$	NR	NR	9RR, 4SP, 1PP 6RR, 1PR, 2PP	16
Hansen et al. (2015b) [26] 2-part study	Pt 1: Cross-sectional study (MS vs. HC) Pt2: Randomised control trial	Pt 1: $n_{MS} = 37$ $n_{HC} = 15$ Pt 2: $n_{INT} = 16$ $n_{CTL} = 11$	Pt 1: 22/15 8/7 Pt 2: 10/6 6/5	Pt 1: $48 \pm 10$ $50 \pm 10$ Pt 2: $46 \pm 11$ $48 \pm 10$	Pt 1: $25.3 \pm 4.7$ $24.5 \pm 2.6$ Pt 2: $26.1 \pm 5.2$ $23.4 \pm 4$	Pt 1: $3.1 \pm 1.3$ NA Pt 2: $3 \pm 1.5$ $3 \pm 1.3$	NR	NR	20RR, 10SP, 3PP, 1PR, 3Unknown 12RR, 2SP, 1PP, 1Unknown	16

Blood Lactate in Multiple Sclerosis: Syst. Review & meta-analysis

	(MS <sub>INT</sub> vs. MS <sub>CTL</sub> )								6RR, 1SP, 2PP, 1PR, 1 Unknown	
Keytsman et al. (2019) [27]	Controlled non-randomised trial	n <sub>MS</sub> = 18 n <sub>HC</sub> = 19	6/12 5/14	41.7 ± 8.5 41.5 ± 9.9	24.8 ± 3.9 24.6 ± 2.8	1.9 ± 1.1 NA	NR	4	All phenotypes	17
Amorini et al. (2014) [7]	Cross-sectional	n <sub>MS</sub> = 613 n <sub>HC</sub> = 625	411/202 420/205	45.4 ± 12.8 44.8 ± 11.7	NR NR	3.3 ± 2.1 NA	0 - 8	NR	430RR, 153SP, 30PP	18
Cleland et al. (2016) [28]	Repeatability study	n <sub>MS</sub> = 16 n <sub>HC</sub> = 11	10/6 6/5	45.7 ± 4.9 42.9 ± 7.5	NR NR	NR NA	1 - 5	NR	9RR, 5CP, 2Unknown	16
Hansen et al. (2013b) [17]	Cross Sectional	n <sub>MS</sub> = 38 n <sub>HC</sub> = 16	22/16 9/7	48 ± 10 51 ± 10	25.1 ± 4.7 24.4 ± 2.5	3.1 ± 1.3 N/A	NR	NR	20RR, 14SP, 3PP, 1PR	14

Blood Lactate in Multiple Sclerosis: Syst. Review & meta-analysis

Hansen et al. (2013a) [29]	Cross sectional	$n_{MS} = 26$ $n_{HC} = 15$	16/10 9/6	$50 \pm 10$ $50 \pm 10$	$25 \pm 3.5$ $24.2 \pm 2.5$	$3.4 \pm 1.3$ NA	NR	NR	14RR, 9SP, 3PP	14
Juybari et al. (2018) [30]	Cross sectional	$n_{MS} = 50$ $n_{HC} = 50$	35/15 37/13	$49.6 \pm 13.5$ $45.3 \pm 15.3$	$26.1 \pm 4.9$ $25.1 \pm 4.9$	$3.8 \pm -$ NA	NR	NR	50RR	13
Kerling et al. (2014) [31]	Cross sectional	$n_{MS} = 60$ ( $n_{EDSS<3} = 38$ $n_{EDSS 3.5-6} = 22$ ) $n_{HC} = 48$	44/16 (28/10 16/6) 36/12	$44 \pm 10$ ( $42.6 \pm 10.5$ $46.6 \pm 9.9$ $40.0 \pm 13.7$ )	$24.6 \pm 3.8$ ( $25.2 \pm 4.3$ $23.6 \pm 2.4$ ) $25.2 \pm 4.6$	NR NR NR NA	0 – 6 (0 – 3 3.5 – 6)	6	NR	18
Langeskov-Christensen et al. (2014) [32]	Repeatability study	$n_{MS} = 20$ $n_{HC} = 20$	11/9 11/9	$39.9 \pm 9$ $40 \pm 8$	$24.6 \pm 4.8$ $24.1 \pm 2.7$	$2.6 \pm 1.6$ NA	NR	6	19RR, 1SP	17

Blood Lactate in Multiple Sclerosis: Syst. Review & meta-analysis

Larson et al. (2014) [33]	Cross sectional	$n_{MS} = 8$ $n_{HC} = 7$	6/2 5/2	$51.1 \pm 9.2$ $49.4 \pm 14.3$	$25 \pm 3.9$ $26.0 \pm 8.3$	NR NA	0 – 5	6.5	8RR	13
Penesova et al. (2015) [34]	Cross sectional	$n_{MS} = 19$ $n_{HC} = 19$	10/9 10/9	$30 \pm 7$ $29 \pm 7$	$23.8 \pm 4.5$ $24.4 \pm 5.3$	$1.1 \pm 0.8$ NA	NR	2	NR	16
Schlüter et al. (2017) [35]	Retrospective analysis	$n_{RR} = 25$ $n_{CP} = 41$	16/9 23/18	$40 \pm 11$ $50 \pm 8$	$24.1 \pm 5.5$ $23.1 \pm 3.3$	NR NR	0 – 3.5 4 – 6	6	25RR 14CP	15
't Eijnde et al. (2014) [36]	Cross sectional	$n_{MS} = 12$ $n_{HC} = 12$	8/4 7/5	$54 \pm 7$ $50 \pm 9$	$24.8 \pm 3.9$ $24.7 \pm 3.3$	$3.5 \pm 1.5$ NA	NR	NR	6RR, 2SP, 1PP	16

226 Abbreviations: Healthy Controls (HC), Multiple Sclerosis (MS), number ( $n_x$ ), intervention (INT), control (CTL), combined training (CMBD), endurance training (END), relapsing remitting (RR), secondary progressive

227 (SP), primary progressive (PP), chronic progressive (CP), progressive relapsing (PR), not reported (NR), not applicable (NA), standard deviation (SD).

228 3.2 Study quality

229

230 With 1 exceptional outlier with a sample size totalling 1238 participants (613 with MS) [7], 6 studies  
231 had less than 30 participants [23–25,28,33,36], 9 studies had between 30 and 70 participants [17–  
232 19,26,27,29,32,34,35] and 2 studies had between 100-110 participants [30,31]. The total number of  
233 participants reported on across all studies was 1986; 1129 were participants with MS. 5 papers failed  
234 to adequately describe their methods adequately for study reproduction [23,24,30,31,33]. Of the  
235 studies conducting intervention trials, 3 did not include a control group [18,23,24]. 1 study failed to  
236 report lactate measurement data for all groups included within the study [32]. Overall AXIS scores may  
237 be found in Table 1. The most common sources of points being deducted (and implicit increase in risk  
238 of bias) included lack of information on or categorisation of non-responders [17–19,23,25,26,28–  
239 31,33–36], lack of justification of sample size used within the study [24,29,30,33–35], potential  
240 inherent bias within or lack of description of the population from which samples were recruited  
241 [17,19,23–26,29,33,36], and inadequate descriptions of methodologies used within the studies to  
242 enable repetition in future research [23,24,28,30,33].

243

244 3.3 MS type

245

246 5 studies [18,23,27,31,34] did not specify which or how many phenotypes of MS they included in their  
247 studies. Of the of the remainder, all included a majority of participants with RR MS, 7 studies involved  
248 participants with PP MS [7,17,24–26,29,36], 8 studies involved participants with SP MS [7,17,24–  
249 26,29,32,36], 3 studies involved participants with CP MS [19,28,35], 3 studies involved 1 participant  
250 with PR MS [17,25,26] and 2 studies involved participants with an undisclosed MS phenotype [26,28].  
251 MS phenotypes included in each study are located in Table 1.

252

253 3.4 EDSS



254

255 All studies provided a description of the EDSS status of participants in the form of a group mean EDSS  
256 value, group EDSS range, and/or maximal EDSS value eligible for inclusion in the study.

257

258 12 studies reported a mean EDSS value between 2 – 4.5 [7,17–19,23–26,29,30,32,36], 2 studies  
259 reported a mean EDSS value less than 2 [27,34] and 4 studies did not report mean EDSS value  
260 [28,31,33,35].

261

262 1 study [34], focusing on people newly diagnosed with MS, limited EDSS value to  $\leq 2$  to be included in  
263 their study, 9 studies were limited to a maximum EDSS score of  $\leq 6$  or included an EDSS score range  
264 within 0-6 [18,23,24,27,28,31–33,35], 1 study included an EDSS range between 0-8 [7] and 7 studies  
265 reported neither range nor maximal inclusion values for EDSS [17,19,25,26,29,30,36]. 3 studies  
266 separated EDSS into lower and higher range brackets for direct comparison of blood lactate  
267 concentration between disease severities for PwMS, with the minimum score for higher EDSS  
268 grouping placed at 3.5 or 4 [24,31,35].

269

270 No studies involving exercise interventions  $\geq 3$  months reported reassessed EDSS scores after  
271 completion of the intervention. However, 1 study [18] stated in their discussion that EDSS remained  
272 stable in participants with MS following a 3 month exercise intervention.

273

274 Mean EDSS scores, EDSS score range, and maximum EDSS scores included in each study are located in  
275 Table 1.

276

277 3.5 Symptom exacerbation

278

279 4 studies [18,19,24,27] reported instances of participants with MS dropping out of an intervention as  
280 a result of symptom exacerbation or relapse. No studies reported on frequency of symptom  
281 exacerbation or relapse as a measure of intervention effectiveness.

282

### 283 3.6 Testing Conditions

284

285 7 papers tested for blood lactate under resting conditions [7,19,25,28,30,34,36], 7 tested during  
286 submaximal exercise [17–19,25,26,29,36] and 8 papers tested during maximal intensity exercise  
287 [18,23,24,27,31–33,35].

288

289 All 14 studies which involved lactate testing during exercise used a cycle ergometer to reach the  
290 desired exercise intensity, however exercise protocol differed between studies. Details of testing  
291 conditions can be found in Table 3 for intervention studies and Table 4 for observational studies.

292

### 293 3.7 Lactate testing equipment

294

295 1 study [33] did not describe their methods of lactate measurement but reported the study as part of  
296 another [37] which did provide a detailed description but could not itself be included in group  
297 comparison. 7 papers measured blood lactate using the Accutrend Plus lactate analyser [17,19,25–  
298 27,29,36], 2 papers with the EBIO 6666 lactate analyser [18,31], 2 reported using the Lactate Pro  
299 [28,33], 1 used the BioSen C-Line [35], 1 used the YSI 1500 Sport [32], 1 used the SUPER GL2 [34], 1  
300 used an unbranded colorimetric assay kit [30], 1 used spectrophotometric determination [7], and 2  
301 did not describe their methods of determining blood lactate [23,24]; those two who did not describe  
302 their method of lactate measurement were testing blood lactate concentration during maximal  
303 intensity exercise. Lactate testing equipment details are located in Table 3 for intervention studies and  
304 Table 4 for observational studies.

305

306 3.8 Lactate testing sites

307

308 9 studies took blood from the fingertip [17,19,25,26,28,29,32,33,36], 3 from the earlobe [18,27,35], 2  
309 from the cubital vein [7,34], and 4 provided no description of the testing site [23,24,30,31].

310

311 3.9 Timings and Workloads of Lactate Testing

312

313 Full details of individual studies' lactate measurement timing are provided in Table 3 for intervention  
314 studies and Table 4 for observational studies.

315

316 3.9.1 Lactate<sub>REST</sub>

317

318 2 studies measured resting blood lactate in participants after 15 minutes rest [7,36], with 1 study [36]  
319 sitting participants on the cycle ergometer before testing blood lactate, 3 studies measured resting  
320 blood lactate with participants on the cycle ergometer within 3 minutes or immediately prior to  
321 beginning a bout of exercise [19,25,28], 1 study tested their participants in an early morning fasted  
322 state but did not rest participants before blood sampling [34], and 1 study provided no description of  
323 resting protocol for participants prior to blood sampling [30].

324

325 3.9.2 Lactate<sub>SUB-MAX</sub>

326

327 4 studies measured blood lactate in participants at the end of submaximal intensity exercise bouts  
328 [17,19,25,29], 2 studies measured blood lactate in participants during submaximal exercise bouts  
329 [26,36], and 1 study measured blood lactate at a 50W workload during increments of a

330 cardiopulmonary exercise test (CPET) [18]. Details of relative or absolute submaximal exercise  
331 intensities are located in Table 3.

332

### 333 3.9.3 Lactate<sub>MAX</sub>

334

335 4 studies measured blood lactate at the end of a CPET [24,32], 2 studies measured blood lactate  
336 concentration both at the end of a CPET (Lactate<sub>MAX</sub>) and the highest value obtained during the  
337 recovery phase following a CPET (Lactate<sub>PEAK</sub>) [23,27], 2 studies measured blood lactate 1 minute into  
338 and every subsequent 3 minutes during a CPET [18,31], 1 study measured blood lactate 3 minutes  
339 after cessation of a CPET [33], and 1 study made multiple blood lactate tests during the recovery phase  
340 post-CPET and reported the highest as Lactate<sub>MAX</sub> [35].

341

### 342 3.10 Power output during lactate testing

343

#### 344 3.10.1 Maximum power output in PwMS compared with HC

345

346 3 studies reported maximum power output (W) reported during CPETs in PwMS compared with HC  
347 [27,31,33]. All studies reported lower absolute (W) mean power output at maximum exercise intensity  
348 in PwMS compared with HC, however only 1 study [31] calculated this difference to be statistically  
349 significant. 1 study [27] did not statistically compare power output between PwMS and HC and  
350 another [33] reported a statistically insignificant difference between PwMS and HC group means.

351

#### 352 3.10.2 Maximum power output in PwMS with low EDSS compared with high EDSS scores

353

354 4 studies reported maximum power output reported during CPETs in PwMS comparing groups with  
355 low and high EDSS scores [24,31,35]. One study [31] reported significantly higher mean absolute (W)

356 and relative (W/kg) power output at maximum intensity exercise in the low EDSS group compared  
357 with the high EDSS group. One study [35] reported significantly higher mean relative power output at  
358 maximum intensity exercise in the low EDSS group compared with the high EDSS group. One study  
359 [24] reported higher mean power output at maximum intensity exercise in the low EDSS group  
360 compared with the high EDSS group but did not statistically analyse the significance of this difference.

361

362 3.10.3 Sub-maximal power output in PwMS pre-intervention compared with post-intervention

363

364 Of the 3 studies investigating blood lactate in PwMS during sub-maximal intensity exercise before and  
365 after a training intervention [19,25,26], all studies calculated target power output for each participant  
366 using calculations based on age, sex, height and body mass. 2 studies used the same absolute (W)  
367 power output during the sub-maximal exercise test before and after the training intervention [25,26]  
368 and 1 study [19] did not specify if the estimated power output was recalculated at baseline and post-  
369 intervention. Intervention exercise prescription details are located in Table 2.

370

371 3.10.4 Maximum power output in PwMS pre-intervention compared with post-intervention

372

373 4 studies reported absolute power output (W) at maximum intensity during a CPET in PwMS before  
374 and after a training intervention [23,24,27,31]. 3 studies reported significantly increased power output  
375 in PwMS post-intervention compared to baseline [23,24,27]. 1 study reported a small increase in mean  
376 power output in both groups of PwMS (combined exercise group and endurance exercise group)  
377 following an exercise intervention but did not provide statistical analysis of these changes[31].  
378 Intervention exercise prescription details are located in Table 2.

379

380 3.11 Oxygen Consumption

381

382 3.11.1 Oxygen consumption at rest ( $VO_{2_{REST}}$ ) in PwMS compared to HC

383 3 studies reported  $VO_{2_{REST}}$  in PwMS and HC [17,28,36], however only 1 of these provided statistical  
384 comparison of these values [17]. Similar or statistically non-significant differences in  $VO_{2_{REST}}$  were  
385 reported in 2 papers [17,28], while 1 paper [36] reported a modestly higher  $VO_{2_{REST}}$  in HC compared  
386 to MS, though confirmation of the statistical significance of this was not provided.

387 3.11.2 Oxygen consumption at maximal intensity exercise ( $VO_{2_{MAX}}$ ) in PwMS compared to HC

388 4 studies reported oxygen consumption during maximal intensity ( $VO_{2_{MAX}}$ ) exercise in PwMS and HC  
389 [31–33,38]. Significantly greater  $VO_{2_{MAX}}$  values were reported in HC compared to PwMS in 3 studies  
390 [31–33], meanwhile 1 study [38] reported higher  $VO_{2_{MAX}}$  values for HC compared to PwMS but did  
391 not report the statistical significance of this difference.

392 3.11.3 Oxygen consumption at sub-maximal intensity exercise ( $VO_{2_{SUB-MAX}}$ ) in PwMS pre- vs. post-  
393 intervention

394 2 studies [25,26] reported  $VO_{2_{SUB-MAX}}$  values for PwMS pre- compared to post-exercise intervention,  
395 neither study reported significant differences in  $VO_{2_{SUB-MAX}}$  following the exercise interventions  
396 provided to their cohorts. Intervention exercise prescription details are located in Table 3.

397 3.11.4  $VO_{2_{MAX}}$  in PwMS pre- vs. post-intervention

398 3 studies [18,24,38] reported  $VO_{2_{MAX}}$  values in PwMS before and after completing an exercise  
399 intervention. 2 studies [24,38] reported significant increases in  $VO_{2_{MAX}}$  in their cohort of PwMS  
400 following an exercise intervention, meanwhile 1 study [18] reported non-significant increases in  
401 mean  $VO_{2_{MAX}}$  in their cohort of PwMS following either a combined-exercise or endurance-exercise  
402 intervention. Intervention exercise prescription details are located in Table 3.

403

404 Table 2. Summary of exercise intervention studies

Study	Study design	Lactate testing state	Intervention type -Supervision level	Intervention progression	Frequency (sessions/2wk)	Intervention duration (months)
Keytsman et al. (2017) [23]	pre-/post no control	Maximal intensity exercise	Resistance training and HIIT - Supervised	Wk 1-6: Cycling HIIT sessions gradually increased from 5x1min bouts > 5x2min bouts at 85-90% HR <sub>MAX</sub> . Wk 7-12: 5x2min bouts at 100%HR <sub>MAX</sub> and resistance training for upper and lower body altering within range of 1x10 and 2x20 repetitions, depending on individual ability.	5	3
Zaenker et al. (2018) [24]	pre-/post no control	Maximal intensity exercise	Resistance training, endurance training and HIIT	Wk1-4: 1 endurance session (intervals and continuous blend) and 1 resistance training session for lower body per week, progressing from 4x10 > 5x15 repetitions. 15-20mins stretching at end of sessions.	1 <sup>st</sup> month: 4	3

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			-Supervised and autonomous	Wk 5-12: One additional autonomous session added, alternating between endurance and resistance each week.	2 <sup>nd</sup> + 3 <sup>rd</sup> months: 5	
Wens et al. (2015) [19]	Randomised control trial	Rest, Submaximal intensity exercise	Cardiovascular and resistance training -Supervised	Control group: no intervention Intervention group: Sessions began with cardiovascular training (walk, run or cycle) followed by upper and lower body resistance training. Cardiovascular training ranged from 1x6min > 3x10min bouts, and resistance training ranged from 1x10 > 4x15 repetitions. Volume and intensity of CV and resistance gradually increasing through the programme.	5	6
Kerling et al. (2015) [18]	Non-controlled	Submaximal, maximal intensity exercise	MS <sub>CMBD</sub> : Endurance and resistance training	MS <sub>CMBD</sub> +MS <sub>END</sub> : 1 <sup>st</sup> phase began with 20 minutes of endurance training on cycle ergometer at 50% workload <sub>MAX</sub> , Borg≤13.	4	3



	randomized trial		<p>MS<sub>END</sub>: Endurance training only</p> <p>-Supervised</p>	<p>MS<sub>END</sub>: 2<sup>nd</sup> phase of endurance training continued for further 20minutes on X-trainer, stepper, arm ergometer, treadmill or rowing ergometer, as chosen by participant. HR kept within parameters determined by the exercise method chosen.</p> <p>MS<sub>CMBD</sub>: 2<sup>nd</sup> phase was a full-body dynamic resistance training programme. Participants aimed for 2x10-15 repetitions for each exercise, intensity was increased once 2x15repetitions were achieved.</p>		
Hansen et al. (2015a) [25]	Randomised control trial	Rest, submaximal intensity exercise	<p>Endurance and resistance training</p> <p>-Supervised</p>	<p>Control group: no intervention</p> <p>Intervention group: Sessions began with cycling, walking or running endurance training, gradually increasing from 1x6 &gt; 3x10 minutes and corresponding to 12-14 Borg scale rating.</p>	5	6

				<p>This was followed by upper and lower body resistance training, gradually increasing from 1x10 &gt; 4x15 repetitions as self-determined by participant with encouragement from supervisor.</p> <p>Intensity corresponded to 12-14 Borg scale rating.</p>		
<p>Hansen et al. (2015b) [26]</p> <p>2-part study</p>	<p>Pt 1: Cross-sectional study (MS vs. HC)</p> <p>Pt2: Randomised control trial (MS<sub>INT</sub> vs. MS<sub>CTL</sub>)</p>	<p>Submaximal intensity exercise</p>	<p>Endurance and resistance training</p> <p>-Supervised</p>	<p>For Pt2 of study:</p> <p>Control group: no intervention</p> <p>Intervention group: Sessions began with cycling or walking endurance training, gradually increasing from 1x6 &gt; 3x10 minutes and corresponding to 12-14 Borg scale rating.</p> <p>This was followed by upper and lower body resistance training, gradually increasing from 1x10 &gt; 4x15 repetitions as self-determined by participant with encouragement from supervisor.</p>	5	6

				Intensity corresponded to 12-14 Borg scale rating.		
Keytsman et al. (2019) [27]	Controlled non-randomised trial	Maximal intensity exercise	Periodized endurance and HIIT -Home-based or group	<p>Training programme progressed through 8 3-week cycles:</p> <p>Wk1 – High intensity endurance training</p> <p>Wk2 – Max intensity interval training</p> <p>Wk3 – Interval training and optional endurance sessions</p> <p>Session structures ranged from 3x60-90 second intervals at 90-100% HR<sub>MAX</sub>, 1-3 hour sessions at 60-80% HR<sub>MAX</sub>, or 40-55km at 60-80% or 70-90% HR<sub>MAX</sub>. All sessions were undertaken on cycle ergometers.</p>	7-8/3weeks	6

405 Abbreviations: High intensity interval training (HIIT), combined exercise (CMBD), endurance exercise (END), heart rate (HR)

406

407

408 Table 3. Blood Lactate conditions and outcomes in exercise intervention studies

Study	Lactate testing state	Test exercise equipment	Exercise test type	Lactate testing apparatus	When was blood lactate measured?	Pre-Intervention Lactate concentration (mmol/L) ± SD	Post-Intervention Lactate concentration (mmol/L) ± SD
Keytsman et al. (2017) [23]	Maximal intensity exercise	Cycle ergometer: eBike BasicVR, General Electric GmbH, Bitz, Germany	CPET	No description given	Not reported, but identical result format as Keytsman et al. (2019): Lactate <sub>MAX</sub> taken at cessation of CPET and Lactate <sub>PEAK</sub> was greatest value recorded during recovery	Lactate <sub>MAX</sub> : MS – 5.8 ± 1.8  Lactate <sub>PEAK</sub> : MS – 8.2 ± 2.8	Lactate <sub>MAX</sub> : MS – 5.7 ± 1.4  Lactate <sub>PEAK</sub> : MS – 10.3 ± 2.2

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Zaenker et al. (2018) [24]	Maximal intensity exercise	Cycle ergometer: Ergoline type Ergoselect 200P	CPET	No description given	End of test	Lactate <sub>MAX</sub> : MS <sub>ALL</sub> – 6.9 ± 2.2 MS <sub>EDSS 0-3</sub> – 7.4 ± 1.6 MS <sub>EDSS 3.5-5</sub> – 5.9 ± 2.8	Lactate <sub>MAX</sub> : MS <sub>ALL</sub> – 9.1 ± 3.1 MS <sub>EDSS 0-3</sub> – 9.3 ± 2.2 MS <sub>EDSS 3.5-5</sub> – 8.5 ± 4.4
Wens et al. (2015) [19]	Rest, Submaximal intensity exercise	Cycle ergometer: eBike Basic; General Electric GmbH, Bitz, Germany	25% W <sub>MAX</sub>	Accutrend Plus	3-minute resting period before exercise and end of exercise bouts 1 (B1) and 2 (B2).	Lactate <sub>REST</sub> : MS <sub>INT</sub> – 2.6 ± 0.54 MS <sub>CTL</sub> – 2.6 ± 0.77 Lactate <sub>SUB-MAX</sub> : MS <sub>INT</sub> – (B1) 3 ± 0.54 (B2) 2.8 ± 1.08 MS <sub>CTL</sub> – (B1) 3.1 ± 0.77 (B2) 3.1 ± 0.77	Lactate <sub>REST</sub> : MS <sub>INT</sub> – 2.2 ± 0.54 MS <sub>CTL</sub> – 2.3 ± 0.77 Lactate <sub>SUB-MAX</sub> : MS <sub>INT</sub> – (B1) 2.6 ± 0.54 (B2) 2.5 ± 0.54 MS <sub>CTL</sub> – (B1) 3.3 ± 1.16 (B2) 3.1 ± 0.77
Kerling et al. (2015) [18]	Submaximal, maximal intensity exercise	Cycle ergometer: Ergometrics 900s, Ergoline, Bitz, Germany	CPET	EBIO 6666	1 minute after beginning of CPET, every 3 minutes during exercise,	Lactate <sub>50W</sub> : MS <sub>CMBD</sub> – 1.45 ± 0.51 MS <sub>END</sub> – 1.57 ± 0.79 Lactate <sub>MAX</sub> :	Lactate <sub>50W</sub> : MS <sub>CMBD</sub> – 1.31 ± 0.51 MS <sub>END</sub> – 1.35 ± 0.52 Lactate <sub>MAX</sub> :

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					and at 50W in CPET	MS <sub>CMBD</sub> – 5.43 ± 2.03 MS <sub>END</sub> – 4.8 ± 2.89	MS <sub>CMBD</sub> – 5.9 ± 1.97 MS <sub>END</sub> – 5.14 ± 2.5
Hansen et al. (2015a) [25]	Rest, submaximal intensity exercise	Cycle ergometer: eBike Basic, General Electric GmbH, Bitz, Germany	25% W <sub>MAX</sub>	Accutrend Plus	3 minute resting period before exercise and end of exercise bout	Lactate <sub>REST</sub> : MS <sub>INT</sub> – 2.7 ± 0.4 MS <sub>CTL</sub> – 2.4 ± 0.7 Lactate <sub>SUB-MAX</sub> : MS <sub>INT</sub> – 3.1 ± 0.9 MS <sub>CTL</sub> – 3.1 ± 0.5	Lactate <sub>REST</sub> : MS <sub>INT</sub> – 2 ± 0.6 MS <sub>CTL</sub> – 2.2 ± 0.6 Lactate <sub>SUB-MAX</sub> : MS <sub>INT</sub> – 2.4 ± 0.7 MS <sub>CTL</sub> – 3.3 ± 1.1
Hansen et al. (2015b) [26]	Submaximal intensity exercise	Cycle ergometer: eBike Basic, General Electric GmbH, Bitz, Germany	MS – 25% W <sub>MAX</sub> HC – 35% W <sub>MAX</sub>	Accutrend Plus	During final minute of exercise	Lactate <sub>SUB-MAX</sub> : Pt 1: MS <sub>ALL</sub> – 3.1 ± 0.8 HC – 3.1 ± 1.1 Pt 2: MS <sub>INT</sub> – 3.2 ± 0.8 MS <sub>CTL</sub> – 3.4 ± 0.6	Lactate <sub>SUB-MAX</sub> : Pt 1: N/A Pt 2: MS <sub>INT</sub> – 2.5 ± 0.7 MS <sub>CTL</sub> – 3.6 ± 1

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Keytsman et al. (2019) [27]	Maximal intensity exercise	Cycle ergometer: eBike Basic, General Electric GmbH, Bitz, Germany	CPET	Accutrend Plus	Lactate <sub>MAX</sub> taken at cessation of CPET and Lactate <sub>PEAK</sub> was greatest value recorded during post-exercise recovery	Lactate <sub>MAX</sub> : MS – 4.5 ± 1.6 HC – 5.3 ± 1.3 Lactate <sub>PEAK</sub> : MS – 8.9 ± 2.3 HC – 9.7 ± 2.4	Lactate <sub>MAX</sub> : MS – 5.9 ± 1.8 HC – 6.9 ± 2.1 Lactate <sub>PEAK</sub> : MS – 10.3 ± 3.1 HC – 10.7 ± 2.2
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409 Abbreviations: not applicable (N/A), not stated (N/S), Multiple Sclerosis group (MS), healthy control group (HC), control group (CTL), intervention group (INT), combined-exercise group  
 410 (CMBD), endurance-only group (END), cardiopulmonary exercise test (CPET), maximal workload ( $W_{MAX}$ ), standard deviation (SD), Lactate value taken at cessation of CPET (Lactate<sub>MAX</sub>), greatest  
 411 lactate value recorded during post-exercise recovery (Lactate<sub>PEAK</sub>).

412

413

414 Table 4. Blood Lactate conditions and outcomes in observational studies

Study	Lactate testing state	Test exercise equipment	Exercise test type	Lactate testing apparatus	When was blood lactate measured?	Lactate concentration (mmol/L) ± SD	
Amorini et al. (2014) [7]	Rest	N/A	N/A	Spectrophotometric determination	Following 15 minutes of complete rest	Lactate <sub>REST</sub> : MS - 3.04 ± 1.26 HC - 1.09 ± 0.25	
Cleland et al. (2016) [28]	Rest	N/A	N/A	Lactate Pro	Seated on cycle ergometer before exercise	Lactate <sub>REST</sub> : Week 1: MS - 2.9 ± 2.6 HC - 2.1 ± 0.8	Lactate <sub>REST</sub> Week 2: MS - 1.9 ± 0.5 HC - 2.2 ± 1.1
Hansen et al. (2013b) [17]	Submaximal intensity exercise	Cycle ergometer: eBike Basic, General Electric GmbH, Bitz, Germany	MS – 25% W <sub>MAX</sub> HC – 35% W <sub>MAX</sub>	Accutrend Plus	End of exercise bouts 1 (B1) and 2 (B2).	Lactate <sub>SUB-MAX</sub> : MS – (B1) 3.1 ± 0.8 (B2) 3.0 ± 0.7 HC – (B1) 3 ± 1.1 (B2) 2.6 ± 1.4	



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Hansen et al. (2013a) [29]	Submaximal intensity exercise	Cycle ergometer: eBike Basic, General Electric GmbH, Bitz, Germany	MS – 25% $W_{MAX}$ HC – 35% $W_{MAX}$	Accutrend Plus	End of exercise bout.	Lactate <sub>SUB-MAX</sub> : MS - 2.9 ± 0.6 HC - 3.1 ± 1.1
Juybari et al. (2018) [30]	Rest	N/A	N/A	Lactate assay kit	N/S	Lactate <sub>REST</sub> : MS - 2.5 ± 0.6 HC - 1.6 ± 0.34
Kerling et al. (2014) [31]	Maximal intensity exercise	Cycle ergometer: Ergoline viasprint 150 P, ergoline, Bitz, Germany	CPET	EBIO 6666	1 minute after beginning of CPET and every subsequent 3 minutes.	Lactate <sub>MAX</sub> : MS <sub>ALL</sub> - 5.09 ± 2.52 HC - 9.17 ± 2.9 MS <sub>EDSS 0-3</sub> - 5.86 ± 2.5 MS <sub>EDSS 3.5-6</sub> - 3.77 ± 2.01

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Langeskov-Christensen et al. (2014) [32]	Maximal intensity exercise	Cycle ergometer: Monark Ergomedic 828E, Monark AB, Sweden	CPET	YSI 1500 Sport	Within 1 minute after cessation of CPET.	Lactate <sub>MAX</sub> : Test 1: MS - 9.5 ± 2.7 Test 2: MS - 9.4 ± 2.6
Larson et al. (2014) [33]	Maximal intensity exercise	Cycle ergometer: Lode; Groningen, the Netherlands	CPET	No description given	3 minutes after CPET [37]	Lactate <sub>MAX</sub> : MS - 5.7 ± 2.5 HC - 6.7 ± 1.8
Penesova et al. (2015) [34]	Rest	N/A	N/A	SUPER GL2	8AM fasted state	Lactate <sub>REST</sub> : MS - 1.16 ± 0.38 HC - 1.00 ± 0.22
Schlüter et al. (2017) [35]	Maximal intensity exercise	Cycle ergometer: Ergofit® 3000	CPET	Biosen C-Line	Multiple blood tests taken during recovery phase following CPET,	Lactate <sub>MAX</sub> : MS <sub>EDSS 0-3.5</sub> - 6.5 ± 2.4 MS <sub>EDSS 4-6</sub> - 4.1 ± 2

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					with the highest value recorded as Lactate <sub>MAX</sub> .		
't Eijnde et al. (2014) [36]	Rest, submaximal intensity exercise	Cycle ergometer: eBike Basic, General Electric GmbH, Germany	MS – 25% W <sub>MAX</sub> HC – 35% W <sub>MAX</sub>	Accutrend Plus	During 3 minutes of rest on bike after 15 minutes of seated rest in laboratory and during exercise bout 1 (B1) and 2 (B2).	Control (Week 1): Lactate <sub>REST</sub> : MS - 2.9 ± 0.8 HC - 2.5 ± 0.8 Lactate <sub>SUB-MAX</sub> : MS – (B1) 2.7 ± 0.9 (B2) 2.7 ± 0.9 HC – (B1) 3 ± 1.1 (B2) 2.3 ± 0.7	Precooling (Week 2): Lactate <sub>REST</sub> : MS – 3.2 ± 0.9 HC – 3.1 ± 1.1 Lactate <sub>SUB-MAX</sub> : MS – (B1) 3.3 ± 0.9 (B2) 2.9 ± 0.8 HC – (B1) 3.2 ± 0.9 (B2) 2.9 ± 1.2

415 Abbreviations: not applicable (N/A), not stated (N/S), Multiple Sclerosis (MS), healthy controls (HC), relapsing remitting (RR), chronic progressive (CP), cardiopulmonary exercise test (CPET),

416 maximal workload (W<sub>MAX</sub>), standard deviation (SD).

417 3.12 Meta Analyses

418

419 15 studies in total were included for 5 meta-analysis comparisons: resting lactate concentrations  
420 between HC and PwMS [7,28,30,34,36]; maximum lactate concentrations between HC and PwMS  
421 [27,31,33]; maximum lactate concentration between higher EDSS scores and lower EDSS scores in  
422 PwMS [24,31,35]; submaximal lactate concentrations pre and post training in PwMS ([19,25,26]; and  
423 maximum lactate concentrations pre and post training in PwMS [18,23,24,27]. The descriptive  
424 statistics of these meta-analyses are located in Table 5. 4 desirable meta-analyses were not possible  
425 to perform due to limited or no data (Lactate<sub>SUB-MAX</sub> in HC vs. MS, Lactate<sub>REST</sub> in High vs. Low EDSS,  
426 Lactate<sub>SUB-MAX</sub> in High vs. Low EDSS, Lactate<sub>REST</sub> in PwMS Pre- vs. Post-intervention).

427

428

429 Table 5: Summary statistics of relationships between the studies included in each meta-analysis,  
 430 illustrated in Figures 2-6.

	HC vs. MS Lactate <sub>REST</sub> Figure 2.	HC vs. MS Lactate <sub>MAX</sub> Figure 3.	High vs. Low EDSS Lactate <sub>MAX</sub> Figure 4.	Pre vs. Post Lactate <sub>SUB-MAX</sub> Figure 5.	Pre vs. Post Lactate <sub>MAX</sub> Figure 6.
Q-value	34.72	7.71	0.617	0.237	8.089
DF	4	2	2	2	4
P-Value	<0.001	0.021	0.735	0.88	0.088
I <sup>2</sup> value	88.48	74.081	0.000	0.000	50.54

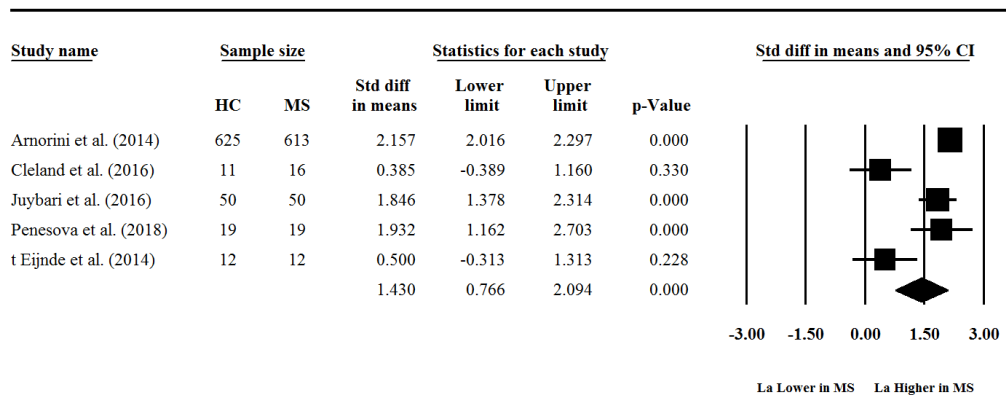
431 Abbreviations: Degrees of Freedom (DF).

432

433 3.12.1 Resting Lactate

434 Compared to HC, blood lactate concentration at rest was significantly higher in PwMS ( $p < 0.001$  with  
435 a standardised difference in means of 1.43 (95% CI [0.788, 2.094]). From the summary statistics (Table  
436 5), the  $I^2$  value of 88.48 and  $p$ -value of  $< 0.001$  illustrate a statistically significant, substantial  
437 heterogeneity between outcomes of the studies included in the meta-analysis. The direction of the  
438 effect was the same across all studies, though the effect was not significant in 2 of the studies included  
439 within the meta-analysis [28,36]. Significant elevation in the pooled resting lactate in PwMS remained  
440 with a standardised difference in means of 1.18 when the results of Amorini et al. (2014) were  
441 removed from the meta-analysis.

442



443

444 **Fig. 2** Forest plot of resting lactate concentrations obtained from PwMS compared with HC, with the

445 final row representing results of data analysis synthesis. Abbreviations: Lactate (La).

446

447

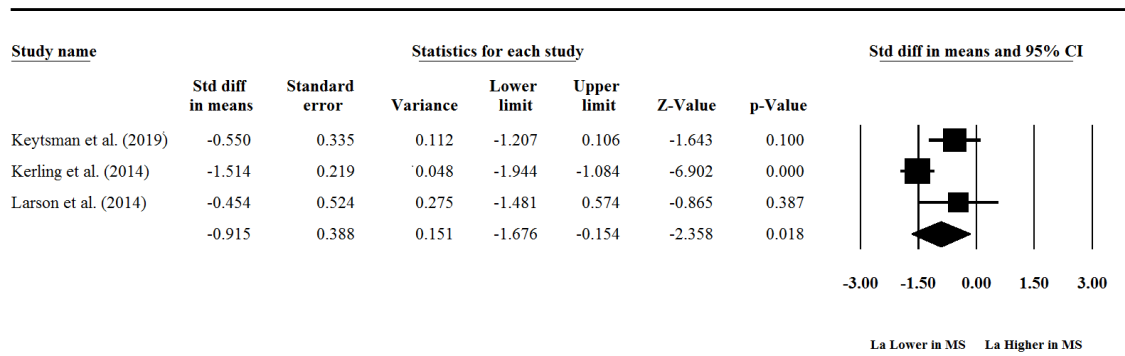
448 3.12.2 Maximal Lactate

449

450 Compared to HC, blood lactate concentration in response to maximal intensity exercise was  
451 significantly lower in PwMS ( $p < 0.001$ ) with a standardised difference in means of 1.141 (95% CI [0.801,  
452 1.480]). From the summary statistics (Table 5), the  $I^2$  value of 74.081 and p-value of 0.021 illustrate a  
453 statistically significant, moderate to substantial heterogeneity between outcomes of the studies  
454 included in the meta-analysis. The same direction of effect was observed across all studies, however,  
455 only the results of Kerling et al (2014) were statistically significant, despite a significant pooled effect  
456 across all studies.

457





458

459 **Fig. 3** Forest plot of maximal lactate concentrations obtained from PwMS compared with HC, with the

460 final row representing results of data analysis synthesis. Abbreviations: Lactate (La)

461

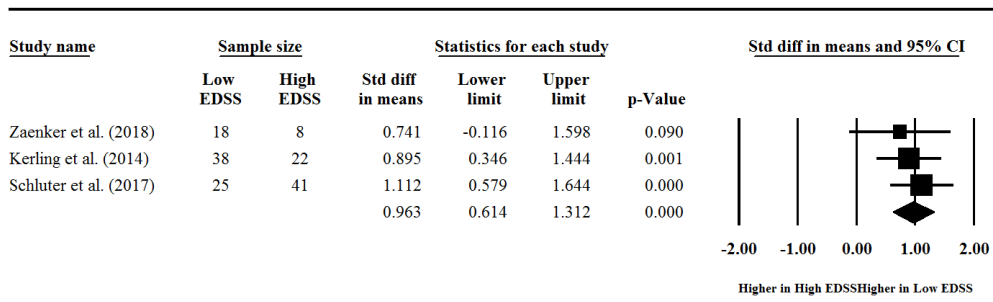
462

463 3.12.3 Maximal Lactate, High EDSS vs. Low EDSS

464

465 Compared to higher EDSS score counterparts, blood lactate concentration in response to maximal  
466 intensity exercise was significantly higher in PwMS with lower EDSS scores ( $p < 0.001$ ) with a  
467 standardised difference in means of 0.963 (95% CI [0.614, 1.312]). From the summary statistics (Table  
468 5), the  $I^2$  value of 0 and p-value of 0.735 illustrate no evidence of significant heterogeneity between  
469 outcomes of the studies included in the meta-analysis with a similar direction and magnitude of effect  
470 within all studies, despite the differences reported within the study by Zaenker et al. (2018) being non-  
471 significant.

472



473

474 **Fig. 4** Forest plot of maximal lactate concentrations obtained in studies which compared groups of  
 475 higher EDSS scoring individuals compared with maximal lactate concentrations obtained in groups of  
 476 lower EDSS scoring individuals, with the final row representing results of data analysis synthesis.

477

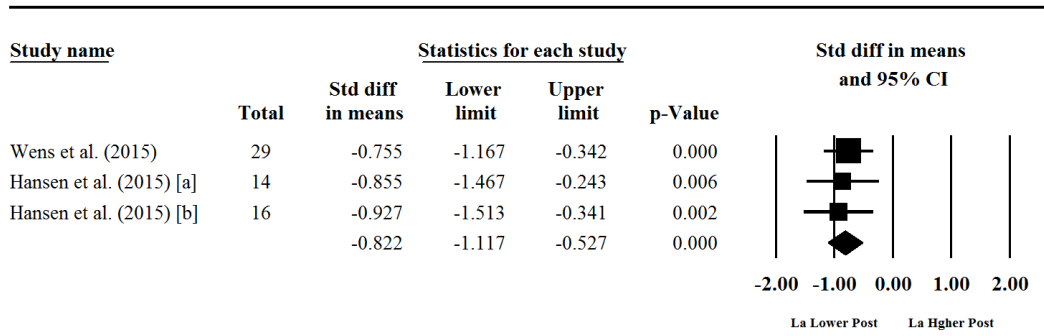
478

479 3.12.4 Sub-maximal Lactate, Pre-/Post-Intervention

480

481 Compared to pre-intervention values, blood lactate concentration in response to sub-maximal  
482 intensity exercise was significantly lower post-intervention in PwMS ( $p < 0.001$ ) with a standardised  
483 difference in means of  $-0.822$  (95% CI  $[-1.117, -0.527]$ ). From the summary statistics (Table 5), the  $I^2$   
484 value of 0 and p-value of 0.88 illustrate no evidence of significant heterogeneity between outcomes  
485 of the studies included in the meta-analysis with a similar direction and magnitude of effect within all  
486 studies.

487



488

489 **Fig. 5** Forest plot of sub-maximal lactate concentrations obtained in PwMS before and after  
 490 completion of exercise intervention, with the final row representing results of data analysis synthesis.

491 Abbreviations: Lactate (La)

492

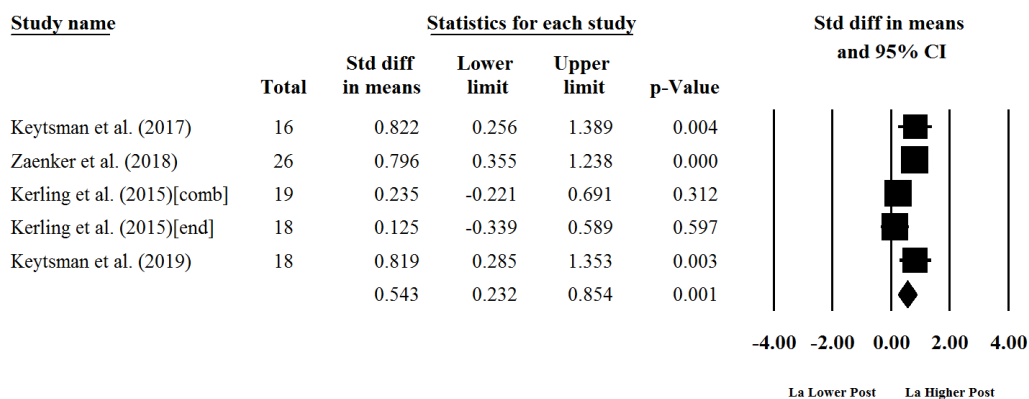
493

494 3.12.5 Maximal Lactate, Pre-/Post-Intervention

495

496 Compared to pre-intervention values, blood lactate concentration in response to maximal intensity  
497 exercise was significantly higher post-intervention in PwMS ( $p < 0.001$ ) with a standardised difference  
498 in means of  $-0.543$  (95% CI  $[0.232, 0.854]$ ). From the summary statistics (Table 5), the  $I^2$  value of 50.54  
499 suggests moderate heterogeneity between outcomes of the studies included in the meta-analysis but  
500 due to a p-value of 0.088 heterogeneity between outcomes is statistically non-significant. The  
501 direction of effect was generally the same across all studies, however, the 2 exercise groups within  
502 the study by Kerling et al. (2015) produced non-significant increases in Lactate<sub>MAX</sub> following exercise  
503 intervention.

504



505

506 **Fig. 6** Forest plot of maximal lactate concentrations obtained in PwMS before and after completion of  
 507 exercise intervention, with the final row representing results of data analysis synthesis. Abbreviations:  
 508 Lactate (La), combined training intervention group ([comb]), endurance training intervention group  
 509 ([end]).

510

511

512 4.0 Discussion

513

514 4.1 Main findings

515

516 The objectives of this systematic review and meta-analysis were to compare blood lactate  
517 concentrations at rest and during exercise in PwMS to HC, and to assess changes in exertional blood  
518 lactate production following an exercise intervention. The main findings are (1) resting blood lactate  
519 is significantly higher in PwMS than in HC, (2) maximal lactate is lower in PwMS than in HC, and (3) in  
520 PwMS exercise training results in reductions in submaximal and increases in maximal lactate  
521 concentrations. Meta-regression analysis to test for associations was precluded, however, as there  
522 were insufficient studies for them to be performed. This is the first systematic review and meta-  
523 analysis on these topics, providing novel findings across all variables and indicating multiple potential  
524 pathways for future research in MS symptom development, disease severity and intervention impact.

525

526 4.2 Lactate in PwMS compared to HC

527

528 4.2.1 Lactate<sub>REST</sub>

529

530 PwMS display significantly higher Lactate<sub>REST</sub> compared to HC with a large effect size (Figure 2). This  
531 supports the conclusions drawn by Amorini et al. [7], who reported higher resting lactate  
532 concentrations in all MS types included within their study compared to HC (RR, PP, SP; see Section  
533 3.3). These findings provide support for blood lactate to potentially find use as a biomarker for MS  
534 and the hypothesis of an association between mitochondrial dysfunction and MS. It is also important  
535 to note that while the effect size was reduced on the removal of one study [7] due to the relative  
536 statistical power of this study's large sample size, the elevation of Lactate<sub>REST</sub> in PwMS remained both  
537 large and statistically significant, further strengthening the integrity of these results.



538

539 The mechanism behind this is unclear and may arise from a variety of causes. From the limited data  
540 provided for  $VO_{2\text{REST}}$  in PwMS compared to HC [17,28,36], there seems to be little evidence to suggest  
541 that the differences in resting lactate are caused by differences in oxygen consumption in PwMS  
542 compared to HC, though evidence is limited and such confounding factors are not completely  
543 eliminated. It is also unlikely that  $Lactate_{\text{REST}}$  is affected by differences in fitness levels between PwMS  
544 and HC given previous research has reported no significant differences in  $Lactate_{\text{REST}}$  between trained  
545 and untrained healthy cohorts [39–41]. Elevated lactate at rest has been recorded as a result of  
546 mitochondrial dysfunction [42] and atherosclerosis [43], as well as others including ischaemia,  
547 malignancy and trauma [44]. It has been hypothesized in previous literature that mitochondrial  
548 dysfunction may be a contributor to progression of MS [45–47], as such, the results of the meta-  
549 analysis (Figure 2) may strengthen the potential link between MS and mitochondrial dysfunction.  
550 Conversely, however, the elevation of resting blood lactate may find a root in impaired peripheral  
551 vascular function. It has been reported that elevated pulse pressure, a marker of arterial stiffness, is  
552 significantly associated with a decreased distance during a 6-minute walk test (6-MWT) in PwMS [48],  
553 a measure which correlates strongly with EDSS score [49]. It has also been reported that vascular  
554 comorbidities are associated with impaired mobility in PwMS [50]. Given the reported association with  
555 lactate and atherosclerosis in previous literature [43] and the association between Multiple Sclerosis  
556 and elevated risk of cardiovascular disease [51,52], elevated lactate in PwMS may be an indicator of  
557 secondary vascular effects, as well as or instead of primary neurological effects of MS. At this stage of  
558 research, however, it is not possible to make firm conclusions.

559

560 More studies are required to measure the association between EDSS score and blood lactate at rest.  
561 Amorini et al. [7] reported a moderate correlation between resting blood lactate and EDSS score ( $R^2 =$   
562  $0.419$ ;  $p < 0.0001$ ) and strong correlation between resting blood lactate and median EDSS score in  
563 PwMS ( $R^2 = 0.972$ ;  $p < 0.0001$ ) but none of the other studies found in this systematic review compared

564 resting blood lactate between groups with higher and lower EDSS scores. A useful direction of future  
565 research would be to focus on intra-individual changes in resting blood lactate concentration for  
566 PwMS throughout disease progression, undergoing relapse and/or fatigue and the specificity and  
567 sensitivity of blood lactate as a biomarker of MS.

568

#### 569 4.2.2 Lactate<sub>MAX</sub>

570 PwMS have significantly lower lactate production at maximal intensity exercise levels compared to HC  
571 (Figure 3). Unfortunately, there were insufficient statistical analyses of power output during the CPETs  
572 used to measure Lactate<sub>MAX</sub> and the authors of this paper were subsequently unable to establish the  
573 contribution of differences in power output to the differences in Lactate<sub>MAX</sub> between PwMS and HC.  
574 One study did report non-significant differences between power output at maximum intensity  
575 exercise in PwMS compared to HC [33], but the small sample sizes within the study (8 PwMS and 7  
576 HC) mean the results cannot be generalised to the wider population with confidence. Previous  
577 literature with larger sample sizes have confirmed significantly reduced peak power output in PwMS  
578 compared to HC [53,54]. Additionally, the evidence from papers reporting VO<sub>2</sub><sub>MAX</sub> in PwMS and HC  
579 [31–33,38], which reported higher VO<sub>2</sub><sub>MAX</sub> values in HC compared to PwMS suggests a reduced  
580 metabolic demand from musculoskeletal tissues in MS compared to HC, a finding supported by a  
581 previous systematic review and meta-analysis on aerobic capacity on PwMS [55]. As such, it may not  
582 be unreasonable to conclude that reduced metabolic demand on the muscles due to reduced muscle  
583 activation is a potentially significant mechanism behind the difference in power output at maximum  
584 intensity exercise and may have an impact on Lactate<sub>MAX</sub> response. This would be supported by  
585 previous literature reporting reduced motor unit activation and recruitment as a symptom of MS [56].  
586 Additionally, a reduced Lactate<sub>MAX</sub> response in higher EDSS scores compared to lower EDSS scores was  
587 observed (Figure 4). As above, the mechanism behind this phenomenon is likely to be affected by  
588 increasing impairment of motor unit recruitment with increasing severity of MS symptoms [56]. This

589 is supported by the data from the literature in this review (Section 3.10.2) as well as in other literature  
590 [53] reporting significantly lower peak power output in higher EDSS scoring groups of PwMS compared  
591 to their lower EDSS scoring counterparts. The presence of this reduced Lactate<sub>MAX</sub> response in PwMS  
592 and its further reduction with increasing EDSS score also suggests potential for Lactate<sub>MAX</sub> to function  
593 as a biomarker itself. What remains unclear is if other hypothesized mechanisms of MS progression,  
594 e.g. mitochondrial dysfunction, are also playing a contributing factor to the impairment of Lactate<sub>MAX</sub>  
595 response in PwMS both compared to HC and with increasing disease severity. Future research is  
596 required to clarify the magnitude of this impairment of Lactate<sub>MAX</sub> due to MS symptoms as well as the  
597 precise mechanisms by which this impairment occurs.

#### 598 4.2.3 Lactate response range

599 Taken together, the findings of the meta-analyses of Lactate<sub>REST</sub> and Lactate<sub>MAX</sub> (Figures 2 and 3)  
600 illustrate an overall lactate range compression from resting state to maximal exertion in PwMS  
601 compared to HC. Moreover, the findings that Lactate<sub>MAX</sub> is further reduced in PwMS with higher EDSS  
602 when compared to those with lower EDSS scoring PwMS (Figure 4) show that Lactate<sub>REST</sub> – Lactate<sub>MAX</sub>  
603 range compression is further exacerbated by MS disease progression. The occurrence of this  
604 compression is largely supportive of the suggestion by Amorini et al. [7] that resting blood lactate in  
605 PwMS is correlated EDSS score, but future research into lactate mechanics in PwMS is still warranted  
606 to confirm the extent of lactate range compression in PwMS and if it could have any clinical relevance  
607 in measuring severity of disease progression.

608

#### 609 4.3 Effects of exercise intervention on Lactate in PwMS

610

##### 611 4.3.1 Lactate<sub>REST</sub>

612

613 Only 2 studies [19,25] reported the effects of training Lactate<sub>REST</sub> in PwMS and so there were not  
614 enough studies to perform a meta-analysis on this variable. However, both studies reported  
615 significant within-group falls in resting blood lactate following a 6-month training intervention,  
616 suggesting that blood lactate in PwMS may be reduced in response to training. Given the association  
617 between EDSS and Lactate<sub>REST</sub> reported by Amorini et al. [7], a key question for future research is  
618 whether changes in Lactate<sub>REST</sub> translate to alterations in EDSS or symptom severity. Without  
619 assessment of resting lactate, it is impossible to determine if any changes in symptom severity occur  
620 in tandem with alterations in metabolic processes.

621

#### 622 4.3.2 Lactate<sub>SUB-MAX</sub>

623

624 A meta-analysis of lactate response to submaximal intensity exercise (Figure 5) showed a reduction in  
625 lactate response in PwMS following an exercise intervention. It is worth noting that for at least 2 of  
626 the 3 studies investigating this lactate response [25,26], power output during the sub-maximal  
627 intensity exercise test was kept the same during baseline and post-intervention. Should this be the  
628 case, it would demonstrate that the causes the change in Lactate<sub>SUB-MAX</sub> response following an exercise  
629 intervention were not down to changes in motor unit and muscular activation but instead down to  
630 other potential factors associated with exercise training, such as increasing mitochondrial density, size  
631 and metabolic enzyme activity, increased capillarisation and flow-mediated dilation, as well as others  
632 [57–59]. However, given a lack of change in VO<sub>2</sub><sub>SUB-MAX</sub> for the cohorts within the studies which  
633 reported it [25,26] it is difficult to confirm if this is the case or if such changes to Lactate<sub>SUB-MAX</sub> in  
634 response to an exercise intervention were clinically meaningful within this cohort. Future research  
635 would be well placed to investigate the precise mechanisms of physiological adaptations to exercise  
636 in PwMS as well as the degree to which sub-maximal lactate response adaptations are associated with  
637 changes in functional capacity and quality of life.

638

639 4.3.3 Lactate<sub>MAX</sub>

640

641 PwMS demonstrate a significant increase in Lactate<sub>MAX</sub> response following a structured exercise  
642 programme over the course of at least 3 months (Figure 6), demonstrating that aerobic capacity in  
643 PwMS can be improved through a structured training programme [55]. Given the variable nature of  
644 each study's training programme, with 5/6 studies incorporating some combination of endurance,  
645 resistance and HIIT training, it is difficult to comment on the precise mechanisms by which adaptations  
646 were made to increase Lactate<sub>MAX</sub> and where training modalities overlap. Nevertheless, it seems likely  
647 that exercise intensity is an important variable in predicting the response. For example, of studies  
648 using HIIT [23,24,27], the greatest changes in Lactate<sub>MAX</sub> (Figure 6) were observed, suggesting a  
649 potential pattern of HIIT adoption and expansion of the compressed Lactate range observed in PwMS.  
650 It has been shown in the general population that HIIT induces greater increases in mitochondrial  
651 content, enzyme activity, potentially skeletal muscle recruitment compared to moderate intensity  
652 training [60,61], and that increased lactate buffering capacity in muscle is observed in both sprint  
653 interval training and continuous training [62]. Additionally, considering 3 studies out of 4 [23,24,27]  
654 reported significant increases in peak power output in PwMS and 2 studies out of 3 [24,31,38]  
655 reported significant increases in VO<sub>2</sub><sub>MAX</sub> in PwMS following an exercise intervention, it can be  
656 reasonably concluded that increased skeletal muscle recruitment and enhanced metabolic activity had  
657 some effect on the increase in Lactate<sub>MAX</sub> response. Further research is needed to determine if HIIT is  
658 indeed more effective at increasing Lactate<sub>MAX</sub> concentration in PwMS and whether the causes by  
659 which this occurs is due to any of the other aforementioned mechanisms. It also remains unclear how  
660 the adaptability of Lactate<sub>MAX</sub> response is affected in PwMS compared to HC, as no studies were found  
661 to investigate these variables. As such, further research into these gaps in the literature are warranted  
662 to expand this knowledgebase.

663

664 It may also be noted that, while the data is too limited to draw confident conclusions, increases in  
665 Lactate<sub>MAX</sub> appear to be associated with increases in both power output and VO<sub>2</sub><sub>MAX</sub> in PwMS. As such,  
666 as has been the case for the researchers of this paper investigating health and fitness-related variables  
667 in PwMS during the COVID-19 pandemic, Lactate testing may be an alternative method of remotely  
668 measuring changes in physical fitness in PwMS in response to an exercise intervention during periods  
669 of limited availability of physical contact with researchers or clinicians. This is due to its widespread  
670 availability to researchers, relatively low equipment costs and potential suitability for PwMS to be  
671 tested at home by either themselves or a carer.

672

#### 673 4.4 Limitations of the literature

674

675 Studies generally had a low sample size, with a third of studies involving less than 30 participants [23–  
676 25,28,33,36], which inherently reduces generalizability of results to the wider population of interest  
677 and risks detecting large sample sizes which cannot be replicated in future studies. A greater number  
678 of studies would allow for a more nuanced analysis, including meta-regression or moderator analysis  
679 to identify areas underpinning any heterogeneity and to minimise influence of results that may have  
680 been duplicated to some degree by repeated use of cohort data across different studies. More studies  
681 would also allow for meta-analyses on the effect of exercise on Lactate<sub>REST</sub> in PwMS or the differences  
682 in Lactate<sub>SUB-MAX</sub> between PwMS and HC exercising at the same power output; both of which were  
683 impossible due to the limited number of studies investigating them.

684

685 It would be beneficial if more studies provided more complete data with regard to power output and  
686 VO<sub>2</sub> during their respective testing conditions. There was limited scope for this review to investigate  
687 potential correlations or confounding factors regarding the interactions between Lactate  
688 concentration, power output and VO<sub>2</sub>; both of which could affect the suitability of blood Lactate  
689 concentration as either a biomarker or as a suitable alternative to more complex physiological

690 measurements of health and fitness in PwMS during times of limited clinician or researcher contact  
691 with PwMS.

692

693 Differences in testing and training protocols, as seen in Tables 2, 3 and 4, made comparisons between  
694 studies difficult. The result of these issues was moderately or highly heterogeneity between studies,  
695 especially for studies comparing Lactate<sub>REST</sub> and Lactate<sub>MAX</sub> between PwMS and HC. An example of this  
696 was in testing site utilised for lactate measurement and the equipment used for lactate testing, where  
697 5 studies failed to adequately describe one or both of these aspects of methodology [23,24,30,31,33].  
698 The most commonly utilised site, the fingertip, was only used by half of the studies included in this  
699 review [17,19,25,26,28,29,32,33,36]. It has been shown that the testing site for blood lactate  
700 measurement makes a significant difference to the concentration measured [63–65] and so the  
701 variability of testing site protocol in studies included in this review may have impacted the reliability  
702 of comparisons between studies. Additionally, 7 studies reported utilising laboratory-based lactate  
703 measurement protocols [7,18,30–32,34,35] and 9 reported using Accutrend Plus and Lactate Pro  
704 portable devices [17,19,25–29,33,36]. While portable lactate testing consumer devices, eg. Accutrend  
705 and Lactate Pro, have been shown to have reasonable accuracy compared with a laboratory-based  
706 method [66], variability in testing equipment may have impacted results across studies. Finally,  
707 findings from Keytsman et al. [23,27] showed a significantly greater blood lactate concentration in  
708 PwMS during post-CPET recovery (Lactate<sub>PEAK</sub>) compared to the moment of CPET cessation  
709 (Lactate<sub>MAX</sub>). Given there were 5 different methods overall for timing Lactate<sub>MAX</sub> measurements  
710 (Section 3.9.3), the differences in post-CPET lactate test timing across studies was a potential cause of  
711 variability in results and would benefit from more uniform measurement procedure in future.

712

713 In terms of reporting EDSS, only one paper [24] provided EDSS data in a form as to be able to establish  
714 interquartile (IQ) range of the EDSS scores of participants, with the rest reporting in the form of mean  
715  $\pm$  SD, or mean  $\pm$  SE. EDSS is not a normally distributed score, therefore such reporting measures

716 provide an incomplete report of EDSS score of the participant group with MS for further research and  
717 it is the recommendation of the authors of this paper to report IQ range of EDSS to provide a clearer  
718 picture of cohorts used within the study.

719

#### 720 4.5 Suggestions for future research

721

722 Given the above, and given the findings in this paper and by Amorini et al. [7], more studies are  
723 required that have blood lactate concentration, changes in blood lactate concentration (if undergoing  
724 a training intervention) and EDSS score/symptom severity as primary outcomes rather than secondary  
725 data reported only as part of a standard exercise /CPET test. With this, longer training interventions,  
726 greater participant numbers and uniform testing procedures, a more nuanced understanding of blood  
727 lactate in PwMS and the mechanisms behind its metabolic response and biomarker reliability would  
728 be available for research.

729

#### 730 4.6 Limitation of this review

731

732 This review was limited primarily by the low number of studies performing blood lactate testing in  
733 PwMS during each condition of rest, and submaximal and maximal exercise intensities. As such, meta-  
734 analyses either could not be performed for a particular condition (eg. Lactate<sub>REST</sub> pre- vs. post-  
735 intervention) or were performed using 3-5 studies. Limiting the publication language to English and  
736 exclusion of non-peer-reviewed literature (e.g. grey literature) means that some studies may have  
737 been missed, however the authors employed a rigorous search strategy to ensure as many studies as  
738 possible were included in the overall review.

739

#### 740 5.0 Conclusion

741



742 Blood lactate occurs in higher concentrations during rest in PwMS compared to HC and Lactate<sub>MAX</sub> is  
743 lower in PwMS compared to HC, with higher EDSS scoring groups displaying lower Lactate<sub>MAX</sub>  
744 concentrations than lower EDSS scoring groups. With a long-term training intervention, Lactate<sub>REST</sub> and  
745 Lactate<sub>SUB-MAX</sub> concentrations may decrease, while Lactate<sub>MAX</sub> concentrations may increase, compared  
746 to baseline values. There is evidence to suggest these changes arise from both increased skeletal  
747 muscle recruitment as well as adaptations to factors such as capillary structure and mitochondrial  
748 number and efficiency. Given the purported role of lactate as both a biomarker of disease severity,  
749 and a prognostic indicator of future relapse, more work is needed to elucidate the association  
750 between resting and exertional metabolism and ongoing disease processes. The primary limitations  
751 to the literature were small sample sizes and heterogeneity of lactate testing protocols due to lactate  
752 testing often being a secondary outcome measure, while the primary limitation to this systematic  
753 review and meta-analysis was a low number of studies included. Future research would be worthwhile  
754 to investigate the changes in blood lactate during fatigue and relapse.

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756

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