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Inflammation and physical activity in multiple sclerosis patients. A systematic review and meta-analysis

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ABSTRACT

Objectives: Due to the inflammatory nature of multiple sclerosis (MS), the most widely used therapeutic approach targets the immune response but can comprise side effects (e.g. secondary immunosuppression). For these reasons, among non-pharmaceutical interventions without known side effects, physical activity (PA) gained importance because it is feasible, safe and a supportive complementary treatment strategy to alleviate symptoms in MS subjects. Consequently, the main aim of this systematic review is to analyze the effect of PA protocols, as a complementary therapy, on inflammatory status in MS patients.

Methods: Four electronic databases (PubMed, Embase, CINAHL, and Cochrane CENTRAL) were systematically searched up to 01 June 2023 (Prospero Protocol ID=CRD42021244418). The refined search strategy was based on three concepts: "MULTIPLE SCLEROSIS" AND "PHYSICAL ACTIVITY" AND "INFLAMMATION".

Results: three main findings emerged: 1) untrained subjects showed a negative modulation of inflammatory biomarkers concentrations when compared to trained people (−0.74, 95 %C.I.–1.16, −0.32); 2) training modulated positively inflammatory biomarkers (+0.47, 95 %C.I. 0.24,0.71); 3) Aerobic PA protocol enhance higher positive influence on inflammation.

Conclusions: Persistent, low-grade inflammation in MS could be upregulated by non-pharmacological complementary therapies, in particular by regular aerobic PA that could reduce and positively modulate inflammation.

1. Introduction

Multiple Sclerosis (MS), a multifactorial immune-mediated disease of the central nervous system (CNS), has increased worldwide by 30 % between 2013 and 2020, with 2.8 million patients estimated.[1] MS is characterized by demyelination, neurodegeneration and chronic inflammation,[2,3] and MS patients (pwMS) can develop different neurological symptoms: muscle spasms, walking difficulties, visual problems, fatigue, pain, depression and poor quality of life.[4–6] The etiology of MS is complex but, inflammation is the major driver of the pathology,[2,7] principally characterized by T cell-mediated reactions[7] against CNS antigens that upregulate pro-inflammatory mediators and activate microglia/macrophages.[5] Increasing evidence suggests that also B lymphocytes and oxidative imbalance may contribute to the pathogenesis and neurodegeneration of MS.[2,8–12]

The most widely used approach to study MS targets the immune responses, analyzing inflammatory biomarkers levels in plasma from pwMS.[13,14] Among the wide options, pro- or anti-inflammatory cytokines are the most reliable and trustworthy choices.[15,16] The mainly studied pro-inflammatory cytokines in MS with are interleukin (IL-6), tumor necrosis factor α (TNF-α), and interferon γ (IFN-γ), while IL-10 plays a role in regulating the pro-inflammatory cascades.[16]

Disease-modifying therapies downregulate immune activation to halt or to partly reverse disease progression and relapses,[17] even with some side effects (secondary immunosuppression or infections).[18] Recently, the focus has shifted on other non-pharmacological interventions supporting traditional therapies, without known side effects. Exercise[19] is a feasible and safe complementary treatment strategy to alleviate symptoms in MS subjects.[20,21] For decades, exercise, hereinafter called Physical Activity (PA), was not recommended because it would increase the risk of exacerbations, symptoms or fatigue.[21] Therefore, PA gained extensive interest in MS management,[22] mainly because it is well known the positive role of an active lifestyle on health[23–26] and, moreover, regular exercise can stabilize or reduce inflammation.[27,28] Evidence-based guidelines have been developed to promote PA in pwMS, also because recent researches showed that these

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subjects are still less active than the healthy population.[24,29] Despite this increasing interest, the association between exercise and inflammatory status in MS are still poorly investigated. Consequently, the main aim of this systematic review is to analyze the effect of applying PA protocols on inflammatory status in pwMS, to provide an overview of the scientific results achieved until today. We included studies that jointly analyzed standardized PA protocols and measured inflammatory biomarkers (IL-6, TNF-α, IFN-γ, and IL-10) in pwMS. A further, not less important, objective is to highlight what kind of PA protocol could be more useful in MS management, to define and integrate preventive strategies and evidence-based guidelines.

2. Materials and methods

The present systematic review is conducted according to the PRISMA 2020 Statement,[30] and its protocol is registered on PROSPERO (Protocol ID=CRD42021244418).

The search strategy evaluates only published studies available in PubMed, Embase, CINAHL, and Cochrane CENTRAL, systematically queried up to 01 June 2023 (first search string: 15 June 2021/second update: 01 June 2023). The search strategy was based on three concepts: "Multiple Sclerosis" AND "physical activity" AND "oxidative stress" OR "inflammation". Further details are reported in supplementary materials (Table A1).

2.1. Inclusion/Exclusion Criteria

Original studies, in English or Italian, on humans suffering from MS (18 + years, no smoking, no other pathologies) and involved in PA protocols, were included. Studies with PA standardized protocols with declared durations, methods, timing, and intensity were included, while PA interventions in extreme conditions (e.g.hypoxia) were excluded. Subjectively (e.g.questionnaire) and objectively measured PA was considered while non-quantitative or explicit data, unpublished research, congress abstracts, reviews, animal/in-vitro studies were excluded. Articles reporting data on drugs or antioxidant supplementation were considered only in presence of control group/time without the supplementation effect, otherwise excluded.

2.2. Data extraction and quality assessment

The screening of included articles was performed by two independent reviewers in a two-step process. The first selection step consists in title/abstracts examination, according to the above-mentioned eligibility criteria. The second phase involves the selection of texts after their integral reading by the reviewers through a double-blind process. When

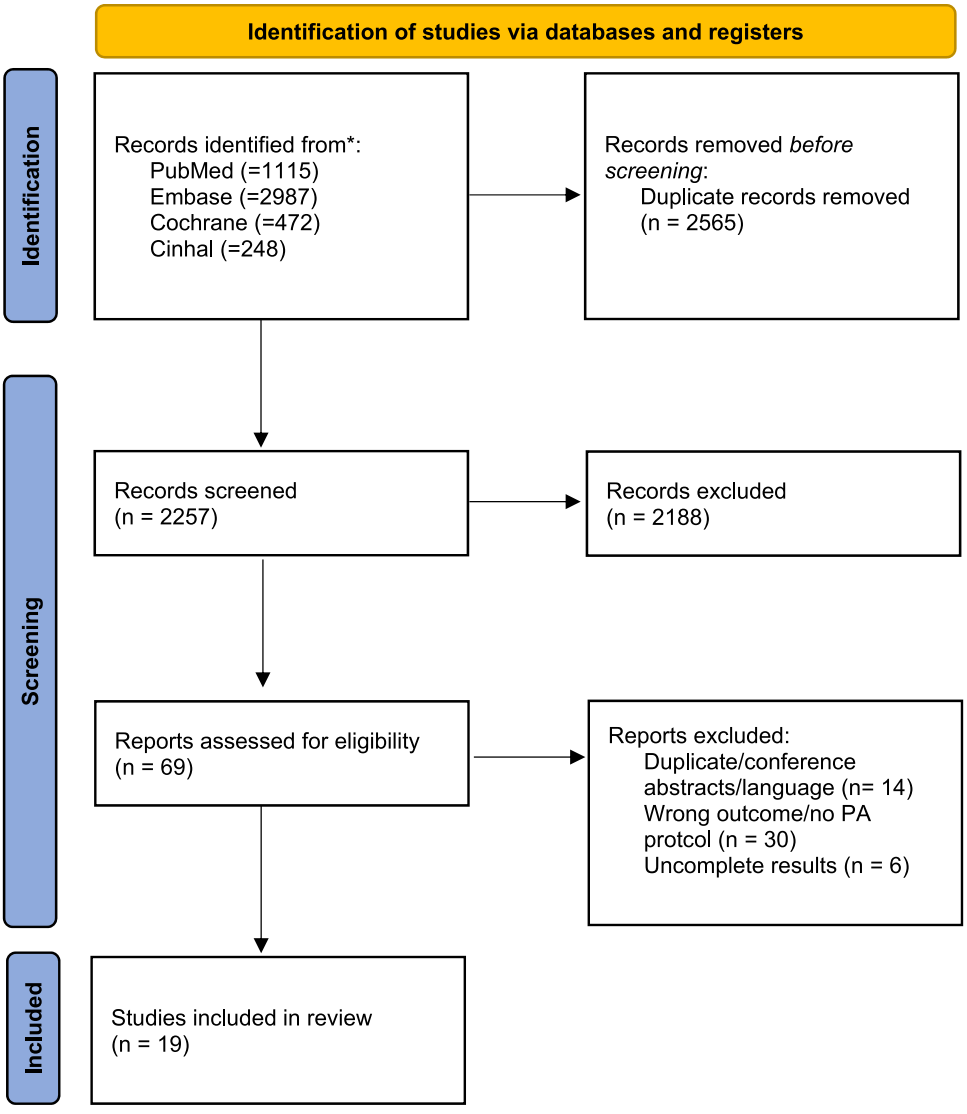

Fig. 1. Systematic Reviews (PRISMA) flow diagram of the studies selection process.

divergences arose, a third reviewer was involved.

The process is summarized in the PRISMA diagram[30] (Figure 1/supplementary Table A1).

Data were collected using a standardized data extraction form, if originally presented by graphs they were extracted by the Webplotdigitizer software. Extracted data included the first author, year of publication, geographical location of the study (supplementary file Fig. A1), study settings, type of MS, sample size, PA protocol type and characteristics, biomarkers analyses (type, analytical methods, matrix, concentration). Study quality were appraised using validated and specifically selected instruments, depending on the study design[31]: i) NIH Quality Assessment Tool for observational, cross-sectional, case--control and before-after studies, ii) PEDro scale to assess the quality of randomized clinical trials, and iii) The Joanna Briggs Institute Critical Appraisal Checklist for quasi-experimental studies (non-randomized).

2.3. Data and statistical analysis

The primary outcomes were: 1) the quantification of inflammatory biomarkers in pwMS attending PA protocols vs pwMS untrained/not-physically active; 2) the comparison between pre and post sessions in pwMS attending PA protocols; 3) the differences in the inflammatory modulation according to PA protocols.

Categorical variables were reported as frequency (n), while continuous variables as Mean ± Standard Deviation (SD), as reported in the original research articles. The statistical analyses and the forest plots were created by Jamovi(1.6.21) and R Studio(RStudio Team 2020).

The analyses were carried out with a random-effect model using the standardized mean difference as the outcome measure. The heterogeneity (i.e.tau^2), was estimated by the restricted maximum-likelihood estimator.[32] In addition, the Q-test for heterogeneity and the I^2 statistic are reported. Studentized residuals and Cook's distances were used to examine outliers and/or influential studies in the model. Studies with a studentized residual larger than a standard normal distribution were considered potential outliers (i.e.Bonferroni correction) while studies with a Cook's distance larger than the median plus six times the IQR were considered influential.

Besides, although the 19 studies belong to different categories of study (randomized studies, case-control studies, pilot studies, etc.), and after different preliminary analyses that showed almost similar results within the different categories, we chose to analyze all the included papers together to strengthen the results, given the small number of studies included in the review.

3. Results

Among the 4822 items initially found, 2565 duplicates were removed, by EndNote and manually, before the first screening. The remaining 2257 were primarily screened, following the inclusion/exclusion criteria declared, and 2188 articles were excluded. Sixty-nine papers were assessed for the full-text screening and, among these, 50 papers were excluded because a) duplicate/conference abstracts or not in English/Italian (n = 14), b) uncompleted or lacking data (n = 6), c) no PA protocol applied (n = 30). In conclusion, 19 research papers were included in the systematic review. All the included studies were assessed by adopting the proper checklist (Supplementary file, table A4) according to the study design and, concerning the quality appraisal, 17 papers out of 19 (89%) were of good quality (High), with a score≥ 7.

3.1. Study and participant characteristics

Table 1 reports the main characteristics of the studies included (SECTION-A), focusing on the population analyzed and the PA protocol adopted by each study, and on the peculiarities of the biomarkers of inflammation (SECTION-B), according to the studies included in the review.

Among the nineteen studies included, n = 13 were randomized trial,[33–45] n = 3 case-control studies[46–48] and n = 3 other typologies (n = 1 before/after study,[49] n = 1 non-randomized single arm,[50] and n = 1 pilot study[51]). Regarding PA protocols, 14 papers used aerobic PA[34–37,39,42–44,46,47,49–51] and 5 used combined PA protocols (aerobic/anaerobic or holistic activities).[33,38,40,41,45] Overall, on average, PA protocols duration lasted 8 weeks, with 2 training sessions per week around 40 min.

About 60% of the studies (Table 1) enrolled pwMS with Relapsing-Remitting (RR) MS[33,37,38,40–44,46,48] and only four studies recruited more severe forms of MS (Primary-Progressive MS (PP)[36,38,44,50] and Secondary-Progressive (SP) MS).[36,38,44,50]

Ten studies[33,34,38,40–45,48] foresaw trained vs untrained subjects, 7 out of the 19 studies enrolled only females,[34,40,41,43,45,46,51] and only 6 studies included healthy control subjects.[33,36,37,46,47,49]

3.2. Biomarkers of Inflammation

In Supplementary file, table A2-A4 summarized the main conclusions and all the inflammatory biomarkers investigated by each study included in this review. To better support our purposes, we chose the most representative, strong and homogeneous biomarkers described in the 19 studies:

- IL-6: covered by 73.7% of the studies;
- TNF-α: covered by 63.1% of the studies;
- IL-10: covered by 57.9% of the studies;
- IFN-γ: covered by 36,8% of the studies.

IL-6 is a soluble mediator with a pleiotropic effect on inflammation, immune response, and hematopoiesis.[52] IL-6 is synthesized in a local lesion in the initial stage of inflammation, followed by a rapid induction of an extensive range of acute phase proteins such as C-reactive protein (CRP), fibrinogen, and haptoglobin.[52] The dysregulated, continual synthesis of IL-6 is a typical bio-molecular mechanism of various immune diseases, included MS.

TNF-α is a homotrimer protein consisting of 157 amino acids, mainly generated by activated macrophages, T-lymphocytes, and natural killer cells.[53] It triggers various inflammatory molecules, including pro-inflammatory cytokines and chemokines. TNF-α has been identified as a major regulator of inflammatory responses and involved in the pathogenesis of some inflammatory and autoimmune diseases, such as MS or rheumatoid arthritis.[54]

IL-10 is a cytokine that exhibits multiple immune modulatory effects in many autoimmune diseases,[55] enhancing the stimulation and survival of autoreactive B cells[56] and influencing the progression of the disease.

IFN-γ is a cytokine produced primarily by activated CD4 + or CD8 + T cells and natural killer cells and is recognized as chief mediator of innate and adaptive immunity.[57] IFN-γ triggers macrophages and upregulates of a variety of pro-inflammatory parameters (e.g. IL-12, TNFα, IL-15).[58]

To date, Table 1-Section B reported the mean concentrations of the four chosen inflammatory parameters. According to Table 1-sectionB.1, five studies (35.7%)[33,34,38,44,45] analyzed IL-6 concentrations both in pre and post training moments and comparing trained vs untrained subjects, six studies (42.8%)[33,36,37,46,47,49] matched pwMS with healthy control and five studies (35.7 %) [36,38,39,50,51] reported no significant results. Table 1-sectionB.2 described six studies (50 %) analyzing TNF-α concentrations both in pre and post training sections and in trained vs untrained subjects, four studies (33.3 %)[33,34,38,42,43,48] matched pwMS with healthy control and four studies (33.3 %)[33,37,46,47] reporting no significant results.[35,38,46,51] Table 1-sectioB.3 described seven studies (63.5 %) analyzing IL-10 concentrations both in pre and post training sections and in trained vs untrained subjects,[33,38,40,42,43,45,48] four studies (36.4 %)[33,46,47,49] matched pwMS with healthy control and three studies (27.3 %)[33,38,40] reporting no significant results. Finally,

Table 1
SECTION A: Formal characteristic of the studies and sample population characteristics of the studies included in the review. **SECTION B:** Biomarkers of inflammation (section B.1 IL-6 / section B.2 TNF-α / section B.3 IL-10 / section B.4 IFN-γ) according to the main characteristics of the studies included in the review.

SECTION A

Study	Country	Study design			PA protocol	PA protocol characteristics			Type of MS	EDSS (mean ±SD)	Sample size	Groups		Age (mean ±SD)	Sex	
		Type	Randomizedgroup	Healthy control group		Total duration (weeks)	Session duration (minutes)	Session frequency (days/weeks)							Male	Female
Alvarenga-Filho H. (2016)	Brazil	controlled trial	No	Yes	Combined	12	60	2	RR	1 ± 1.5	18	8	Trained	41.1 ± 12.9	1	7
												10	Untrained	35.2 ± 7.6	2	8
Bahmani E. (2022)	Iran	randomized controlled study	Yes	No	Aerobic	8	20-40	3	(NA)	(NA)	40	20	Trained	27.2 ± 2.5	-	20
												20	Untrained	26.8 ± 2.9	-	20
Bansi J.(2012)	Switzerland	randomized controlled trial	Yes	No	Endurance/ Aerobic	3	30	1.5	(NA)	4.7 ± 0.6	52	28	Trained 1	52 ± 5.3	10	18
												24	Trained 2	50 ± 5.4	8	17
Barry A.(2019)	Ireland	before and after	No	Yes	Endurance/ Aerobic	8	30	2	(NA)	(NA)	9	All Trained		35.3 ± (NA)	(NA)	
Berkowitz S. (2019)	Israel	case-control study	No	Yes	Aerobic	2	15	1	RR	1.5 ± 1.3	14	All Trained		33.8 ± 7.8	-	14
Briken S.(2016)	Germany	randomized controlled trial	Yes	Yes	Endurance/ Aerobic	9	15-45	2-3	PP/ SP	4.9 ± 0.9	32	All Trained		49.9 ± 7.6	13	19
Castellano V. (2008)	USA	controlled trial	No	Yes	Aerobic	8	30	3	RR	3.4 ± (NA)	11	All Trained		40 ± 10	3	8
Deckx N.(2016)	Belgium	randomized controlled trial	Yes	No	Combined	12	(NA)	5	RR/ SP	3 ± 0.4	45	29	Trained	47 ± 2	13	16
												16	Untrained	50 ± 3	6	10
Devasahayam A.J. (2020)	Canada	non-randomized single arm	No	No	Aerobic	10	50	3	PP/ SP	6 ± 1	10	All Trained		53.2 ± 15.6	1	9
Donia Scott A. (2019)	Germany	controlled trial	No	No	Aerobic	8	30	(NA)	(NA)	4.5 ± 1.5	13	All Trained		57.2 ± 7.6	3	10
Eftekhari E.(2018)	Iran	randomized controlled trial	Yes	No	Pilates/ Combined	8	45	3	RR	(NA)	25	13	Trained	34.5 ± 7.3	-	13
												12	Untrained	31.4 ± 8.9	-	12
Golzari Z.(2010)	Iran	randomized controlled trial	Yes	No	Combined	24	60	3	RR	2.1 ± 1.1	20	10	Trained	32.1 ± 7.6	-	10
												10	Untrained	33.7 ± 8.2	-	10
Kjølhede T.(2016)	Denmark	randomized controlled trial	Yes	No	Resistance training/ Aerobic	8	30	2	RR	2.9 ± 1	32	16	Trained	44.6 ± 7	4	12
												16	Untrained	42.2 ± 8	4	12
Majdinasab N. (2018)	Iran	case-control study	No	Yes	Interval training/ Aerobic	8	40	(NA)	RR	2.4 ± 0.8	35	All Trained		28.2 ± 3.6	(NA)	
Mokhtarzade M. (2018)	Iran	case-control study	No	No	Interval training/ Aerobic	8	40	(NA)	RR	2.4 ± 0.8	63	35	Trained	31.6 ± 2.6	12	23
												28	Untrained	30.6 ± 3.2	9	18
Mokhtarzade M. (2017)	Iran	randomized controlled trial	Yes	No	Interval training/ Aerobic	8	42-66	3	RR	1.8 ± 0.3	40	22	Trained	32 ± 2.8	-	22
												18	Untrained	31.3 ± 3.3	-	18
Schulz K.(2004)	Germany	randomized waitlist trial	Yes	No	Aerobic	8	30	2	RR/ PP/ SP	2.5 ± 0.8	28	15	Trained	39 ± 9	4	11
												13	Untrained	40 ± 11	5	8

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Table 1 (*continued*)

SECTION A																	
White L.(2006)	USA	pilot study	No		No	Resistance training/ Aerobic	3	30	2	RR	3.8 ± 0.9	10	All Trained	47 ± 12	-	10	
Zadeh F.T.(2021)	Iran	randomized trial	Yes		No	Combined	8	60	2	(NA)	5 ± 1	30	15 Trained	32.1 ± 2.1	-	15	
													15 Untrained	32 ± 3.1	-	15	

SECTION B										
Section B.1 Study	Biological sample	Analytical method	IL-6 (pg/mL) **Pre-training (mean±SD)**		**Post-training (mean±SD)**		**Healthy control group (mean±SD)**	Statistical analyses		Notes
Alvarenga-Filho H. (2016)	Blood cells supernatants	ELISA	Trained	12.6 ± 5.2	Trained	11.8 ± 5	7.9 ± 1.8	Control vs trained p = 0.03Control vs untrained p = 0.04		Data extrapolated
			Untrained	15 ± 4.6	Untrained	-				
Bahmani E. (2022)	Blood (serum)	ELISA	Trained	3.9 ± 0.03	Trained	2.4 ± 0.04	No	Pre vs post training p < 0.001		
			Untrained	4.04 ± 0.06	Untrained	-				
Bansi J.(2012)	Blood (serum)	multiplexed cytometric cytokine assay	Trained 1	2.8 ± 2.9	Trained 1	2.5 ± 1.7	No	Pre vs post training(2) = 0.04		
			Trained 2	3.3 ± 2.3	Trained 2	4.2 ± 4				
Barry A.(2019)	Blood (plasma)	V-PLEX pro-inflammatory assays	Trained	1.6 ± 2.1	Trained	1.6 ± 1.4	1.1 ± 1.4	Control vs pwMS p < 0.05		Data extrapolated
Berkowitz S.(2019)	Blood (serum)	High Sensitivity Immunoassay Mix	Trained	5.9 ± 10.4	Trained	4 ± 7.7	2.6 ± 3.2	Pre vs post training p = 0.02Control vs pwMSp= 0.02		
Briken S.(2016)	Blood (plasma)	ELISA	Trained	12.7 ± 3.8	Trained	9.8 ± 2.8	11.9 ± 3.1	NS		Data extrapolated
Castellano V.(2008)	Blood (plasma)	multiplex immunoassay	Trained	13.8 ± 16.5	Trained	10.5 ± 15.5	14.2 ± 15.4	Pre vs post training p < 0.05		Data extrapolated
Deckx N.(2016)	Blood (serum)	ELISA	Trained	0.6 ± 0.1	Trained	0.4 ± 0.1	No	NS		
			Untrained	0.5 ± 0.1	Untrained	-				
Devasahayam A.J. (2020)	Blood (serum)	ELISA	Trained	0.5 ± 0.1	Trained	0.7 ± 0.6	No	NS		
Donia Scott A.(2019)	Blood (serum)	ELISA	Trained	10.1 ± 3.3	Trained	9.5 ± 3	No	NS		
Majdinasab N.(2018)	Blood (serum)	ELISA	Trained	3.2 ± 0.5	Trained	2.3 ± 0.4	2.4 ± 0.4	Pre vs post training p < 0.001		Data extrapolated
Schulz K.(2004)	Blood	ELISA	Trained	1.6 ± 2.5	Trained	1.3 ± 2.1	No	Pre vs post training p < 0.05		
			Untrained	1.9 ± 0.7	Untrained	-				
White L.(2006)	Blood	ELISA	Trained	6.6 ± 4	Trained	6.4 ± 9	No	NS		
Zadeh F.T.(2021)	Blood (serum)	ELISA	Trained	6.8 ± 1.5	Trained	3.2 ± 0.9	No	Trained pre vs post p = 0.001		
			Untrained	6.8 ± 1.8	Untrained	-				
Section B.2 Study	**Biological sample**	**Analytical method**	**TNF-α (pg/mL) Pre-training (mean±SD)**		**Post-training (mean±SD)**		**Healthy control group (mean±SD)**	**Statistical analyses**		**Notes**
Alvarenga-Filho H. (2016)	Blood (cells supernatants)	ELISA	Trained	17.7 ± 6	Trained	13.3 ± 4.8	9.2 ± 5	Control vs Trained p < 0.001Control vs untrained p = 0.025		Data extrapolated

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Table 1 (*continued*)

SECTION A									
			Untrained	15.6 ± 4.8	Untrained	-			
Bahmani E. (2022)	Blood (serum)	ELISA	Trained	8.9 ± 1.4	Trained	6.9 ± 0.9	No	Pre vs post training $p < 0.05$	
			Untrained	9.7 ± 1	Untrained	-			
Bansi J.(2012)	Blood (serum)	multiplexed cytometric cytokine assay	Trained 1	11.4 ± 5.2	Trained 1	10.6 ± 5.6	No	NS	
			Trained 2	10.5 ± 6.1	Trained 2	9.3 ± 6.4			
Berkowitz S.(2019)	Blood (serum)	High Sensitivity Immunoassay Mix	Trained	1.6 ± 2.2	Trained	1.2 ± 1.9	3.1 ± 3.8	NS	
Castellano V.(2008)	Blood (plasma)	multiplex immunoassay	Trained	6.8 ± 3.7	Trained	9.2 ± 5.3	4.9 ± 3.4	Pre vs post training $p < 0.05$	Data extrapolated
Deckx N.(2016)	Blood (serum)	ELISA	Trained	1.1 ± 0.1	Trained	1 ± 0.1	No	NS	
			Untrained	1.4 ± 0.2	Untrained	-			
Donia Scott A.(2019)	Blood (serum)	ELISA	Trained	26 ± 8.3	Trained	24.9 ± 7.9	No	Pre vs post training $p < 0.001$	
Kjølhede T.(2016)	Blood (plasma)	Multiplex (Bio-rad)	Trained	2 ± 0.6	Trained	2.2 ± 0.7	No	Trained vs untrained $p < 0.05$	Data extrapolated
			Untrained	2.8 ± 1.1	Untrained	-			
Majdinasab N.(2018)	Blood (serum)	ELISA	Trained	4.9 ± 0.6	Trained	4.4 ± 0.5	3.4 ± 0.5	Pre vs post training $p < 0.001$	Data extrapolated
Mokhtarzade M.(2018)	Blood (serum)	ELISA	Trained	4.2 ± 1.4	Trained	-	No	Trained vs untrained $p < 0.05$	
			Untrained	4.5 ± 0.9	Untrained	-			
Mokhtarzade M., (2017)	Blood (serum)	ELISA	Trained	4.1 ± 1.2	Trained	3.8 ± 1.7	No	Pre vs post training $p < 0.05$	
			Untrained	4.2 ± 1.2	Untrained	-			
White L. (2006)	Blood	ELISA	Trained	6.2 ± 4	Trained	3.6 ± 1	No	NS	
Section B.3 Study	**Biological sample**	**Analytical method**	**IL-10 (pg/mL) Pre-training(mean±SD)**		**Post-training (mean±SD)**		**Healthy control group (mean±SD)**	**Statistical analyses**	**Notes**
Alvarenga-Filho H. (2016)	Blood (cells supernatants)	ELISA	Trained	15.6 ± 6.3	Trained	14.3 ± 7.6	14.1 ± 5.2	NS	Data extrapolated
			Untrained	18.9 ± 5.9	Untrained	-			
Barry A.(2019)	Blood (plasma)	V-PLEX pro-inflammatory assays	Trained	1.7 ± 1.4	Trained	2.3 ± 2.2	2.3 ± 1.6	control vs MS $p < 0.05$; pre vs post in MS subjects $p < 0.05$	Data extrapolated
Berkowitz S.(2019)	Blood (serum)	High Sensitivity Immunoassay Mix	Trained	5.05 ± 8.8	Trained	2.1 ± 4.9	3.1 ± 3.8	Pre vs post training $p < 0.05$	
Deckx N.(2016)	Blood (serum)	ELISA	Trained	1.3 ± 0.2	Trained	0.5 ± 0.1	No	NS	
			Untrained	0.6 ± 0.2	Untrained	-			
Eftekhari E. (2018)	Blood (serum)	ELISA	Trained	13.1 ± 5.4	Trained	12.4 ± 7.2	No	NS	

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Table 1 (*continued*)

SECTION A									
			Untrained	13.3 ± 4.8	Untrained	-			
Kjølhede T.(2016)	Blood (plasma)	Multiplex (Bio-rad)	Trained	7.2 ± 5.4	Trained	9.6 ± 6.3	No	Trained vs untrained $p < 0.05$	Data extrapolated
			Untrained	11.8 ± 10.2	Untrained	-			
Majdinasab N.(2018)	Blood (serum)	ELISA	Trained	2.5 ± 0.4	Trained	2.2 ± 0.7	2.6 ± 0.7	Pre vs post training $p = 0.023$	Data extrapolated
Mokhtarzade M.(2018)	Blood (serum)	ELISA	Trained	9.2 ± 1.3	Trained	-	No	Trained vs untrained $p < 0.05$	
			Untrained	5 ± 0.6	Untrained	-			
Mokhtarzade M. (2017)	Blood (serum)	ELISA	Trained	2.6 ± 1.9	Trained	3 ± 1.6	No	Pre vs post training $p < 0.05$	
			Untrained	2.7 ± 1.3	Untrained	-			
White L.(2006)	Blood	ELISA	Trained	26 ± 13	Trained	15 ± 9	No	Pre vs post training $p = 0.011$	
Zadeh F.T.(2021)	Blood (serum)	ELISA	Trained	16.4 ± 2.8	Trained	23.2 ± 2.1	No	Trained pre vs post $p = 0.001$	
			Untrained	17.2 ± 1.1	Untrained	-			
Section Study	**Biological sample**	**Analytical method**	**IFN-γ (pg/mL) Pre-training (mean±SD)**		**Statistical analyses**		**Healthy control group (mean±SD)**	**Statistical analyses**	**Notes**
Alvarenga-Filho H. (2016)	Blood (cells supernatants)	ELISA	Trained	17.1 ± 7.4	Trained	15 ± 5	11.3 ± 7.4	Control vs untrained $p = 0.048$Control vs Trained $p = 0.04$;	Data extrapolated
			Untrained	16.3 ± 6.9	Untrained	-			
Berkowitz S.(2019)	Blood (serum)	High Sensitivity Immunoassay Mix	Trained	5.9 ± 10.4	Trained	4 ± 7.7	2.6 ± 3.2	Pre vs post training $P < 0.05$	
Castellano V.(2008)	Blood (plasma)	multiplex immunoassay	Trained	25.1 ± 21.7	Trained	40.1 ± 20.9	16.4 ± 16.9	Pre vs post training $p < 0.05$	Data extrapolated
Donia Scott A.(2019)	Blood (serum)	ELISA	Trained	40.7 ± 13.2	Trained	37.6 ± 11.1	No	NS	
Golzari Z. (2010)	Blood (plasma)	ELISA	Trained	8.8 ± 2.6	Trained	5.5 ± 7.2	No	Pre vs post training $p < 0.05$	Data extrapolated
			Untrained	7.4 ± 1.8	Untrained	-			
Kjølhede T.(2016)	Blood (plasma)	Multiplex (Bio-rad)	Trained	4.3 ± 2.4	Trained	6.3 ± 3.7	No	Trained vs untrained $p < 0.05$	Data extrapolated
			Untrained	9.7 ± 5.6	Untrained	-			
White L.(2006)	Blood	ELISA	Trained	8.4 ± 6.2	Trained	4.1 ± 3.1	No	Pre vs post training $p = 0.008$	

according to Table 1-sectionB.4, three studies (42.8 %)[33,41,42] analyzed IFN-γ concentrations both in pre and post training moments and comparing trained vs untrained subjects, three studies (42.8 %)[33,37,46] matched pwMS with healthy control and only one study (14.3 %)[39] reported no significant results.

3.3. Meta-analyses

We meta-analyzed data according to: 1) the levels of inflammatory biomarkers in trained vs untrained patients; 2) pre-post training change of concentrations of inflammatory biomarkers; 3) the modulation of inflammatory biomarkers by different PA protocols. In addition, complementary analyses were reported in the Supplementary materials (Supplementary materials figure A.2).

3.3.1. Inflammatory biomarkers concentrations in trained vs untrained subjects

The observed standardized mean differences ranged from − 2.82 to 1.10, with the majority of estimates being negative (90 %) (Fig. 2). The estimated average standardized mean difference was − 0.74 (95%C.I. −1.16,−0.32), the average outcome differed significantly from zero ($z = -3.47$, $p = 0.0005$) and the prediction interval for the true outcomes is given by − 2.52 to 1.04. As a whole, untrained subjects showed a negative modulation of inflammatory biomarkers concentrations when compared to trained people (−0.74, 95%C.I.–1.16,−0.32).

3.3.2. Pre-post training changes of inflammatory biomarkers concentrations

The observed standardized mean differences ranged from − 0.72 to 2.76, with the majority of estimates being positive (79 %) (Fig. 3). The average outcome differed significantly from zero ($z = 3.93$, $p < 0.0001$), and the prediction interval for the true outcomes is given by − 0.87 to 1.81. Hence, although the average outcome is estimated to be positive, in some studies the true outcome may be negative. Overall, training modulated positively inflammatory biomarkers (+0.47, 95%C.I.0.24,0.71), showing a positive influence of training on inflammatory biomarkers after training.

3.3.3. Differences in inflammatory biomarkers according to different PA protocols

The observed standardized mean differences of AEROBIC PA protocol ranged from − 5.54 to − 0.06, with the majority of estimates being

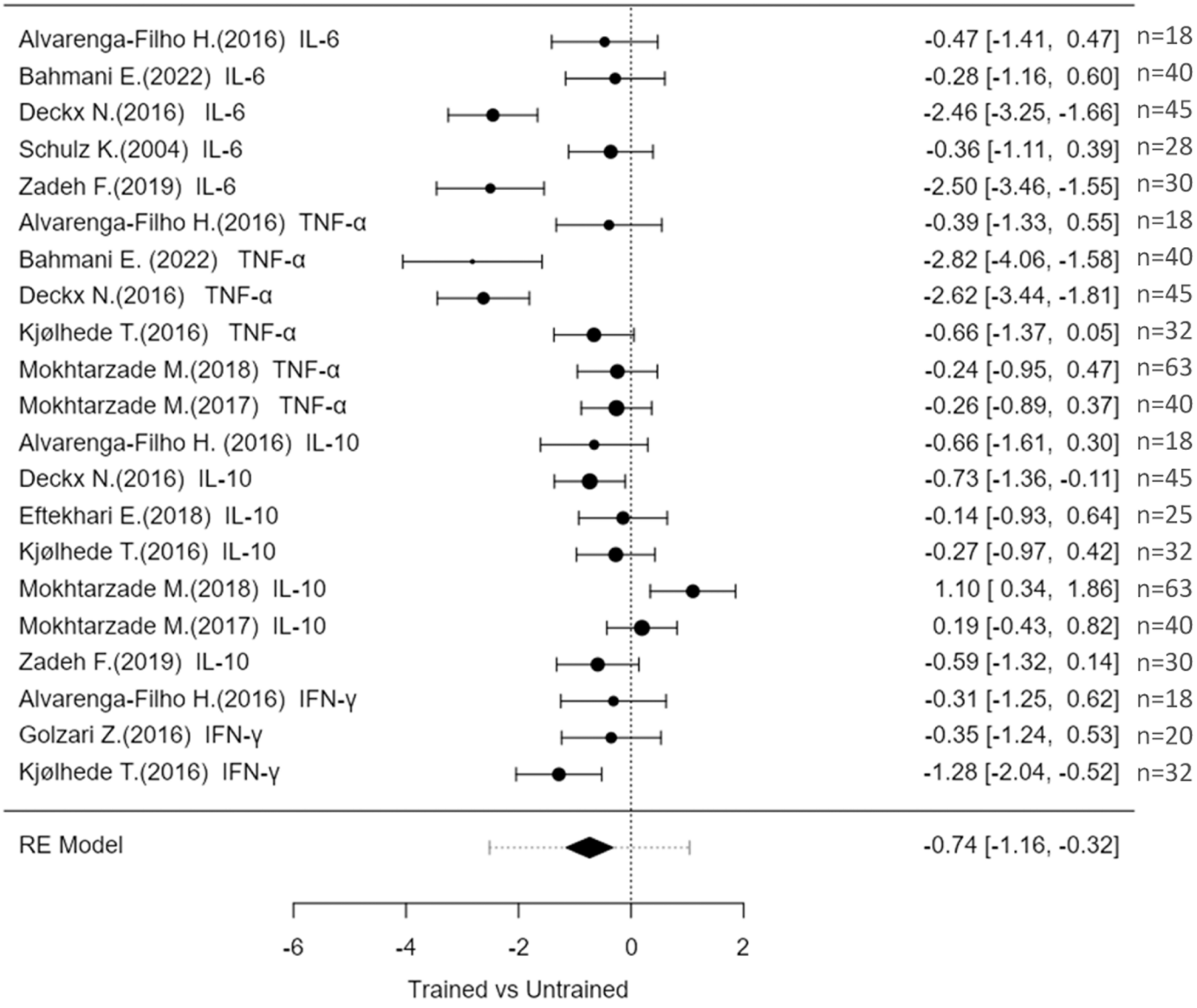

Fig. 2. Forest plot of inflammatory biomarkers (IL-6/ TNF-α/IL-10/IFN-γ) on trained vs untrained pwMS resulting from meta-analysis.

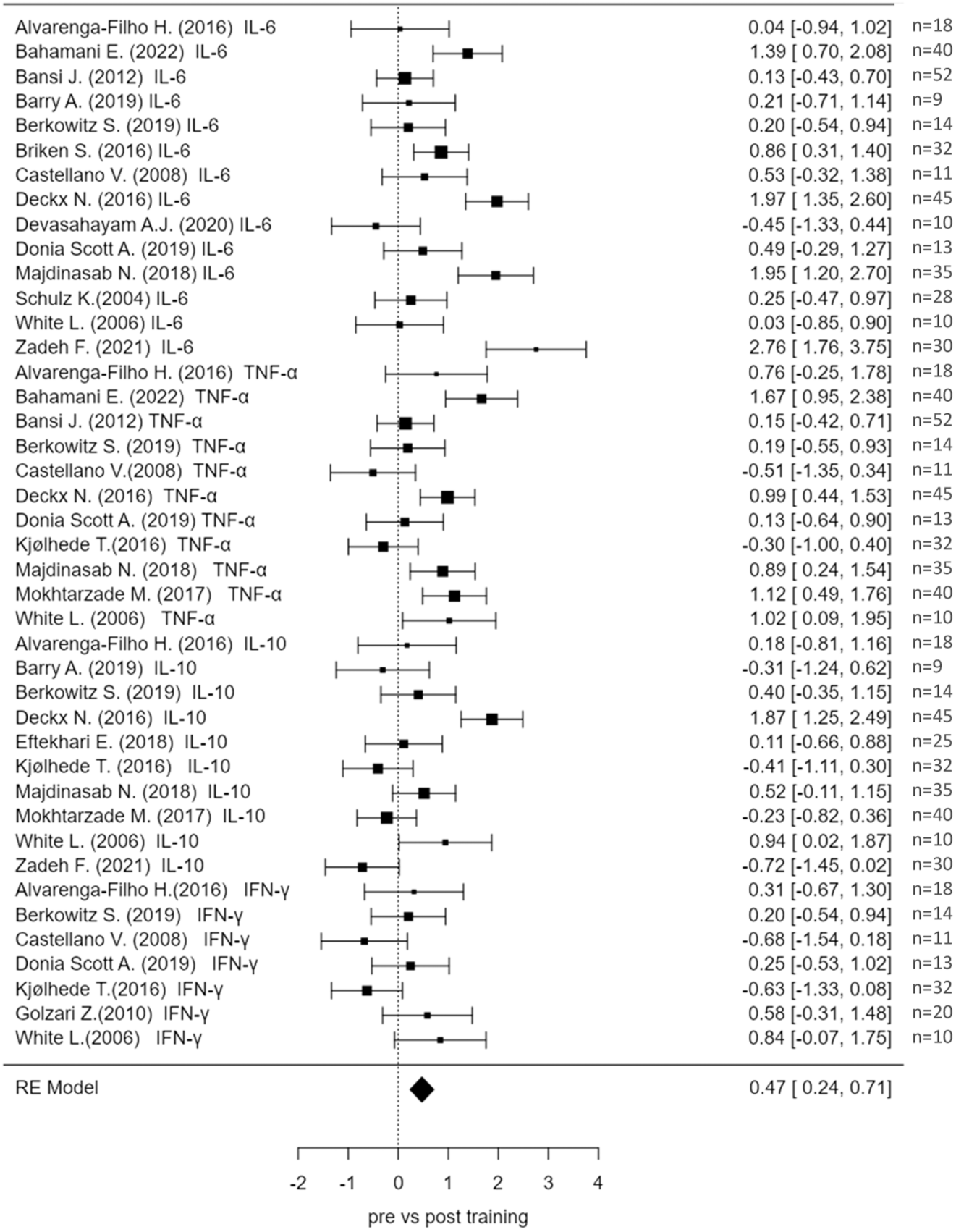

Fig. 3. Forest plot regarding pre vs post training changes of inflammatory biomarkers concentrations (IL-6/ TNF-α/IL-10/IFN-γ) on trained pwMS resulting from meta-analysis.

negative (100 %) while the observed standardized mean differences of COMBINED PA protocol ranged from − 2.50 to 3.98, with the majority of estimates being negative (64 %) (Fig. 4). The estimated average standardized mean difference based on the random-effects model was − 1.14 (95%C.I.–2.13,−0.16) and − 0.109 (95%C.I.–1.04,0.83) for AEROBIC and COMBINED PA protocol, respectively. Therefore, the average outcome differed significantly from zero (z = −2.27, p = 0.02). Overall, AEROBIC PA protocol resulted to be more efficient, positively modulating and decreasing (−1.14, 95%C.I.–2.13,−0.15) the inflammatory biomarkers. Regarding COMBINED PA protocol, the meta-analysis resulted to be not statistically significant, even if the trend showed was similar to AEROBIC PA protocol, enhancing the positive influence and upregulation of PA training on inflammation and inflammatory biomarkers.

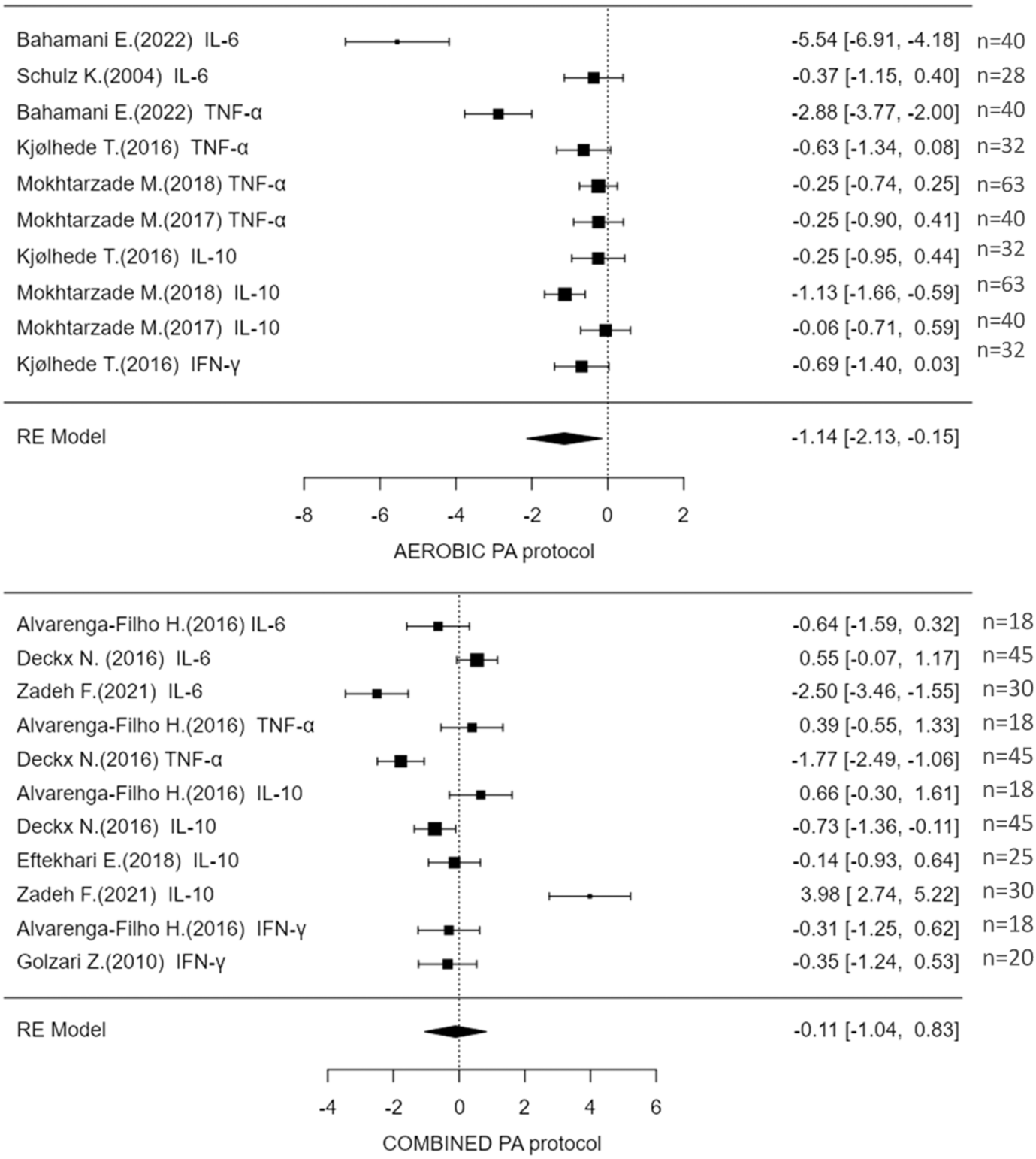

Fig. 4. Forest plot of inflammatory biomarkers concentrations (IL-6/ TNF-α/IL-10/IFN-γ) according to PA protocols, AEROBIC PA protocols (top) and COMBINED PA protocols (down), resulting from meta-analysis.

4. Discussion and conclusion

The promotion of active lifestyle is a major issue in the treatment of a broad range of immune-mediated diseases, including autoimmune diseases such as MS.[27,59] For decades, PA was not in MS management because it was thought that exercise could negatively modulate the risk of symptoms exacerbations and fatigue,[21] even if PA was considered a feasible and safe supportive strategy to alleviate symptoms[20,21] in pwMS.[60] Although the development of ad hoc guidelines for promoting PA,[24,29] recent literature[18,24,29,61] confirms that pwMS are still physically less active than the healthy population. This seems plausible because neuromuscular functioning decreases with disease progression,[22,62] accumulating fatigue and disability and diminishing the motivation to be physically active.[22]

From the biomolecular perspectives, the human physiology is inherently associated with PA which affects many interconnected cellular systems such CNS or the immune system.[63] Since MS is a neuroinflammatory disease, mitigation of both peripheral/central inflammation is of high importance and exercise may represent a complementary therapy approach without side effects.[21,64] Regular exercise has been shown to decrease and upregulate the systematic inflammation in pwMS,[33,34] ameliorating disease-specific symptoms and activity.[65,66] Interestingly, only high-intensity interval training reached significant results,[34,43,47,48] adding evidence that a steady high cardiovascular could reach anti-inflammatory and neuroprotective effects.

To our knowledge, this is the first review study that systematically investigated the role of PA as a complementary therapy in the management of MS. In particular, this review aimed to underline if and how PA protocol could positively upregulate inflammatory biomarkers. Despite the large availability of measures, our review highlights the lack of standardized PA protocols among researchers, making inter-studies comparison challenging. These issues mainly concern also the selecting criteria used both for sampling and biological protocols, as well as the comparability of analytical methods and procedures. Furthermore, this review identified a lack of high-quality evidence that PA exerts consistent effects on inflammatory biomarkers measures, mainly due to different biological protocols and type of PA adopted. This could be due to the low sample size, heterogeneity of the included study population, and short duration of existing studies (averaging 8 weeks). This raises the question if the available studies are appropriate to address whether PA positively upregulates inflammation levels, since longer observations periods are needed to validly evaluate the late-terms effect of exercise on pwMS.

Despite these considerations, the abundance of cross-sectional studies in this review showed beneficial short effects of a high level of PA on inflammatory biomarkers concentrations in pwMS. While the exact mechanisms by which PA may reduce and upregulate inflammation are not still entirely understood, some data suggest that PA may lead to improvements in inflammatory status over time. These factors include: 1) reductions in immune cell production and mediators and 2) locally immune function adaptations. The activation of inflammatory pathways elicited by exercise makes it almost counterintuitive that regular PA would reduce chronic inflammation, even if it is now evident that an acute inflammatory response plays a role in physiological adaptations, such as muscle resistance. Contracting skeletal muscle produces several cytokines, most notably IL-6, which mediate metabolic changes during exercise.[33,34,36,38,40,44,45,48,49,66] Thus, acute exercise activates an immune response, but the effects are primarily anti-inflammatory, enhancing lipid and glucose metabolism. In turn, regular exercise can lead to lower basal levels of circulating pro-inflammatory markers, also reducing the inflammatory response to acute exercise. Many works[33,34,43] well support that a long, structured, aerobic PA protocol positively modulated pro-inflammatory biomarkers concentrations.

There are several limitations in this systematic review. First, we did not assess the risk of bias for included cross-sectional, non-randomized controlled, and pilot studies. Second, the majority studies examined the effect of a predominantly exercise intervention while the improvement of daily activities were not included. Third, the relatively short duration of the studies needs to be considered. Fourth, the study populations of the included studies were not uniform in size, sex-quote representation and MS subtype, meaning inhomogeneity.

In conclusion, persistent, low-grade inflammation is an important contributor to the pathophysiology of several chronic diseases, such as MS. Given these widespread deleterious health effects of an augmented inflammatory state, identification of non-pharmacological complementary therapies that could reduce and upregulate inflammation is critical. Consistent data from the recent literature on pwMS show a link between higher levels of PA and a reduction of inflammatory biomarkers, as well as increasing aerobic PA could be more effective for reducing chronic inflammation.

Despite the wide heterogeneity among studies, the present review concluded that PA seems more prone to positively modulate inflammation and inflammatory levels in pwMS. Indeed, even if all subjects were pwMS, trained subjects had lower inflammatory biomarkers levels if compared to untrained ones. In addition, aerobic PA is more effective in the upregulation of inflammatory biomarker concentrations when compared to the aerobic one. These peculiar hallmarks strengthen the role of PA as an effective and positive non-pharmacological complementary therapy in MS and continue stimulating research efforts. Epidemiologic approaches focusing on long-term effects and follow-up and large-scale studies could support further research in this field, adding trials targeting the magnitude and persistence in the long-period of the effect of PA on inflammatory mediators, and the amount of exercise necessary to produce clinically meaningful reductions in inflammation and, possibly, in neuroinflammation.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.ctim.2024.103040.

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