Review Article

Efficacy of electrotherapy in Bell's palsy treatment: A systematic review

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Abstract.

BACKGROUND: Up to now there is not enough evidence that supports the use of electrotherapy in the treatment of Bell's palsy. **OBJECTIVE:** Through a systematic review, we aimed to verify whether the use of electrotherapy is effective for treating Bell's palsy or peripheral paralysis.

METHODS: Publications were searched in PubMed, EBSCO and Web of Science. The present systematic review included studies that analyzed the electrotherapy as a therapeutic method for treating individuals with Bell's palsy, in order to recover the function of facial muscles.

RESULTS: Seven studies involving a total of 131 cases and 113 controls were included in this systematic review. In the studies analyzed, patients received electrotherapy combined with other treatments such as hot-wet facial napkins, massages and muscle reeducation. Although the effect of electrotherapy alone was not evaluated, the use of electrotherapy combined with other treatments produced a significant improvement in the individuals evaluated.

CONCLUSIONS: Due to the diverse methodologies used and the small number of individuals included in the studies, we could not fully prove the efficacy of electrotherapy for treating Bell's Palsy. Future studies with larger samples and homogenous populations should be performed to obtain conclusive results.

Keywords: Bell's palsy, peripheral paralysis, electrotherapy

1. Introduction

Facial peripheral paralysis, or Bell's palsy, is an acute mononeuropathy of the facial nerve. It is of un-

known cause and can affect a single nerve; it starts with pain in the mastoids region and partial or total paralysis of one side of the face [1,2]. Bell's palsy affects equally males and females, with an incidence of 11.5 to 40.2/100 000 [3,4]. The incidence is higher in individuals with Diabetes Mellitus, immunocompromised patients, individuals with arterial hypertension, patients who have had a viral infection of the upper respiratory tract, and pregnant woman [5]. Although Bell's palsy can happen at any age, there are peaks of incidence

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between 15–45 years of age, mean age 40 years [6]. Bell's palsy is usually idiopathic (75% of cases), and less frequently a secondary paralysis (25%) [2]. Several etiologic mechanisms have been proposed to explain the development of Bell's palsy including herpes virus, infectious mechanisms, ischemic mechanisms and autoimmune mechanisms; nevertheless, these are not completely understood [7–9].

The treatment of Bell's palsy is divided into acute and maintenance treatments. The acute treatment consists of using corticosteroids and antivirals that must be initiated within the first 72 hours after the onset of clinical signs [8]. The use of corticosteroids represents a highly-recommended intervention with evidence of improvement [3,10]. Regarding the use of antivirals, it is recommended that antiviral drugs are not prescribed in isolation [3,11]. The maintenance treatment includes interventions such as eye care, mouth care, physical therapy, Botulinum toxin injections and even complementary medicine treatments such as acupuncture [8].

Another type of treatment is physical therapy. However, according to the general-international Guidelines and some systematic reviews, physical therapy is not a highly recommended treatment due to the scarce evidence of improvement observed in individuals with Bell's palsy who received any type of physical therapy [3,12-14]. Nonetheless, in a recent systematic review, it was reported that the combination of pharmacological treatment with some modalities of physical therapy, favored a better recovery than pharmacological treatment alone [15]. Therefore, our systematic review will determine if the use of electrotherapy is recommendable when treating individuals with Bell's palsy. The objective of this systematic review is to demonstrate the benefits and efficacy of electrotherapy for treating patients with facial paralysis (Bell's palsy), in comparison to patients who did not receive electric stimulation.

2. Methods

This systematic review followed the Preferred Reporting Items for Systematic Review and Meta-analysis (PRISMA) criteria. The protocol of this systematic review was registered in PROSPERO (https://www.crd. york.ac.uk/PROSPERO/index.php), registration number 42014014938.

2.1. Inclusion and exclusion criteria

2.1.1. Inclusion criteria

The studies had to be published in peer-reviewed journals; had to be written in English; had to be cross-sectional or case-control designs. The studies had to provide information regarding time of exposure and the effects of the electrotherapy when treating patients with Bell's facial paralysis. Finally, the electrical stimulation had to be performed with electrodes.

2.1.2. Exclusion criteria

Controlled clinical trials that did not include electrotherapy as part of the Bell's facial paralysis treatment. Protocol articles for future studies were also excluded [16].

2.2. Search and selection of articles

We performed a search in PUBMED and Web of Science databases. We used the terms "Bell's palsy", "electrical stimulation", "rehabilitation" and combinations such as "Bell's palsy and electrical stimulation" and "Rehabilitation and facial paralysis". The search was concluded in January 2017. Initially, the electronic search generated 1,148 potentiality relevant papers; of those, we excluded 517 because they were duplicates. Then, we excluded 580 more, as they were of no relevance for this systematic review, or were written in other language than English, or were systematic reviews. Additionally, 27 papers were excluded after reading the abstract. Finally, we considered 24 papers; however, after reading and analyzing the contents, only 7 papers were included in this systematic review (Fig. 1).

2.3. Data extraction

Data was extracted by two independent investigators (EGBP and MSM). Each reviewer extracted the following data: author, year of publication, location, number of controls, number of cases, diagnoses and evolution of the palsy, characteristics of the treatment in the clinical trials and also additional treatments.

2.4. Data analysis

The selected studies were assessed using the GRADE system (Grading of Recommendations Assessment, Development and Evaluation Scale) (http://www.gradeworkinggroup.org) for quality evaluation (Table 1).

Reference	Design	Number of patients	Quality of the evidence (GRADE)	Publication bias
[23] Gittins J (1998)	Cross-sectional	10	$\oplus \oplus \oplus \ominus$ Moderate	Undetected
[17] Targan R (2000)	Case-control	12	$\oplus \oplus \oplus \oplus High$	Undetected
[18] Manikadan N (2006)	Case-control	29	$\oplus \oplus \oplus \oplus High$	Undetected
[22] Hyvärinen A (2008)	Cross-sectional	10	$\oplus \oplus \oplus \ominus$ Moderate	Undetected
[19] Alakram P (2010)	Case-control	8	$\oplus \oplus \oplus \ominus$ Moderate	Undetected
[20] Tuncay F (2015)	Case-control	32	$\oplus \oplus \oplus \oplus High$	Undetected
[21] Kim J (2016)	Case-control	30	$\oplus \oplus \oplus \oplus High$	Undetected

Table 1
Quality assessment of the studies using the GRADE system

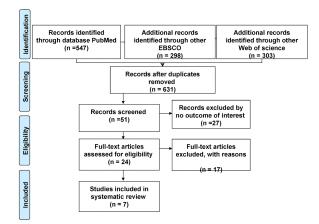


Fig. 1. Flowchart showing the search strategy used in the systematic review.

3. Results

This systematic review analyzed seven studies related to facial paralysis or Bell's palsy and rehabilitative treatment. The descriptive characteristics of each study are shown in Table 2.

3.1. Brief description of each study included

The study by Targan et al. in the USA [17] included twelve individuals with idiopathic facial paralysis and five individuals with a history of surgically affected nerves were recruited [17]. Diagnoses were based on latency and the House-Brackmann scale. They obtained the correlation coefficients between clinical residuals and nerve conduction latency (r=0.44). The House-Brackmann scale and nerve conduction latency were low (r=0.51), but statistically significant (p=0.036 and p=0.02, respectively).

Another study performed in India included 59 individuals diagnosed with Bell's palsy [18]. Patients were divided into a control group (n=30) and a group of neuromuscular facial re-education (n=29). The control group was treated following a standard protocol, while the facial neuromuscular re-education group

received a treatment that included 3 sets of 5 to 10 repetitions of electrical stimulation. The facial scale ratings in the control group were 32 (9.7–54) in pretreatment, and 54.5 (42.2–71.7) in post-treatment, $p \le 0.01$. While the group of neuromuscular reeducation scored 33 (18–43.5) in pretreatment, and 66 (54–76.7) in post-treatment, $p \le 0.01$.

In a recent study performed in the region of South Africa [19], 16 individuals were recruited and divided into a re-education group (n=8) and an experimental group (n=8) within the early phase of Bell's palsy. Both groups had the same treatment; however, the experimental group additionally received 30 minutes of electrical stimulation of facial muscles using a TENS unit (Frequency 10 Hz, pulse width 10 μ sec). According to the House-Brackmann scale, the recovery rates in the experimental group were higher than in the control group $(37.6 \pm 18.1 \text{ versus } 29.6 \pm 12.5, \text{ respectively})$; nevertheless, these differences were not statistically significant (p=0.36).

In a different study [20], individuals with Bell's palsy were divided into a group that received electro stimulation (n=32) and another group that did not (n=28). Significant differences were observed between the groups when the FDI scale was evaluated. The electro stimulation-group showed a major improvement of physical function (p=0.02) and social welfare function (p=0.03).

Finally, in a study performed by Kim et al. in Korea [21], 60 individuals with Bell's palsy in early phase were selected and separated into two groups, control (n=30) and experimental (n=30). The treat ment in both groups consisted of drugs, with additional electrical stimulation for the experimental group. Prednisolone was administrated during the first 5 days (1 mg/kg/day), and then during 10 days. They also received acyclovir (1500 mg/day) during five days. The overall rate of patient recovery in the experimental group was 96%, while in the control group was 88%.

Table 2
Descriptive and clinical characteristics of the studies

Study	Location	Controls Cases	Cases	Diagnosis	Characteristic	Characteristics of the treatment	Additional treatment	Result
(Special Control of the Control of t			-		The state of the s			ATT COLU
Gittins J	United	I	10^{1}	Secondary		12 to 24 months	None	Voluntary closing of the eyelid
(1998) [23]	Kingdom			palsy of the 7th	Current type and wave	Constant voltage currents com-		increased an average of 2.5 mm
				cranial nerve ²	form	pensated monophasic current		and improved the speed 0.9 sec-
					Frequency	2 Hz-200 Hz		onds
					Intensity	Not specified		Lagophthalmus: 2.9 mm
					Pulse width	$50 \mu sec - 200 \mu sec$		decrease (mean) in 8 patients
					Duration	3 months		Eyelid displacement ($P <$
					Application time	1 hour daily		0.005)
					Combination with drugs	None		
					during treatment			
Targan R	EE. UU	17	12^{3}	Chronic facial	Evolution of the paralysis	3.7 (Bell's palsy group) and	None	Decreased facial motor nerve la-
(2000) [17]				nerve damage		7.2 years for acoustic neuroma		tency 1.13 ms ($P = 0.001$)
				caused by		group)		Improvement on the House-
				Bell's palsy or	Electrical stimulation	Monophasic current		Brackmann Scale ($P = 0.003$)
				acoustic	Intensity	Sub-motor level (sensory		Decrease in Clinical
				neuroma		threshold)		Impairment score 28.7 ($P =$
				excision	Pulse width	86 μ sec (1 pulse every 700 ms)		0.0005)
					Ramp up	1 second		
					Ramp down	0.5 second		
					Duration	6 months		
					Application time	Starting with 30 minutes for		
						muscle, progressively increas-		
						ing to 6 hours in the 6 th month		
					Combination with drugs	None		
					during treatment			
Manikadan N India	India	30	29	Facial paralysis	Evolution of the paralysis	Acute ⁴	Control group: Gross facial	Significant improvement in the
(2006) [18]				idiopathic	Current type and wave	Galvanic to muscle and faradic	exercises, massage, orthotic	total score of the Facial Grading
					form	current to motor trunk	devices or taping to lift droop-	Score and in the movement sub-
					Frequency and pulse width	Not specified	ing flaccid faces plus electri-	component in favor of the con-
					Intensity	Motor level	cal stimulation	trol group was observed (P <
					Duration	2 weeks (6 days for week)	Re-education group:	0.01)
					Application time	10 visible contractions for mus-	Techniques tailored to each	There were no significant
						cle, 3 times daily	patient	differences in synkinesis
					Combination with drugs	None		subcomponents
					during treatment			

Table 2, continued

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Study	Location	Controls Cases	Cases	Diagnosis	Characteristic	Characteristics of the treatment	Additional treatment	Kesuit
Hyvärinen A	Finland	ı	10	Chronic facial	Evolution of the paralysis	Chronic 1–24 years	None	Improvement in motor latency
(2008) [22]				nerve palsy of	Wave form	Monophasic current		of the facial nerve, significantly
				idiopathic	Frequency	20 Hz		only in the upper nerve branch
				origin or by	Intensity	Sub-motor level (sensory		(P = 0.02)
				Herpes Zoster		threshold)		Improvement in evaluation with
					Pulse width	$100 \mu sec$		the House-Brackmann scale of
					Duration	6 months		at least 1 degree in all patients
					Application time	Starting with 30 minutes for		
						muscle, progressively increas-		
						ing to 6 hours in the 6th month		
					Combination with drugs	No. The use of corticosteroids		
					during treatment	and acyclovir were previous to		
						the treatment with electrical		
						stimulation		
Alakram P	South	∞	∞	Facial paralysis	Evolution of the paralysis	Acute (less than 30 days post	Early re-education	The comparison of recovery
(2010) [19]	Africa			idiopathic		onset)	Control group: Heath,	rates for the House-Brackmann
					Wave form	Not specified	massage and exercises. pred-	scale indicates that the individ-
					Frequency	10 Hz	nisolone (2 mg per kg daily	ual rates in the experimental
					Intensity	Motor level	and weaned off within	group were higher than that of
					Pulse width	$10 \mu \text{sec}$	2 weeks	the control group
					Combination with drugs	All individuals received pred-	Experimental orono: Same as	However there was no
					during treatment	nisolone (2 mg ner kg daily and	control aroun plus electros-	statistically significant
					anima a cannon	weaned off within	timulation	difference in rate of recovery
						wealled on within	CIIII atlation	unicicine in rate of recovery
						2 weeks)	For patients with eye	between the experimental and
							problems were given eye	control group $(P = 0.36)$
							drops and those with pain	
1	,	,	;	,		•	were given Panado's	
Tuncay F	Turkey	28	32	Facial paralysis	Evolution of the paralysis	Acute (four weeks)	Control group: Hot pack,	Significant improvement in the
(2015) [20]				idiopathic	Wave form	Monophasic current (only for	massage, facial expression	House-Brackmann scale in both
						group with electrical stimula-	and exercise through a mirror	
						tion)	Experimental group: Same	the experimental group ($P =$
					Intensity	Motor level	treatment plus facial electros-	0.0001)
					Frequency	2.5 Hz	timulation	Significant improvement in the
					Pulse width interpulse in-	$100 \mu sec$	All patients received	amplitude and latency of the fa-
					terval	300 µsec	corticosteroid treatment for	cial nerve motor in the experi-
					Duration	3 weeks (5 day for week)	10 days	mental group
					Application time	3 sets of 30 contractions per		Improvement in the Facial
						treated muscle		Disability Index in both
					Combination with drugs	No. The use of corticosteroids		treatment groups, in favor of the
						was previous to the treatment		experimental group
						with electrical stimulation		

Table 2, continued

Study	study Location Controls Cases Dia	Controls	Cases	Diagnosis	Characteristic	Characteristics of the treatment	Additional treatment	Result
Kim J	Korea	30^{5}	30^{6}	Facial paralysis	30 ⁵ 30 ⁶ Facial paralysis Evolution of the paralysis One week	One week	Control group: Treated	An improvement in facial per-
(2016) [21]				idiopathic	Wave form	Monophasic current	during the first 5 days with	formance was noted in the first
					Frequency	20 Hz-5 KHz	prednisolone (1 mg/kg/day),	2 weeks in the experimental
					Intensity	Sub-motor level (sensory	and decreased during 10 days.	and decreased during 10 days. group, and the time for com-
						threshold)	They were also treated with	plete recovery was significantly
					Pulse width	$100 \mu sec every 50 ms$	acyclovir (1500 mg at day)	shorter than that in the control
					Duration	6 months	during five days	group $(P < 0.05)$
					Combination with drugs	Prednisolone (1 mg/kg/day for	Experimental group: Same	All patients except one showed
						the first 5 days, weaned off	drug treatment of the control	complete recovery in the experi-
						within 2 weeks) for 10 days,	group plus continuous, low	mental group in three months.
						plus acyclovir (1500 mg daily) frequency-impulse electrical	frequency-impulse electrical	In the control group, 5 patients
						for 5 days	stimulation (SCLES)	did not recover normal facial
								function within 6 months

¹Voluntary closure of the eyelid was measured only in 7 individuals, but the Lagophthalmus was used in 10 individuals. ²Eight patients secondary to Neurinoma, one with surgical lesion and one patient with trauma. ³Twelve patients with Bell's palsy and five patients with surgical resection of the facial nerve by acoustic neuroma. ⁴11.4 days for control group and 12.5 days for experimental group. ⁵30 patients used medical treatment only (control group). ⁶30 patients use medical treatment plus electrical stimulator.

3.2. Studies without a comparison group

In a study conducted in Finland by Hyvärinen et al., ten individuals with chronic facial paralysis were enrolled [22]. When electrical stimulation was applied in the facial nerve, patients showed improvement, and the distal latency obtained improved (p = 0.02). Moreover, in a study conducted in the UK, ten individuals with chronic paralysis of the seventh cranial nerve were recruited [23]. They were assessed with EMG system for measuring eyelid function. The results showed that electrical stimulation therapy improved voluntary movement, increasing the displacement of the eyebrows in a range of 1.4 mm to 4.1 mm with an average of 2.5 mm. However, due to their exclusion criteria the study group was reduced to seven individuals. Four of them showed significant improvement in range of motion ($p \le 0.01$).

4. Discussion

This study was conducted in order to determine if the use of electrotherapy is helpful in the treatment of Bell's palsy. The results of this systematic review showed that there is an improvement in patients who received electrotherapy, in both phases, acute and chronic. Although there is not enough evidence regarding the effectiveness of electrotherapy for treating Bell's palsy [24], the results found in this systematic review are oriented towards a positive response to the treatment.

In the present study, we analyzed 131 cases and 113 controls. We included seven publications, while both of the previous systematic reviews only included three or four publications. For instance, the review about physical therapy for treating Bell's palsy performed by Teixeira et al. [25], analyzed 3 trials and reported no significant improvement in patients who received electrical stimulation, questioning its cost-effectiveness. In the update of the same review [14], four trials were analyzed and the results obtained were similar. It is necessary to emphasize that these previous reviews did not aim to specifically evaluate electrotherapy, but multiple physical therapies. In addition, the trials analyzed were not very recent.

On the other hand, there are several countries without an up to date guide for the long term treatment of Bell's palsy [26]. Although it has been stated that receiving electrotherapy in the acute phase of Bell's palsy is beneficial for patients [19–21,27], and it is

highly used in the Mexican clinical practice (for diagnosis and management of Bell's palsy), there is not enough evidence to support the efficacy of electrotherapy in acute cases.

It is necessary to mention that although patients who received electrical stimulation improved their condition in all the studies evaluated, the methodology used in each one was different. For instance, in the study by Tuncay et al. [20], the electro stimulation with a current wave phase began four weeks after diagnosis. The results showed differences in facial re-education according to the House-Brackmann scale. Furthermore, Hyvärinen [22] used 20 Hz electro stimulation and 100 μ seg pulse duration, 30 min/day, for two weeks, increasing the amount of time per day during 6 months until reaching 6 hrs of stimulation. Gittins [23] on the other hand, used a frequency of 2 Hz to 200 Hz and 50 μ seg-200 μ sec pulse length. Clearly, the frequency applied in the study by Hyvärinen [22] was lower than the frequency applied in the study by Gittins [23]; nevertheless, in the study by Hyvärinen [22] the results obtained were satisfactory, since all patients showed significant improvement according to the House-Brackmann scale. In the study by Gittins [23] however, the entire study population showed no improvement after treatment. Thus, we can infer that the improvement observed when using electrotherapy did not depend on the intensity or frequency used.

Unlike previous studies, Manikandan [18] used electrostimulation via galvanic and faradic current for facial muscles stimulation. Although patients showed improvement, we must emphasize that electro stimulation therapy was combined with facial rehabilitation exercises, through which much of this improvement was obtained. In more recent studies [19-21], the results of electrical stimulation treatment in the early phase of Bell's Palsy demonstrated a significant recovery. It is important to mention that this improvement was observed in studies with a considerable n, [20,21] with more than 20 patients in the control and experimental groups, unlike a recent study with a poor n, [19] in which statistical significance was not observed. Another important aspect in the methodology of all the studies was the stimulation level, while some used a motor level [18-20] others applied sub-motor level (sensory threshold) [17,21,22], this represents a controversy, because literature indicates that stimulation at motor level, could favor secondary effects such as synkinesia. It is considered that patients with chronic peripheral paralysis receiving daily treatment with electrostimulation for at least 3 months improve significantly [23]. Nonetheless, depending on the nerve injury and latencies deficiency in nerve conduction, patients with facial paralysis will be considered for a prolonged electro stimulation combined with exercise programs or drug treatment, in order to increase the success of the intervention.

Given the importance of the treatment in Bell's palsy, our findings suggest that the use of electrotherapy may play an important role in the improvement of patients. It would be necessary to develop further studies with similar characteristics such as parameters of frequency, intensity, pulse duration, treatment time, number of sessions, number of contractions and even the same area of stimulation, with the purpose of clarifying the genuine role that electrotherapy plays in Bell's palsy treatment.

Conflict of interest

The authors have no competing interests to report.

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Appendix: PRISMA checklist

Section/topic	#	Checklist item	Reported
	;		on page #
Title	,		į
Title Abstract	_	Identify the report as a systematic review, meta-analysis, or both.	Title page
Structured summary	7	Provide a structured summary including, as applicable: background; objectives; data sources; study eligibility criteria, participants, and interventions; study appraisal and synthesis methods; results; limitations; conclusions and implications of key findings; systematic review registration number.	Abstract
Introduction			
Rationale	ϵ	Describe the rationale for the review in the context of what is already known.	3
Objectives	4	Provide an explicit statement of questions being addressed with reference to participants, interventions, comparisons, outcomes, and study design (PICOS).	4
Methods			
Protocol and registration	S	xists, if and where it can be accessed (e.g., Web address), and, if available, provide registration information	NA
Eligibility criteria	9	including registration number. Specify study characteristics (e.g., PICOS, length of follow-up) and report characteristics (e.g., years considered, language, publication	4-5
Information sources	7	dates of coverage, contact with study authors to identify additional studies) in the	4-5
Search	×	strateov for at least one database including any limits used such that it could be reneated	4-6
Study selection	6	ded in the	2-6
Data collection process	10		9 9
Data conection process	10	Describe mentor of data extraction from reports (e.g., priored rollins, interprincinty, in duplicate) and any processes for confirming and agrangement and agrangement agrangement and agrangement agrangement agrangement and agrangement	0
Data items	11	ch data were sought (e.g., PICOS, funding sources) and any assumptions and simplifications made.	4
Risk of bias in individual	12	ling specification of whether this was done at the study or	4-6
studies			
Summary measures	13		NA
Synthesis of results	7	ethods of handling data and combining results of studies, if done, including measures of consistency (e.g., I²) for each	2–6
;	,		:
Risk of bias across studies Additional analyses	15	Specify any assessment of risk of bias that may affect the cumulative evidence (e.g., publication bias, selective reporting within studies). Describe methods of additional analyses (e.g., sensitivity or subgroup analyses, meta-regression), if done, indicating which were	Table 1 6, Table 2
		pre-specified.	
Results			
Study selection	17	Give numbers of studies screened, assessed for eligibility, and included in the review, with reasons for exclusions at each stage, ideally with	9
Study obsessemetics	18	mescent characteristics for which data were averaged (a or sends size DICOS fallow in nerical) and movide the citations	6 0 Table 2
Risk of bias within studies	19	present characteristics for which data were contacted (e.g., start) size, i i i CO, i onow up period, and provide me characteristics by bias of each study and, if available, any outcome level assessment (see item 12).	Table 1
Results of individual	20	s considered (benefits or harms), present, for each study: (a) simple summary data for each intervention group (b) effect	8-9
studies		estimates and confidence intervals, ideally with a forest plot.	
Synthesis of results	21	Present results of each meta-analysis done, including confidence intervals and measures of consistency.	NA
Risk of bias across studies	22		Table 1
Additional analysis	23	Give results of additional analyses, if done (e.g., sensitivity or subgroup analyses, meta-regression (see Item 16)).	NA

Section/topic	# Checklist item	Reported on page #
Discussion		
Summary of evidence	24 Summarize the main findings including the strength of evidence for each main outcome; consider their relevance to key groups (e.g.,	9–11
	healthcare providers, users, and policy makers).	
Limitations	25 Discuss limitations at study and outcome level (e.g., risk of bias), and at review-level (e.g., incomplete retrieval of identified research,	9–11
	reporting bias).	
Conclusions	26 Provide a general interpretation of the results in the context of other evidence, and implications for future research.	9–11
Funding		
Funding	27 Describe sources of funding for the systematic review and other support (e.g., supply of data); role of funders for the systematic review.	12

Moher D, Liberati A, Tetzlaff J, Altman DG, The PRISMA Group (2009). Preferred Reporting Items for Systematic Reviews and Meta-Analyses: The PRISMA Statement. PLoS Med 6(7): e1000097. doi: 10.1371/journal.pmed1000097.