Inflammopharmacology (2024) 32:927–944
https://doi.org/10.1007/s10787-023-01414-7

Inflammopharmacology

REVIEW

The effects of melatonin supplementation on neurobehavioral outcomes and clinical severity in rodent models of multiple sclerosis; a systematic review and meta-analysis

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Received: 25 June 2023 / Accepted: 13 December 2023 / Published online: 22 January 2024

Abstract

Background Through the antioxidant and anti-inflammation pathways, melatonin is proposed as a safe and effective intervention in neurological diseases. This study aims to evaluate the effects of melatonin supplementation on the neurobehavioral and clinical outcomes in animal models of multiple sclerosis (MS).

Methods This study was conducted following the PRISMA statement. Animal studies that reported the effects of melatonin in preclinical MS models, including the experimental autoimmune encephalomyelitis (EAE) and cuprizone model for demyelination are included in this study. A systematic search in PubMed, Web of Science, Embase, and Scopus up was conducted in April 2023. The collaborative Approach to Meta-Analysis and Review of Animal Experimental Studies (CAMARADES) critical appraisal tool was used for the quality assessment of the studies and the quantitative synthetizes were conducted using the comprehensive meta-analysis software.

Results Out of 542 studies, finally 21 studies, including 14 studies in the EAE model and 7 studies of the toxic demyelination method with cuprizone were included. The route of administration was intraperitoneal in 18 studies, oral in 2 studies, and subcutaneous in 1 study. The quantitative synthesis of the EAE clinical severity scale was associated with significant differences (standardized mean difference [SDM]: − 2.52; − 3.61 to − 1.42; p value < 0.01). In subgroup analyses, the difference was statistically significant in the mouse subgroup (SMD: − 2.60; − 3.74 to − 1.46; p value < 0.01).

Discussion This study encountered that melatonin may be associated with improved behavioral and cognitive outcomes of preclinical models of MS with acceptable safety profiles.

Funding The research was supported by the Student Research Committee, Tabriz University of Medical Sciences (grant number: 71005).

Keywords Melatonin · Multiple sclerosis · Rodent · Systematic review · Meta-analysis

Introduction

Multiple sclerosis (MS) is a progressive inflammatory autoimmune disease of the myelin that influences around 2.3 million people worldwide (Alghamdi and AboTaleb 2020). It is the most common demyelination disease of the central nervous system (CNS) and usually affects young adults (Alghamdi and AboTaleb 2020; Long et al. 2018). Demyelination interrupts neuronal signals, causing abnormalities correlated with the affected neurons (Alghamdi and AboTaleb 2020). The exact etiology of MS is still not comprehended, but it has been revealed that genetic, environmental, and viral factors may contribute to the initiating of an aberrant autoimmune attack, which may cause myelin and axonal damage (Long et al. 2018).

Animal models of MS have been crucial for dissecting the effects of interventions (Burrows et al. 2019). Experimental autoimmune encephalomyelitis (EAE) is an animal model that resembles demyelinating in the CNS and autoimmune inflammation, which is a well-known model to discover the pathogenesis of MS (Gold et al. 2006). The cuprizone model is also a representative of toxic demyelination which is a valuable tool for MS preclinical studies (Torkildsen et al. 2008), especially for the progressive phenotype of the disease (Zhan et al. 2020).

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Melatonin, N-acetyl-5-methoxytryptamine, is a neuromodulator produced in the pineal gland (Ramos Gonzalez et al. 2021). It is a small, nonpolar, amphiphilic molecule that can traverse any membrane in the body, involving cell membranes, mitochondrial membranes, and the blood–brain barrier (BBB) (Abo Taleb & Alghamdi 2020; Long et al. 2018). Melatonin receptors are expressed on the cell membranes of monocytes, B cells, and CD4 + and CD8 + T cells (Ramos Gonzalez et al. 2021). It has been revealed that melatonin displays multifunctional properties, involving antiaging, as an antioxidant, and also has modulatory effects on both adaptive and innate immunity (Long et al. 2018). Numerous studies have described alterations in melatonin levels and its leading metabolite, 6-sulphatoxymelatonin, in MS patients (Long et al. 2018). The neuroprotective effects of melatonin through antioxidant and anti-inflammatory efficacy may lead to improved neurobehavioral, motor, and cognitive functions in various neurological pathologies (Potes et al. 2023).

Multiple studies examined the effects of melatonin administration in rodent models of MS, which encouraged us to perform this systematic review of the preclinical studies. This study is focused on the severity of the disease, as well as the cognitive and behavioral outcomes of melatonin supplementation on rodent models of MS.

Methods

This systematic review was performed based on the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) statement (Page et al. 2021).

Inclusion and exclusion criteria

All original animal study publications that were related to our review topic and reported the severity of the disease, cognitive and behavioral outcomes of melatonin supplementation in MS models are included in this study. Letters to the editors, conference papers, studies with overlapping data, human studies, reviews, and articles published in languages other than English were excluded.

Search strategy

Two reviewers (A.N. and S.M.) separately conducted a systematic literature search in the online databases including MEDLINE via PubMed, Web of Science, Embase, and Scopus up to April 2023. (melatonin) AND ((experimental AND autoimmune AND encephalomyelitis) OR "multiple sclerosis" OR EAE OR (allergic AND encephalomyelitis) OR Cuprizone) keywords were searched without any limitation. Also, to find possibly suitable studies, researchers explored the reference lists of related studies found.

Selection process and data extraction

Three authors (S.M., H.R., and A.N.) independently evaluated the titles and abstracts of the publications attained after duplicate removal by the EndNote reference manager. The full texts of appropriate studies were independently checked for eligibility by two authors (A.N. and E.N.). The following information was extracted from the included articles: first author, year of publication, number of animals, animal strain, study duration, method of MS induction, age of animals, melatonin dosage, route and duration of administration, outcome assessment scales, results, and main conclusion. Data extraction was conducted by one author (E.N., A.M., A.E., or H.R.) and double-checked by two other authors (A.N., R.M-H., S.M., or S.R.). A third author controlled any disagreements between authors in all stages (S.S-E.).

Quality assessment

Two authors (P.V. and R.M-H.) independently assessed the quality of the included studies using the Collaborative Approach to Meta-Analysis and Review of Animal Experimental Studies (CAMARADES 2023) checklist and referred the disagreement to another author (S.S-E.). The checklist includes 10 items that assess publication in a peer-reviewed journal, control of temperature, random allocation to treatment and control groups, allocation concealment, blinded assessment of outcome, use of anesthetics without significant intrinsic neuroprotective activity, animal model, sample size calculation, compliance with animal welfare regulations, and a statement of potential conflict of interests.

Meta-analysis

The quantitative synthetizes was conducted using the third version of comprehensive meta-analysis (CMA3) software with 95% confidence intervals (CIs) and 0.05 level of significance for p value. Studies that have reported the clinical severity scale scores after melatonin supplementation in the EAE model, were included in the meta-analysis. Mean, standard devotion (SD), and the number of animals in each group were utilized for calculating the effect size in each study. Standard error values are also converted in SD with the following formula:

$$SD = SE * \sqrt{n}$$

Also, subgroup analyses based on strain type (mouse or rat) were conducted and the outcome is reported in

standardized mean difference (SDM) with 95% CIs. Due to the high level of heterogeneity based on the I^2 index, random effect model analysis was utilized. The final results of the meta-analyses are presented in the forest plot.

Results

Literature search and study description

542 studies were screened in the title/abstract stage after the duplicate studies were taken out of the Eq. 26 articles' full texts were examined, and 21 studies were found to be included in this systematic review. Three studies were excluded as did not assess the effects of melatonin in the EAE model (Dokoohaki et al. 2017; Mahmoodi et al. 2020; Sharif et al. 2017) and two studies did not report any severity, behavioral or cognitive outcomes (Majid Ghareghani et al. 2023a, b; Kashani et al. 2014). Further information about the selection process is indicated in the PRISMA flow diagram (Fig. 1).

General characteristics of the studies and risk of bias

Out of 21 included studies; 14 were conducted based on the EAE model of MS and the rest 7 studies, utilized the toxic demyelination method with cuprizone. Melatonin was administered orally in two studies, subcutaneously in one, and intraperitoneally in the rest of the studies (18 studies). Detailed methods of MS inductions, the duration of the treatment, and the dosage of the medication varied between studies, as presented in Table 1. The following tests were used to assess the cognitive and behavioral outcomes of rodent models of MS: Rotarod performance test, Grid floor activity cage test, Y-Maze test, Grip strength test, Open field test

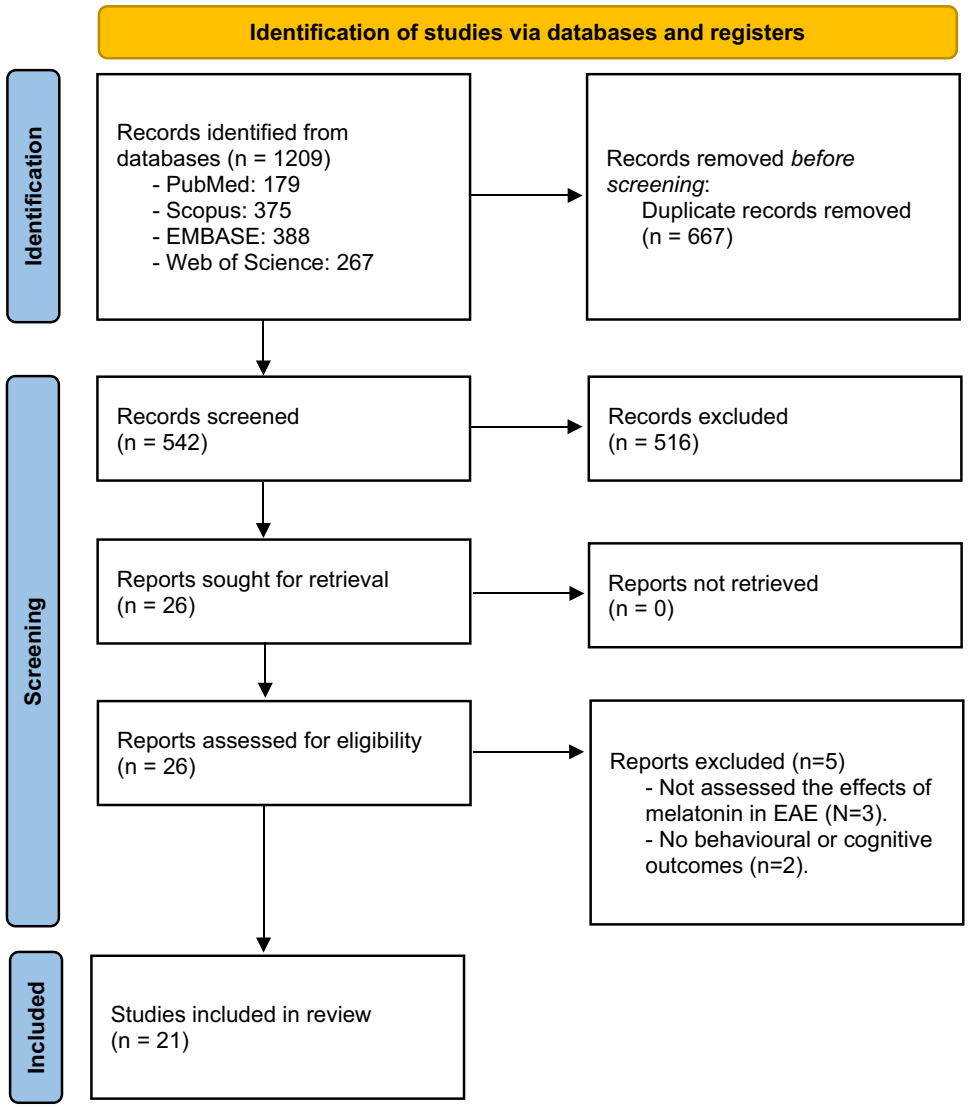

Fig. 1 PRISMA flow diagram

Table 1 The characteristics and main findings of the included studies

study (APA citation)	Number of animals	Animal strain	Study duration	Model and method of induction	Age	Melatonin			Outcome assessment scales	Results
						Route	dosage	duration		
Ortíz et al., 2022 (Ortiz et al. 2022)	72	Male adult Sprague–Dawley rats	20 days	EAE; Rats were immunized by a single IP injection of a homogenate of the spinal cord and pig brain (25 mg each one) suspended in sterile physiological saline solution and emulsified with 50 µL of complete Freund's adjuvant, containing 1 mg/mL heat-killed Mtb strain H37Ra	–	IP	20 mg/kg/day	20 days	Cumulative disease index score, Clinical EAE score	The cumulative index score was significantly reduced in EAE rats treated with melatonin alone or in combination with glatiramer acetate and IFNβ-1b. Treatments using glatiramer acetate and IFNβ-1b, alone or in combination with melatonin, also showed a decrease in clinical symptoms, although their effect was lower than that observed with the administration of melatonin alone
Escribano et al. 2022 (Escribano et al. 2022)	25	Male Dark Agouti rats	65 days	EAE; Injecting SQ, at the dorsal base of the tail, 100 µl of a solution containing 150 µg of MOG in emulsified 1:1 in complete Freund's adjuvant completed with 400 µg of heat-inactivated Mtb, during 14 days	2 months	IP	1 mg/kg, 5 days a week	51 days	severity scale	At 65 days of the experiment, the EAE score had increased concerning 14 days, whereas the treatment with melatonin made the difference in values at 65 days compared to 14 days

Table 1 (continued)

study (APA citation)	Number of animals	Animal strain	Study duration	Model and method of induction	Age	Melatonin			Outcome assessment scales	Results
						Route	dosage	duration		
Chen et al. 2016 (Chen et al. 2016)	21	C57BL/6 mice	Induction + 13 days	EAE; Each C57BL/6 mouse was immunized with 100 µg of MOG in 100 µL of an emulsion of Complete Freund's adjuvant and 400 µg of Mtb H37Ra. Each mouse also received an IP injection of 500 ng of pertussis toxin on days 0 and 2 after immunization	6–8 weeks	SQ	200 mg/kg (3-times injections)	13 days	Clinical EAE score	The clinical score was significantly lower in the melatonin-treated, MOG-induced EAE mice compared with the solvent-treated controls
Ghareghani et al. 2018a, b, c (Ghareghani et al. 2018a, b, c)	40	Adult female C57BL/6 mice	26 days	EAE; The induction kit contains MOG35–55 antigen in emulsion with complete Freund's adjuvant, enriched Mtb + IP injection of pertussis toxin	6–8 weeks	IP	10 mg/kg/day	10 days	Clinical signs score	Melatonin alone or in combination with baclofen reduced clinical scores of EAE
Ghareghani et al. 2017 (Ghareghani et al. 2017)	15	Female Lewis rats	18 days	EAE; a mixture of 1 g Guinea Pig Spinal Cord in 1 ml PBS and Complete Freund's adjuvant and 1 mg/ml Mtb enriched	5–6 weeks	Oral	10 mg/kg/day	6 days	EAE severity scale	Melatonin resulted in a considerable increase in EAE severity, at the onset of EAE symptoms. Paralysis of the melatonin-treated rats progressed to both hind limbs in addition to forelimb weakness, with a maximal score of 6

Table 1 (continued)

study (APA citation)	Number of animals	Animal strain	Study duration	Model and method of induction	Age	Melatonin			Outcome assessment scales	Results
						Route	dosage	duration		
Gonzalez et al., 2021 (Ramos Gonzalez et al. 2021)	128	Male Sprague–Dawley rats	12 days	EAE; administering 150µL of a homogenate containing 25µL of pig spinal cord homogenate, 25µL of pig brain homogenate, and 100µL of Freund's Complete adjuvant	60 days	IP	10 mg/kg/day	12 days	Clinical scale	EAE appeared sooner in the animals treated with melatonin than in the group with no treatment. Treatment with melatonin improved symptoms more rapidly than GA, achieving total recovery of the animals
Taleb & Alghamdi, 2020 (Abo Taleb & Alghamdi 2020)	78	SWR/J mice	9 weeks	cuprizone administration for 5 weeks	8–10 weeks	IP	0.5 ml (80 mg/kg)	9 weeks	Grip strength test, Open field	Melatonin ameliorated weight gain, and improved motor activity in mice during the demyelination and re-myelination stages
I. F. Labunets, N. A. Utko, O. K. Toporova, et al., 2021 (I. F. Labunets, N. A. Utko, O. K. Toporova, et al., 2021)	80	Male 129/Sv mice (H-2b genotype)	3 weeks	cuprizone model of demyelination	6–7 months	IP	1 mg/kg/day	11 days	Open field, Rotarod test	Melatonin injection enhanced the effect of hMMSCs on grooming activity and increased the glutathione reductase activity
I. Labunets et al. 2021a, b, c (I. Labunets et al. 2021a, b, c)	100	129/Sv mice	3 weeks	cuprizone model of demyelination	15–17 months	IP	1 mg/kg/day	11 days	Open field, Rotarod test	Melatonin injections enhance the effects of UC-MMSCs on the motor and emotional activity of animals

Table 1 (continued)

study (APA citation)	Number of animals	Animal strain	Study duration	Model and method of induction	Age	Melatonin			Outcome assessment scales	Results
						Route	dosage	duration		
Long et al. 2018 (Long et al. 2018)	32	Female C57BL/6 mice	4 weeks	EAE; Mice were injected SQ at one side of the flank with 250 µg MOG35–55 peptide in complete Freund's adjuvant and 64 mg/ml of heat-killed Mtb. At 0 h and 48 h after MOG injection, 500 ng pertussis toxin in 100 uL PBS was injected IP	8– 10 weeks	IP	10 mg/kg	to peak of disease	Clinical scores	The incidence of EAE was insignificantly higher in the EAE-untreated group than in the melatonin group. The neurological deficit at the peak stage of EAE in the EAE-untreated group was greater than that of the melatonin group
Ghareghani et al. 2019 (Ghareghani et al. 2019)	32	Adult female C57BL/6 mice	30 days	EAE; Mice were immunized with MOG35–55. MOG35–55 was emulsified in complete Freund's adjuvant, enriched Mtb. Briefly, on day 1, each mouse was injected with 10 µl of MOG emulsion SQ over the flank and then injected IP with 200 ng of pertussis toxin, diluted in sterile PBS. On day 3, a second 200 ng booster pertussis toxin injection was given	6–8 weeks	IP	low-dose: 476 µg/kg/day high-dose: 10 mg/kg/day	13 days	Clinical score, Cumulative neurological disability	The mean peak clinical score in untreated EAE mice was as high as 3.5 at 30 days, which declined to 2.1 and 1.6 in low and high-melatonin-treated mice, respectively. cumulative neurological disability was significantly lower in mice treated with low or high melatonin, compared to untreated EAE mice

Table 1 (continued)

study (APA citation)	Number of animals	Animal strain	Study duration	Model and method of induction	Age	Melatonin			Outcome assessment scales	Results
						Route	dosage	duration		
Álvarez-Sánchez et al. 2015 (Álvarez-Sánchez et al. 2015)	35	Female C57BL/6 mice	15 days	EAE; Mice immunized SQ in each hind leg with 100 µg of MOG35–55, hereafter referred to as pMOG, emulsified in complete Freund's adjuvant containing 50 µg of heat-killed Mtb (H37Ra, ATCC 25177). EAE was induced as above, followed by two IP doses of 400 ng of pertussis toxin on days 0 and 2	8-week	IP	doses ranging from 20 to 80 mg/kg	15 days	EAE severity scale, Clinical signs, Cumulative scores	Both the maximum clinical signs and the cumulative scores were significantly lower in melatonin-treated mice compared to control animals
Vakilzadeh et al. 2016 (Vakilzadeh et al. 2016)	28	Male C57BL/6 mice	6 weeks	Mice were fed with a diet containing 0.2% mixed cuprizone into ground standard rodent chow for 5 weeks	8—9 weeks	IP	50 or 100 mg/kg	7 days	Open field, Tail-flick test	Two different concentrations of melatonin dose-dependently enhanced distance moved and movement velocity in mice compared to the cuprizone group. Application of melatonin at both 50 and 100 mg/kg dose-dependently and significantly increased the nociception latency

Table 1 (continued)

study (APA citation)	Number of animals	Animal strain	Study duration	Model and method of induction	Age	Melatonin			Outcome assessment scales	Results
						Route	dosage	duration		
Wen et al. 2016 (Wen et al. 2016)	48	Female C57BL/6 J mice	28 days	EAE; Animals were injected SQ with 200 µg MOG 35–55 peptide emulsified in complete Freund's adjuvant containing 500 µg Mtb. Immediately and 24 h after immunization, mice were administered 200 ng pertussis toxin IP	7-week	IP	20 mg/kg	-	Clinical severity of EAE	Treatment with melatonin after symptom onset improved motor function, but the effect was not as great as seen with the groups pre-treated with the two compounds. The clinical scores were significantly reduced from days 22 to 28 after EAE induction in groups treated with melatonin
Alghamdi & AboTaleb 2020 (Alghamdi & AboTaleb 2020)	30	Male SWR/J mice	5 weeks	cuprizone model of demyelination	-	IP	80 mg/kg	-	Tail-flick, Locomotor activity, Novel object recognition test	Melatonin prolonged nociception latency, improved locomotor activity, and improved novel object recognition in the MS mouse model

Table 1 (continued)

study (APA citation)	Number of animals	Animal strain	Study duration	Model and method of induction	Age	Melatonin			Outcome assessment scales	Results
						Route	dosage	duration		
I. F. Labunets, N. A. Utko, & O. K. Toporova, 2021 (I. F. Labunets, N. A. Utko, & O. K. Toporova, 2021)	76	129/Sv male mice (Genotype H-2b)	3 weeks	Toxic cuprizone model of demyelination	6–7 months/ 15–17 months	IP	1 mg/kg	11 days	Open field, Rotarod test	After melatonin injections, the number of crossed squares and boluses in experimental mice increases to the values of intact animals. The injection of hMMSCs in combination with melatonin enhanced their effect on the behavior parameters in mice of both age groups and, in addition, increased the number of boluses and rotarod retention time in adult mice and the number of crossed squares and grooming activity in aging animals
Gharib et al., 202 (Gharib et al. 2022)	50	male C57BL/6	8 weeks	feeding 0.2% (w/w) cuprizone (Sigma) in-ground breeder chow	8-week	IP	10 mg/kg	3 weeks	Rotarod test, Grid Floor activity cage, Y-Maze test	The efficacies of melatonin single dose were 45.32% across the activity cage. Treatment with Melatonin didn't improve Y-Maze test results

Table 1 (continued)

study (APA citation)	Number of animals	Animal strain	Study duration	Model and method of induction	Age	Melatonin			Outcome assessment scales	Results
						Route	dosage	duration		
Ghareghani et al. 2022 (Ghareghani et al. 2022)	32	Adult female C57BL/6	23 days	EAE; The immunization solution containing the immunogenic epitope MOG35-55 was emulsified with complete Freund's adjuvant (CFA, Sigma Aldrich) and Mtb, and was injected SQ over the flank. Booster pertussis toxin was injected IP on the day of immunization and 3 days later	10–12-week	IP	10 mg/kg	10 days	Clinical EAE score	Results of neurological disability scores showed lower scores in the whole experimental period for melatonin treatment
Jand et al. 2022 (Jand et al. 2022)	68	Female C57BL/6 mice	21 days	EAE; Mice were immunized with 300 µg of MOG35-55 dissolved in PBS and emulsified with an equal volume of complete Freund adjuvant. Additionally, 300 ng of pertussis toxin was injected IP, into all animals, on days 0 and 2	6–8 weeks	IP	0.1, 1, 5, 10 mg/kg	9 days	clinical severity of EAE, Cumulative disease index score	Melatonin lowers EAE clinical score but there was no difference between groups that were treated with different dosages of melatonin

Table 1 (continued)

study (APA citation)	Number of animals	Animal strain	Study duration	Model and method of induction	Age	Melatonin			Outcome assessment scales	Results
						Route	dosage	duration		
Ghareghani et al. 2018a, b, c (Ghareghani et al. 2018a, b, c)	24	Adult female C57BL/6 mice	30 days	EAE; C57BL/6 mice were immunized with MOG35 –55 emulsified in complete Freund's adjuvant, enriched with Mtb. on day 1, each mouse was injected with 10 µl MOG emulsion SQ over the flank and injected with 200 ng of pertussis toxin PTX IP. On day 3, a second 200 ng booster PTX injection was given	6–8 weeks	IP	10 mg/kg	13 days	Clinical severity of EAE	melatonin decreased EAE severity and had a beneficial role in improving EAE outcomes
Kang et al. 2001 (Kang et al. 2001)	17	Sprague–Dawley rats of both sex	21 days	EAE; each rat was inoculated in both hind footpads with 100 µl of an emulsion containing 1 mg of fresh rat spinal cord homogenate in PBS per ml of complete Freund's adjuvant containing Mtb	8–12 weeks	oral	5 mg/kg	14 days	Clinical severity of EAE	Melatonin significantly reduced the severity and duration of paralysis compared with the control

PBS: phosphate-buffered saline; *EAE*: experimental autoimmune encephalomyelitis; *MS*: Multiple sclerosis; *hMMSCs-2*: human fibroblast growth factor-2; *hMMSCs*: human umbilical cord multipotent mesenchymal stromal cells; *MOG*: myelin oligodendrocyte glycoprotein; *Mtb*: Mycobacterium tuberculosis; *SQ*: subcutaneously; *IP*: intraperitoneal

(Locomotor activity), Severity score, Clinical signs score, EAE severity scale, Cumulative neurological disability, Tail-flick test, and Novel Object Recognition (NOR). The results of quality assessments are presented in Table 2.

Results of individual studies

Melatonin treatment at pharmacological doses (10 mg/kg/d), compared to phosphate-buffered saline (PBS) treated rats, significantly increased the severity of EAE at the onset of symptoms in female Lewis rats (Ghareghani et al. 2017). This study was the only one which reported harmful outcomes with melatonin treatment in the acute phase of MS and found that melatonin exacerbates EAE by enhancing the serum levels of lactate. Another study of this research group assessed the effects of the combination of 10 consecutive days of treatment with melatonin (10 mg/kg/day) and muscle relaxant drug, baclofen (10 mg/kg/day) in EAE. This study found the effectiveness of melatonin treatment alone, or in combination with baclofen, by enhancing re-myelination and reducing inflammation and oxidative stress (Ghareghani et al. 2018a, b, c). Another investigation of the EAE severity as well as factors affecting bone metabolism in MS found that melatonin reduced EAE severity and improved outcomes related to EAE. Additionally, EAE mice treated with melatonin had a reduction in osteoporosis risk (Ghareghani et al. 2018a, b, c). A 2019 study compared untreated EAE mice, with animals treated with physiological or pharmacological melatonin. Animals in the experimental group had a considerably decreased overall level of neurological impairment. This study indicated a negative impact of melatonin on EAE recovery by delaying the re-myelination process by inhibiting the pyruvate dehydrogenase complex (Ghareghani et al. 2019). In light of the previously mentioned study, the same research team in a study of 32 adult (10–12 weeks old) female C57BL/6 mice, showed melatonin (10 mg/kg/day) alone and in combination with PDK4 inhibition by diisopropylamine dichloroacetate, may complement one another's therapeutic effects and offer potential as a novel treatment strategy for MS and other demyelinating disorders (Ghareghani et al. 2022).

Ortiz et al. showed that the injection of melatonin alone or in combination with interferon β-1b (IFNβ-1b) or glatiramer acetate (GA) reduced the maximal disease score

Table 2 Quality assessment of included studies based on CAMAREDES checklist

Author, years	(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)	Score
(Ramos Gonzalez et al. 2021)	X	X	–	–	–	–	–	–	X	–	**3**
(Ortiz et al. 2022)	X	X	X	–	–	–	–	–	X	–	**4**
(Labunets, Utko, Toporova, 2021c)	X	X	–	–	–	–	–	–	X	–	**3**
(Wen et al. 2016)	X	X	–	–	–	–	–	–	X	–	**3**
(Labunets et al. 2021a, b, c)	X	–	–	–	–	–	–	–	X	–	**2**
(Álvarez-Sánchez et al. 2015)	X	–	–	–	X	–	–	–	X	–	**3**
(Chen et al. 2016)	X	–	–	–	–	–	–	–	X	–	**2**
(Ghareghani et al. 2017)	X	X	X	–	–	–	–	–	X	–	**4**
(Alghamdi and AboTaleb 2020)	X	X	X	–	–	–	–	–	X	–	**4**
(Ghareghani et al. 2019)	X	X	X	–	–	–	–	–	X	–	**4**
(Abo Taleb and Alghamdi 2020)	X	X	X	–	–	–	–	–	X	–	**4**
(Long et al. 2018)	X	X	–	–	X	–	–	–	X	–	**4**
(Muñoz-Jurado et al. 2022)	–	X	–	–	–	–	–	–	X	–	**2**
(Ghareghani et al. 2018a, b, c)	X	X	X	–	–	–	–	–	X	–	**4**
(Vakilzadeh et al. 2016)	X	X	X	–	–	–	–	–	X	–	**4**
(I. Labunets et al. 2021a, b, c)	–	X	–	–	–	–	X	–	X	–	**3**
(Kang et al. 2001)	X	–	–	–	–	–	–	–	–	–	**1**
(Jand et al. 2022)	X	X	X	–	X	–	–	–	X	–	**5**
(Gharib et al. 2022)	X	X	–	–	–	–	–	–	X	–	**3**
(Ghareghani et al. 2018a, b, c)	–	X	X	–	–	–	–	–	X	–	**3**
(Ghareghani et al. 2022)	X	X	X	–	X	–	–	–	X	–	**5**

Bolds are the overall scores of the studies

Studies fulfilling the criteria of: (1) peer-reviewed publication; (2) control of temperature; (3) random allocation to treatment or control; (4) allocation concealment; (5) blinded assessment of outcome; (6) use of anesthetic without significant intrinsic neuroprotective activity; (7) animal model (aged, diabetic, or hypertensive); (8) sample size calculation; (9) compliance with animal welfare regulations; and (10) statement of potential conflict of interests

and the cumulative index score in EAE and ameliorated immune-mediated Brain damage, delaying the onset and progression of the damage (Ortiz et al. 2022). Fifty-one days of administration of melatonin (1 mg/kg) either used alone or in combination with other conventional medications was found to be an effective treatment for MS in Escribano et al.'s study of 25 male Dark Agouti rats (Escribano et al. 2022). The clinical score was considerably lower in the melatonin-treated EAE animals compared to the solvent-treated controls after 13 days after subcutaneous injection, according to Chen et al., study. This study found that high-dose melatonin (1 mg/kg, 5 days a week) significantly improves the severity of established EAE by modulation of adaptive immunity (Chen et al. 2016). Gonzalez et al.'s study found that melatonin treatment reduced symptoms more quickly than GA in male Sprague–Dawley rats with relapsing–remitting EAE. Regarding the first relapse-remission cycle, IFN-β and GA had better result than melatonin (Ramos Gonzalez et al. 2021).

Additional effects of melatonin on the cuprizone model of demyelination are also investigated. A study on 80 129/Sv mice, found that melatonin enhances the effect of umbilical cord-derived multipotent mesenchymal stromal cells (UC-MMSCs) in cuprizone-treated mice. This study also found that after melatonin injections, the number of crossed squares and boluses in experimental mice increases to the values of intact animals (Labunets, Utko, Toporova, et al., 2021a). In a related work on aged rats, melatonin injections improved the impact of human UC-MMSCs on the behavioral responses of elderly mice fed a cuprizone diet and also cause favorable changes in the parameters of motor and emotional activity (I. Labunets et al. 2021a, b, c). The same researchers found that the manifestations of the beneficial effect of transplanted UC-MMSCs in combination with melatonin on the behavioral outcomes of mice with a cuprizone model of demyelination in both adults (6–7-month-old) and aged (15–17-month-old) mice (Labunets, Utko, Toporova, 2021b).

Melatonin treatment (10 mg/kg i.p.) slowed the onset and progression of $MOG_{35–55}$ EAE, according to a study that examined the neuroprotective effects of melatonin on 32 female C57BL/6 mice in Long et al.'s study (Long et al. 2018). Álvarez-Sánchez et al., in an evaluation of immunomodulatory activities of melatonin, demonstrated that, without changing the beginning of the disease, melatonin decreased both the severity and incidence of EAE in 8-week-old female C57BL/6 mice (Álvarez-Sánchez et al. 2015). Based on an evaluation of 28 male C57BL/6 mice in a study by Vakilzade et al., melatonin (at two doses of 50 and 100 mg/kg for 7 days) improved motor performance and increased cell survival in a cuprizone toxic model of demyelination (Vakilzadeh et al. 2016). In a study comparing the therapeutic effects of melatonin (20 mg/kg i.p.) and N-acetylserotonin, Wen et al. found that melatonin is equally efficient as N-acetylserotonin at reducing clinical signs in the EAE model of MS. Melatonin supplementation was associated with reduced clinical scores when administered before or after symptom onset (Wen et al. 2016). As a result of a study of melatonin at various doses on EAE mice, it was determined that melatonin, at all doses tested, can alleviate the severity of EAE (Jand et al. 2022). The last study on 17 Sprague–Dawley rats of both sexes found that melatonin significantly decreased disease severity and duration (Kang et al. 2001).

After 9 weeks of melatonin therapy (80 mg/kg/day), researchers evaluated the grip strength test and the open field test and concluded that melatonin reduced weight gain and enhanced motor activity in male and female SWR/J mice during the demyelination stage. In the re-myelination stage, protective effects only observed in male mice suggest an interaction between exogenous melatonin therapy and female sex hormones (Abo Taleb & Alghamdi 2020). This study was the only one which shed light on the sex-specific effects of melatonin therapy in animal models of toxic demyelination. In another study, Alghamdi and Abo Taleb injected 80 mg/kg of melatonin daily for 5 weeks in 30 Male SWR/J mice to examine the hormone's impact on memory. They found that melatonin increased locomotor activity, prolonged nociception latency, and improved novel object recognition in the MS mouse model (Alghamdi & AboTaleb 2020). An experiment involving 50 mice administered melatonin (10 mg/kg) for three weeks demonstrate that melatonin improves locomotion, psychological condition, and cognition impairment (Gharib et al. 2022).

Meta-analysis

To perform a meta-analysis, we included studies that compared the clinical severity scale in melatonin-treated groups with the EAE group treated with the ineffective substance. In eight studies (Chen et al. 2016; Ghareghani et al. 2017, 2022, 2019; Ghareghani et al. 2018a, b, c; Jand et al. 2022; Kang et al. 2001; Ortiz et al. 2022), a significant difference was reported between the melatonin and placebo groups, but in the other two studies, it was not statistically significant (Álvarez-Sánchez et al. 2015; Wen et al. 2016). In the meta-analysis, the difference between the melatonin and placebo groups was significant (SMD: − 2.52; 95%CI: − 3.61 to − 1.42; p value < 0.01). In subgroup analyses, the difference between melatonin and EAE was only meaningful in the mouse subgroup (SMD: − 2.60; 95%CI: − 3.74 to − 1.46; p value < 0.01). Figure 2 presents the details and the forest plot of the meta-analyses.

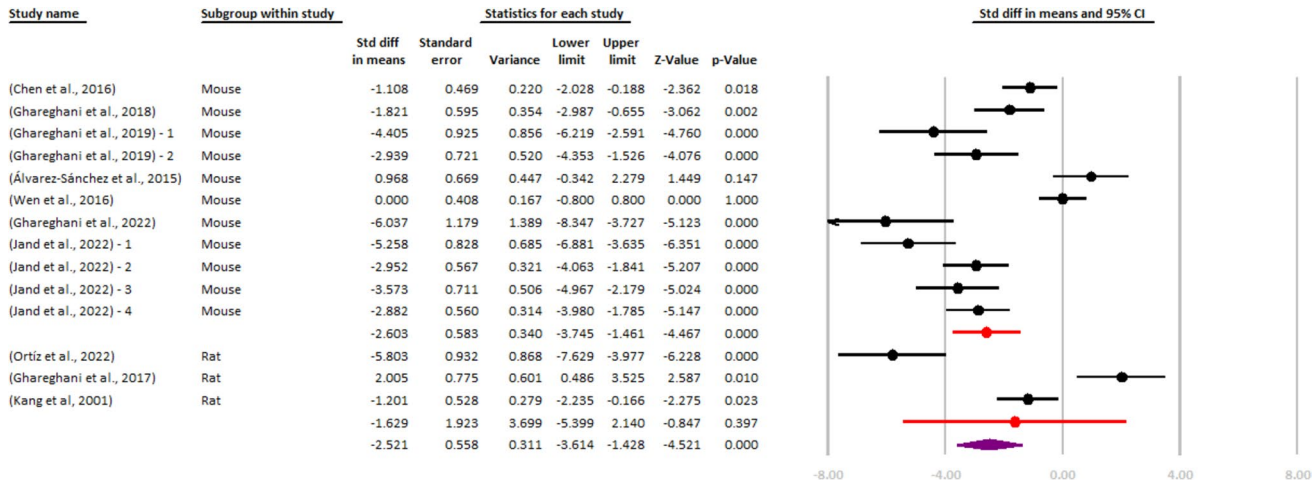

Fig. 2 The forest plot and details of meta-analysis of the effects of melatonin supplementation on the clinical severity scale in EAE

Discussion

This systematic review of preclinical studies includes 21 animal studies, which examines the effects of melatonin administration on behavioral and cognitive outcomes in rodent models of MS. Based on the findings, treatment with melatonin may be associated with improved behavioral and cognitive outcomes of rodents with acceptable safety profile. Results of our quantitative synthesis found statistically significant effects for the use of melatonin in animal models of MS in improving the clinical severity scale, which was evident only in studies involving mice in the sensitivity analysis.

The discovery of unique gene variations linked to aberrant melatonin secretion as well as the well-established relationship between melatonin and the immune system support the idea that dysregulation of the melatonin pathway may play a role in the etiology of MS (Skarlis & Anagnostouli 2020). Melatonin levels in MS patients are often unaltered, however, lower levels are related to the severity and clinical features of the disease, so low melatonin levels have been linked to severe exacerbations of MS (Kern et al. 2019).

Melatonin due to its antioxidant function, can attenuate oxidative stress by reducing the main oxidative stress markers, activation of the Nrf2/antioxidant responsive element signaling pathway, and protecting against mitochondrial dysfunction. Moreover, melatonin could induce Nrf2 to stop apoptosis and restore the cellular redox potential. Melatonin also regulates the response of regulatory and effector T cells and reduces the amount of pro-inflammatory cytokines due to its immunomodulatory and anti-inflammatory properties, providing a more protective cytokine microenvironment (Muñoz-Jurado et al. 2022). Hawkes et al. found that melatonin can decrease neutrophil function, boost NK cell and monocyte levels, control CD4 +, and CD8 + cells differentiation, and regulate B-cell activation while investigating the impact of melatonin on the immune system in MS. Melatonin can also influence the NO/NOS pathway, enhance mitochondrial activity, and prevent NF-κB from moving into the nucleus, all of which have an impact on how the immune system functions (Hawkes et al. 2021).

Most of the studies suggested the positive effects of melatonin on behavioral and cognitive outcomes in rodent models of MS. In addition, considering the antioxidant effects of melatonin, as well as the oxidative stress basis of MS (Naseri et al. 2022; Tobore 2021), future clinical trials are suggested to assess the effects of exogenous melatonin supplementation in patients with different phenotypes of MS, with different levels of disability.

While the prevalence of MS is significantly higher at latitudes above 40 degrees North and South, populations in these areas receive little sunlight, which may cause a longer increase in melatonin synthesis and release. These findings are made by Ghareghani et al. in their study on the relationship between latitude, vitamin D, melatonin, and gut microbiota. They contend that melatonin therapy would not be an effective strategy for treating MS patients with low melatonin levels or vitamin D deficiency because the functions and synthesis of these two hormones are at odds with one another, and they recommend that a balance should exist between these two hormones (Ghareghani et al. 2018a, b, c). According to a recent study by Ghareghani et al., taking vitamin D or melatonin may help people with MS, though doing so over the long term can upset their balance, which is crucial for the immune cells' fine-tuning of performance (Ghareghani et al. 2023a, b).

The positive effects of melatonin on cognitive outcomes of animal models of MS are reported in two studies (Alghamdi and AboTaleb 2020; Jallouli et al. 2022). Considering the

correlation between inflammation, oxidative stress, and cognitive outcomes of MS (Talebi et al. 2021), as well as the beneficial effects of melatonin supplementation in improving cognitive outcomes previously reported in other neurological conditions such as Alzheimer's Disease (Majidazar et al. 2022). In addition, Morsali et al. implied that literature-derived evidence is in favor of melatonin's benefits in clinical studies (Morsali et al. 2023), which shed light on the importance of future well-designed clinical studies on this topic.

The function of melatonin in cognitive and behavioral outcomes in mouse models of MS was examined in this study as the first systematic review and meta-analysis of preclinical studies. The main advantages of this study were its systematic approach and thorough database scanning across four major databases. Due to differences between the groups, and assessment scales, there were insufficient studies that considered other outcomes other than the clinical severity scale, which prevented us from a comprehensive meta-analysis.

Conclusion

Melatonin is beneficial and efficient in rodent models of MS and has an appropriate safety profile. Melatonin as a supplement may be associated with improved outcomes in MS; however, there is a need for future well-designed clinical studies on this topic to achieve an evidence-based conclusion and clinical recommendations.

Acknowledgements The research protocol was approved and supported by the Student Research Committee, Tabriz University of Medical Sciences (grant number: 71005). Also, we would like to appreciate of the cooperation of Clinical Research Development Unit, Imam Reza General Hospital, Tabriz, Iran in conducting of this research.

Author contributions HR, EN, PV, SM, AM AE, RMH, and AN: systematic search; study selection; data extraction; risk of bias assessment; preparing the figures and writing the first draft of the manuscript; SS: conceptualization; supervision; critically editing the manuscript. All authors approved the final version for submission.

Funding The research was supported by the Student Research Committee, Tabriz University of Medical Sciences (grant number: 71005).

Data availability All data generated or analyzed during this study are included in this published article and its supplementary information files.

Declarations

Conflict of interest None.

Ethical approval Ethics code: IR.TBZMED.VCR.REC.1401.377.

Consent to participate Not applicable.

Consent for publication Not applicable.

References

Abo Taleb HA, Alghamdi BS (2020) Neuroprotective Effects of Melatonin during Demyelination and Remyelination Stages in a Mouse Model of Multiple Sclerosis [Article]. J Mol Neurosci 70(3):386–402. https://doi.org/10.1007/s12031-019-01425-6

Alghamdi BS, AboTaleb HA (2020) Melatonin improves memory defects in a mouse model of multiple sclerosis by up-regulating cAMP-response element-binding protein and synapse-associated proteins in the prefrontal cortex. J Integr Neurosci 19(2):229–237

Álvarez-Sánchez N, Cruz-Chamorro I, López-González A, Utrilla JC, Fernández-Santos JM, Martínez-López A, Lardone PJ, Guerrero JM, Carrillo-Vico A (2015) Melatonin controls experimental autoimmune encephalomyelitis by altering the T effector/regulatory balance [Article]. Brain, Behavior Immunity 50:101–114. https://doi.org/10.1016/j.bbi.2015.06.021

Burrows DJ, McGown A, Jain SA, De Felice M, Ramesh TM, Sharrack B, Majid A (2019) Animal models of multiple sclerosis: From rodents to zebrafish. Mult Scler 25(3):306–324. https://doi.org/10.1177/1352458518805246

Chen SJ, Huang SH, Chen JW, Wang KC, Yang YR, Liu PF, Lin GJ, Sytwu HK (2016) Melatonin enhances interleukin-10 expression and suppresses chemotaxis to inhibit inflammation in situ and reduce the severity of experimental autoimmune encephalomyelitis [Article]. Int Immunopharmacol 31:169–177. https://doi.org/10.1016/j.intimp.2015.12.020

Dokoohaki S, Ghareghani M, Ghanbari A, Farhadi N, Zibara K, Sadeghi H (2017) Corticosteroid therapy exacerbates the reduction of melatonin in multiple sclerosis. Steroids 128:32–36. https://doi.org/10.1016/j.steroids.2017.10.006

Escribano BM, Muñoz-Jurado A, Caballero-Villarraso J, Valdelvira ME, Giraldo AI, Paz-Rojas E, Gascón F, Santamaría A, Agüera E, Túnez I (2022) Protective effects of melatonin on changes occurring in the experimental autoimmune encephalomyelitis model of multiple sclerosis [Article]. Mult Scler Relat Disord. https://doi.org/10.1016/j.msard.2022.103520

Ghareghani M, Dokoohaki S, Ghanbari A, Farhadi N, Zibara K, Khodadoust S, Parishani M, Ghavamizadeh M, Sadeghi H (2017) Melatonin exacerbates acute experimental autoimmune encephalomyelitis by enhancing the serum levels of lactate: A potential biomarker of multiple sclerosis progression [Article]. Clin Exp Pharmacol Physiol 44(1):52–61. https://doi.org/10.1111/1440-1681.12678

Ghareghani M, Reiter RJ, Zibara K, Farhadi N (2018a) Latitude, Vitamin D, Melatonin, and Gut Microbiota Act in Concert to Initiate Multiple Sclerosis: A New Mechanistic Pathway. Front Immunol 9:2484. https://doi.org/10.3389/fimmu.2018.02484

Ghareghani M, Scavo L, Arnoult D, Zibara K, Farhadi N (2018b) Melatonin therapy reduces the risk of osteoporosis and normalizes bone formation in multiple sclerosis. Fundam Clin Pharmacol 32(2):181–187. https://doi.org/10.1111/fcp.12337

Ghareghani M, Zibara K, Sadeghi H, Farhadi N (2018c) Spasticity Treatment Ameliorates the Efficacy of Melatonin Therapy in Experimental Autoimmune Encephalomyelitis (EAE) Mouse Model of Multiple Sclerosis [Article]. Cell Mol Neurobiol 38(5):1145–1151. https://doi.org/10.1007/s10571-018-0580-y

Ghareghani M, Scavo L, Jand Y, Farhadi N, Sadeghi H, Ghanbari A, Mondello S, Arnoult D, Gharaghani S, Zibara K (2019) Melatonin therapy modulates cerebral metabolism and enhances remyelination by increasing PDK4 in a mouse model of multiple sclerosis. Front Pharmacol 10:147

Ghareghani M, Farhadi Z, Rivest S, Zibara K (2022) PDK4 Inhibition Ameliorates Melatonin Therapy by Modulating Cerebral Metabolism and Remyelination in an EAE Demyelinating Mouse Model

of Multiple Sclerosis. Front Immunol 13:862316. https://doi.org/10.3389/fimmu.2022.862316

Ghareghani M, Pons V, Laflamme N, Zibara K, Rivest S (2023a) Author Correction: Inhibiting nighttime melatonin and boosting cortisol increase patrolling monocytes, phagocytosis, and myelination in a murine model of multiple sclerosis. Exp Mol Med 55(2):485–486. https://doi.org/10.1038/s12276-023-00948-8

Ghareghani M, Zibara K, Rivest S (2023b) Melatonin and vitamin D, two sides of the same coin, better to land on its edge to improve multiple sclerosis. Proc Natl Acad Sci U S A 120(14):e2219334120. https://doi.org/10.1073/pnas.2219334120

Gharib D, Rashed L, Yousuf A, El-Shawwa M, Saber S, Ibrahim B, Kamel M (2022) Therapeutic Effect of Microvesicles Derived From BM-MSCS Transplantation And/Or Melatonin In Cuprizone Model of Multiple Sclerosis: A Pharmacodynamic Biochemical Assay. Egypt J Chem 65(10):153–169. https://doi.org/10.21608/ejchem.2022.112611.5107

Gold R, Linington C, Lassmann H (2006) Understanding pathogenesis and therapy of multiple sclerosis via animal models: 70 years of merits and culprits in experimental autoimmune encephalomyelitis research. Brain 129(Pt 8):1953–1971. https://doi.org/10.1093/brain/awl075

Hawkes CH, Baker MD, Pohl D, Lechner-Scott J, Levy M, Giovannoni G (2021) Melatonin and multiple sclerosis. Mult Scler Relat Disord 51:103032. https://doi.org/10.1016/j.msard.2021.103032

Jallouli S, Ghroubi S, Dhia IB, Yahia A, Elleuch MH, Sakka S, Mhiri C, Hammouda O (2022) Effect of melatonin intake on postural balance, functional mobility and fall risk in persons with multiple sclerosis: a pilot study. Int J Neurosci. https://doi.org/10.1080/00207454.2022.2090353

Jand Y, Ghahremani MH, Ghanbari A, Ejtemaei-Mehr S, Guillemin GJ, Ghazi-Khansari M (2022) Melatonin ameliorates disease severity in a mouse model of multiple sclerosis by modulating the kynurenine pathway. Sci Rep 12(1):15963. https://doi.org/10.1038/s41598-022-20164-0

Kang JC, Ahn M, Kim YS, Moon C, Lee Y, Wie MB, Lee YJ, Shin T (2001) Melatonin ameliorates autoimmune encephalomyelitis through suppression of intercellular adhesion molecule-1. J Vet Sci 2(2):85–89

Kashani IR, Rajabi Z, Akbari M, Hassanzadeh G, Mohseni A, Eramsadati MK, Rafiee K, Beyer C, Kipp M, Zendedel A (2014) Protective effects of melatonin against mitochondrial injury in a mouse model of multiple sclerosis. Exp Brain Res 232(9):2835–2846. https://doi.org/10.1007/s00221-014-3946-5. ([**RETRACTEDARTICLE**])

Kern S, Geiger M, Paucke M, Kästner A, Akgün K, Ziemssen T (2019) Clinical relevance of circadian melatonin release in relapsing-remitting multiple sclerosis. J Mol Med (Berl) 97(11):1547–1555. https://doi.org/10.1007/s00109-019-01821-w

Labunets I, Utko N, Toporova O, Pokholenko I, Panteleymonova T, Litoshenko Z, Butenko G (2021a) The effects of combined administration of human umbilical cord-derived multipotent mesenchymal stromal cells and melatonin or fibroblast growth factor-2 to aged mice with a toxic cuprizone model of demyelination [Article]. Cell Organ Transplant 9(1):4–11. https://doi.org/10.22494/cot.v9i1.116

Labunets IF, Utko NA, Toporova OK (2021b) Effects of Multipotent Mesenchymal Stromal Cells of the Human Umbilical Cord and Their Combination with Melatonin in Adult and Aging Mice with a Toxic Cuprizone Model of Demyelination [Article]. Adv Gerontol 11(2):173–180. https://doi.org/10.1134/s2079057021020077

Labunets IF, Utko NA, Toporova OK, Savosko SI, Pokholenko IO, Panteleymonova TN, Butenko GM (2021c) Melatonin and fibroblast growth factor-2 potentiate the effects of human umbilical cord multipotent mesenchymal stromal cells in mice with cuprizone-induced demyelination [Article]. Biopolym Cell 37(5):369–378. https://doi.org/10.7124/bc.000A62

Long T, Yang Y, Peng L, Li Z (2018) Neuroprotective Effects of Melatonin on Experimental Allergic Encephalomyelitis Mice Via Anti-Oxidative Stress Activity. J Mol Neurosci 64(2):233–241. https://doi.org/10.1007/s12031-017-1022-x

Mahmoodi M, Sedghy F, Jafarzadeh A (2020) Effect of Treatment with Thyme Extract on Urinary Levels of Melatonin in an Experimental Autoimmune Encephalomyelitis Mouse Model. Rep Biochem Mol Biol 8(4):407–412

Majidazar R, Rezazadeh-Gavgani E, Sadigh-Eteghad S, Naseri A (2022) Pharmacotherapy of Alzheimer's disease: an overview of systematic reviews. Eur J Clin Pharmacol 78(10):1567–1587. https://doi.org/10.1007/s00228-022-03363-6

Morsali S, Sabahi Z, Kakaei J, Hakimzadeh Z, Hamidi S, Gholipour-Khalili E, Sanaie S, Talebi M, Naseri A (2023) Clinical efficacy and safety of melatonin supplementation in multiple sclerosis: a systematic review. Inflammopharmacology 31(5):2213–2220. https://doi.org/10.1007/s10787-023-01271-4

Muñoz-Jurado A, Escribano BM, Caballero-Villarraso J, Galván A, Agüera E, Santamaría A, Túnez I (2022) Melatonin and multiple sclerosis: antioxidant, anti-inflammatory and immunomodulator mechanism of action. Inflammopharmacology 30(5):1569–1596. https://doi.org/10.1007/s10787-022-01011-0

Naseri A, Forghani N, Sadigh-Eteghad S, Shanehbandi D, Asadi M, Nasiri E, Talebi M (2022) Circulatory antioxidant and oxidative stress markers are in correlation with demographics but not cognitive functions in multiple sclerosis patients. Mult Scler Relat Disord 57:103432. https://doi.org/10.1016/j.msard.2021.103432

Ortiz GG, Briones-Torres AL, Benitez-King G, Gonzalez-Ortiz LJ, Palacios-Magana CV, Pacheco-Moises FP (2022) Beneficial Effect of Melatonin Alone or in Combination with Glatiramer Acetate and Interferon beta-1b on Experimental Autoimmune Encephalomyelitis. Molecules. https://doi.org/10.3390/molecules27134217

Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffmann TC, Mulrow CD, Shamseer L, Tetzlaff JM, Akl EA, Brennan SE, Chou R, Glanville J, Grimshaw JM, Hróbjartsson A, Lalu MM, Li T, Loder EW, Mayo-Wilson E, McDonald S, Moher D (2021) The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. BMJ 372:n71. https://doi.org/10.1136/bmj.n71

Preclinical Systematic Reviews & Meta-Analysis Wiki (2023) CAMARADES Berlin, QUEST-BIH Charité. https://www.CAMARADES.de

Potes Y, Cachán-Vega C, Antuña E, García-González C, Menéndez-Coto N, Boga JA, Gutiérrez-Rodríguez J, Bermúdez M, Sierra V, Vega-Naredo I, Coto-Montes A, Caballero B (2023) Benefits of the Neurogenic Potential of Melatonin for Treating Neurological and Neuropsychiatric Disorders. Int J Mol Sci. https://doi.org/10.3390/ijms24054803

Ramos Gonzalez EJ, Ramirez Jirano LJ, Garcia Martinez DZ, Ortiz GG, Jave Suarez LF, Leal Cortes CA, Bitzer Quintero OK (2021) A comparative study of melatonin and immunomodulatory therapy with interferon beta and glatiramer acetate in a mouse model of multiple sclerosis. Neurologia (Engl Ed) 36(4):262–270. https://doi.org/10.1016/j.nrl.2018.01.007

Sharif R, Aghsami M, Gharghabi M, Sanati M, Khorshidahmad T, Vakilzadeh G, Mehdizadeh H, Gholizadeh S, Taghizadeh G, Sharifzadeh M (2017) Melatonin reverses H-89 induced spatial memory deficit: Involvement of oxidative stress and mitochondrial function. Behav Brain Res 316:115–124. https://doi.org/10.1016/j.bbr.2016.08.040

Skarlis C, Anagnostouli M (2020) The role of melatonin in Multiple Sclerosis. Neurol Sci 41(4):769–781. https://doi.org/10.1007/s10072-019-04137-2

Talebi M, Majdi A, Nasiri E, Naseri A, Sadigh-Eteghad S (2021) The correlation between circulating inflammatory, oxidative stress, and

neurotrophic factors level with the cognitive outcomes in multiple sclerosis patients. Neurol Sci 42(6):2291–2300. https://doi.org/10.1007/s10072-020-04807-6

Tobore TO (2021) Oxidative/Nitroxidative Stress and Multiple Sclerosis. J Mol Neurosci 71(3):506–514. https://doi.org/10.1007/s12031-020-01672-y

Torkildsen O, Brunborg LA, Myhr KM, Bø L (2008) The cuprizone model for demyelination. Acta Neurol Scand Suppl 188:72–76. https://doi.org/10.1111/j.1600-0404.2008.01036.x

Vakilzadeh G, Khodagholi F, Ghadiri T, Ghaemi A, Noorbakhsh F, Sharifzadeh M, Gorji A (2016) The Effect of Melatonin on Behavioral, Molecular, and Histopathological Changes in Cuprizone Model of Demyelination [Article]. Mol Neurobiol 53(7):4675–4684. https://doi.org/10.1007/s12035-015-9404-y

Wen J, Ariyannur PS, Ribeiro R, Tanaka M, Moffett JR, Kirmani BF, Namboodiri AM, Zhang Y (2016) Efficacy of N-Acetylserotonin and Melatonin in the EAE Model of Multiple Sclerosis. J Neuroimmune Pharmacol 11(4):763–773. https://doi.org/10.1007/s11481-016-9702-9

Zhan J, Mann T, Joost S, Behrangi N, Frank M, Kipp M (2020) The Cuprizone Model: Dos and Do Nots. Cells. https://doi.org/10.3390/cells9040843

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