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REVIEW PAPER

Effects of forest bathing (shinrin-yoku) on levels of cortisol as a stress biomarker: a systematic review and meta-analysis

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Abstract

Forest bathing is a traditional practice characterized by visiting a forest and breathing its air. This review aims to investigate the effects of forest bathing on levels of salivary or serum cortisol as a stress biomarker in order to understand whether forest bathing can reduce stress. Medline/PubMed, Embase, Scopus, Web of Science, Cochrane Library, and Google Scholar were systematically searched for relevant articles. The quality of included trials was assessed following the criteria of the NIH dedicated tools. Afterwards, a qualitative and quantitative synthesis of retrieved evidence was performed. A total of 971 articles were screened; 22 of them were included in the systematic review and 8 in the meta-analysis. In all but two included studies, cortisol levels were significantly lower after intervention in forest groups if compared with control/comparison groups, or a significant pre-post reduction of cortisol levels was reported in the forest groups. The main results of the meta-analysis showed that salivary cortisol levels were significantly lower in the forest groups compared with the urban groups both before (MD = − 0.08 μg/dl [95% CI − 0.11 to − 0.05 μg/dl]; $p < 0.01$; $I^2 = 46\%$) and after intervention (MD = − 0.05 μg/dl [95% CI − 0.06 to − 0.04 μg/dl]; $p < 0.01$; $I^2 = 88\%$). Overall, forest bathing can significantly influence cortisol levels on a short term in such a way as to reduce stress, and anticipated placebo effects can play an important role in it. Further research is advised because of the limited available data.

Keywords Forest bathing · Cortisol · Stress · Placebo effects · Review · Meta-analysis

Introduction

Forest bathing, also called "Shinrin-yoku" in Japanese, is a traditional practice characterized by walking in a forest environment, watching it, and breathing its air (Park et al. 2010). In Japan, forest bathing is considered an "art" and a type of meditation, with its origins deeply rooted in local traditions (Miyazaki 2018).

It is demonstrated that exposure to an outdoor environment with green areas may reduce the experience of stress and ultimately improve health (Kondo et al. 2018). Moreover, studies involving participants from deprived urban communities show, even with evidence obtained from cortisol measurements, that more green areas around living places are associated with lower stress levels (Roe et al. 2013; Thompson et al. 2012). Previous reviews of the scientific literature have indicated that forest bathing in particular can be useful for the immune system function, for cardiovascular and respiratory diseases, and for depression, anxiety, attention deficit hyperactivity disorder, and for stress (Hansen et al. 2017; Lee et al. 2017; Oh et al. 2017). Therefore, "Shinrin-yoku," due to all its potential beneficial effects on health, has been hypothesized to

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have a role in medical prevention, especially for stress and stress-related disorders (Hansen et al. 2017; Morita et al. 2007).

"Stress" can be defined as a response to different stimuli characterized by physiological arousal (Folkman 2013). Cortisol is a glucocorticoid hormone released by adrenal glands, and it represents a sensitive and reliable stress biomarker, even if patient-specific characteristics (age, gender, individual differences in daily pattern of this hormone secretion) should be taken into consideration when thoroughly assessing the effects of any intervention on its levels (Van Cauter et al. 1996; Smyth et al. 1997). Levels of salivary cortisol reflect the concentration of free serum cortisol (Kirschbaum and Hellhammer 1994). Stressful stimuli activate the pituitary-adrenal axis, leading to an increased production of cortisol (Ranabir and Reetu 2011), and have the potential to disrupt the circadian release of this hormone (Tsigos et al. 2016), whereas anti-stress practices like balneotherapy or meditation, which are considered useful to improve stress resilience, are associated with short-term significant lowering of cortisol levels and maybe improvement of cortisol awakening response on a longer term (Antonelli and Donelli 2018; Matousek et al. 2010; Thirthalli et al. 2013; Turakitwanakan et al. 2013). Therefore, this hormone levels can be useful to study psycho-physical benefits of a specific intervention on the so-called stress system (Chrousos 2009), since they help to objectively estimate the integrated effect of an anti-stress practice upon a person's neuro-endocrinological system (Antonelli and Donelli 2018).

The present systematic review and meta-analysis aim to investigate the effects of forest bathing on levels of salivary or serum cortisol as a stress biomarker in order to understand whether forest bathing can reduce stress.

Methods

The PRISMA guidelines were followed for the present systematic review (Liberati et al. 2009).

Eligibility criteria

All studies, regardless of study design, involving healthy participants or patients with a previously diagnosed disease were included in the review. Forest bathing (intervention) was defined as staying in a forest, either walking or simply resting and watching it, and taking in its air for a specified amount of time. Trials were excluded when the study location was a city park or an urban green area. Studies comparing cortisol levels of communities living in places with larger versus smaller green areas were excluded since it was not possible to determine the precise length of exposition and the type of green area (garden, city park, cultivated field, forest), but their results were mentioned in the "Introduction" section. All studies were included regardless of comparison/control group type (control was defined as no intervention, while comparison was defined as any intervention other than forest bathing). All studies measuring the participants' salivary or serum cortisol levels with validated laboratory techniques were included. All articles written in English, French, Spanish, or Italian were included. Articles written in other languages (such as Japanese) but with an English abstract, as well as conference proceedings, were reported in a supplementary table (Supplementary Table A) and mentioned in the "Discussion" section.

The following list summarizes the applied PICOS criteria for inclusion and exclusion of studies in the systematic review:

- P (population): healthy participants or patients with a previously diagnosed disease
- I (intervention): forest bathing
- C (comparison): all types of comparison/control, including no comparison/control
- O (outcomes): measurement of the participants' salivary or serum cortisol levels
- S (study design): all types of study design

Information sources

Medline via PubMed, Embase, Scopus, Web of Science, Cochrane Library, and Google Scholar were systematically searched for relevant articles describing studies on the effects of forest bathing on salivary or serum levels of cortisol. Additionally, three reviews on "Shinrin-yoku" (Hansen et al. 2017; Oh et al. 2017; Park et al. 2010) and one on stress and outdoor environments (Kondo et al. 2018) were also searched for relevant articles matching the inclusion criteria. Finally, a Google search was performed to screen gray literature (mainly conference proceedings) on the topic.

Search

Databases were searched up to February 13, 2019. The following search strategies were used:

- Medline/PubMed (79 results): (shinrin*[Title/Abstract] OR forest bath*[Title/Abstract] OR forest therapy[Title/Abstract] OR forest-therapy[Title/Abstract] OR nature therapy[Title/Abstract])
- Embase (95 results): ("shinrin*":ti,ab,kw OR "forest bath*":ti,ab,kw OR "forest therapy":ti,ab,kw OR "forest-therapy":ti,ab,kw OR "nature therapy":ti,ab,kw)

- Scopus (10 results): TITLE-ABS-KEY ((“shinrin*” OR “forest bath*” OR “forest*therapy” OR “nature therapy”) AND (“cortisol” OR “corticosteroid”))
- Web of Science (30 results): TOPIC: (“shinrin*” OR “forest bath*” OR “forest*therapy” OR “nature therapy”) AND (“cortisol” OR “corticosteroid”) Timespan: All years. Indexes: SCI-EXPANDED, SSCI, A&HCI, CPCI-S, CPCI-SSH, ESCI.
- Cochrane Library (4 results): (forest OR shinrin*) AND cortisol
- Google Scholar (753 results): (“forest bath” OR “shinrin*”) AND “cortisol”
- Google: “shinrin yoku” OR “forest bathing”

Study selection and data collection process

Study screening and selection were performed by two authors independently (M.A. and G.B.). Disagreements were discussed with the third author (D.D.) until consensus was reached. Details of the study selection and data collection process were summarized in a flowchart (Fig. 1).

Among studies comprised in the systematic review, it was decided to include in the meta-analysis only randomized controlled trials (RCTs) describing the effects of forest bathing on salivary cortisol levels of healthy participants compared with the effects of the same activities performed in an urban environment. The following list summarizes the applied PICOS criteria for inclusion and exclusion of studies in the meta-analysis:

- P (population): healthy participants
- I (intervention): forest bathing (forest watching and/or walking)
- C (comparison): visiting a city environment (urban watching and/or walking)
- O (outcomes): measurement of the participants’ salivary cortisol levels
- S (study design): randomized controlled trials

Data were manually extracted from included articles, collected in tables, and then critically appraised. When numerical data were only graphically displayed, a dedicated software was used for data extraction from graphs (Rohatgi 2014). Authors were also contacted by email to retrieve all missing data, even though it was not possible to collect any additional useful information in this way.

Data items

Data items extracted from included studies were the following ones: study design, number of participants (at enrollment and at the end of the trial, when data analysis was performed), characteristics of study population (gender, age, occupation, and diseases, if any), whether salivary or serum cortisol levels were measured and when these measurements were taken, a full description of intervention, plant composition and location of forests used for research purposes, and study results. In particular, the focus was on the significant differences in cortisol levels within the intervention (forest bathing) group (pre-post) ($p < 0{,}05$), significant differences in cortisol levels between groups at baseline ($p < 0{,}05$), and significant differences in cortisol levels between groups after intervention ($p < 0{,}05$). When details on the study forest type and characteristics (plant composition) were not available in included articles, the scientific literature was searched with PubMed and Google Scholar to collect this information.

The number of study participants and means and standard deviations of salivary cortisol levels before and after intervention were extracted from articles included in the meta-analysis to perform the quantitative synthesis. When not directly available, standard deviations were calculated with reported standard errors and number of study participants.

Risk of bias in individual studies

The risk of bias and overall quality of included studies was independently assessed by two authors (M.A., G.B.) following the criteria of two NIH-dedicated tools and taking into consideration the differences in study designs (National Institutes of Health 2014a, 2014b). Disagreements were discussed with the third author (D.D.) until consensus was reached. These tools were chosen due to high heterogeneity of the study design across included trials in order to optimize the quality assessment of their evidence. To evaluate the quality of one study whose cortisol outcome was reported in a letter to the editor (Jia et al. 2016), another article with a more detailed description of the study design was consulted (Mao et al. 2012a).

A dedicated NIH tool was used for the quality assessment of controlled intervention studies (National Institutes of Health 2014a). The overall evaluation was based on the answers to 14 questions, regarding the presence and methods of randomization, the treatment allocation concealment, the blinding of study participants and outcome assessors, the absence of significant differences between groups at baseline, the attrition and dropout rate, the adherence to intervention protocol, the presence of confounding factors, the use of valid and reliable measures, the recruitment of a sufficient number of participants, and other potential sources of bias. Every question could be answered in three ways, either “yes,” “no,” or “other” (which indicates that data are not reported, the answer cannot be determined, or the question is not applicable). Studies were assessed individually and their overall quality was scored as poor if 6 or less of the items were positive (answered with “yes”), fair if positive items ranged from

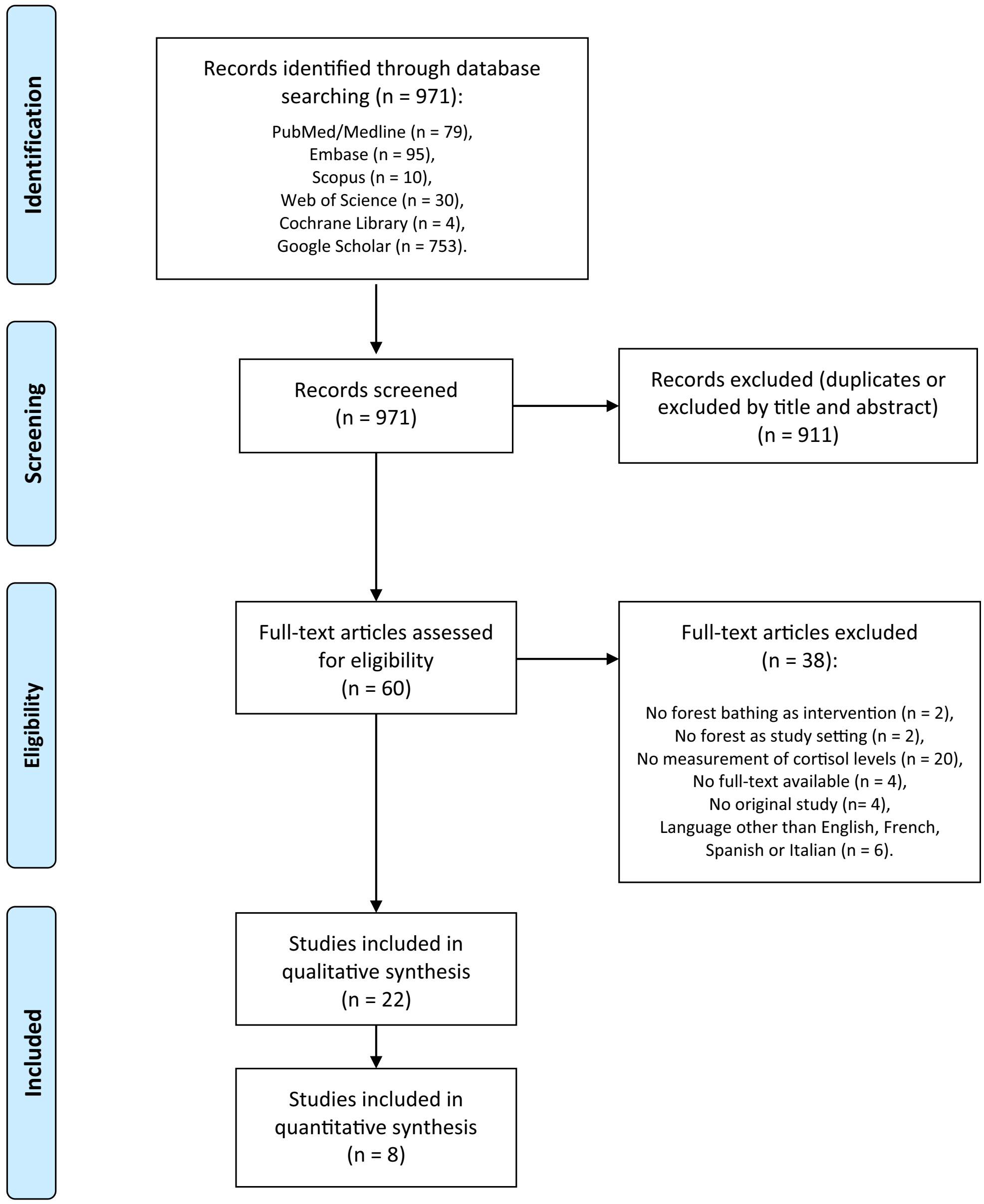

Fig. 1 PRISMA flow diagram of the systematic review and meta-analysis (adapted from Moher et al. 2009)

7 to 9, and good if at least 10 items were positive. When items could not be determined, were not applicable, or were not reported, the overall quality was decided on the basis of available data.

Another dedicated NIH tool was used for the quality assessment of pre-post studies with no control group (National Institutes of Health 2014b). The overall evaluation was based on the answers to 12 questions, regarding the clarity of study objectives, the eligibility criteria of participants and tested intervention, the validity and reliability of outcome measures, the recruitment of a sufficient number of participants, the blinding of study outcome assessors, the attrition and dropout rate, the appropriateness of statistical methods used to analyze data, and other potential sources of bias. Every question could be answered in three ways, either “yes,” “no,” or “other” (which indicates that data are not reported, the answer cannot be determined, or the question is not applicable). Studies were assessed individually and their overall quality was scored as poor if 4 or less of the items were positive (answered with “yes”), fair if positive items ranged from 5 to 7, and good if at least 8 items were positive. When items could not be determined, were not applicable, or were not reported, the overall quality was decided on the basis of available data.

Results of the overall quality assessment were reported in a specific column of Table 1.

The quality of studies included in the quantitative synthesis was also evaluated with the Cochrane Risk-of-Bias tool for randomized controlled trials (Higgins et al. 2011). The risk of bias was independently assessed by two authors (M.A., G.B.) and disagreements were discussed with the third author (D.D.)

Table 1 Characteristics of studies included in the systematic review

Reference	Study design	Number of participants (enrollment/analyzed), study population characteristics and age (years)	Stress marker (salivary/serum cortisol) and hormone sampling time	Full description of intervention	Group 1	Group 2
Dettweiler et al. 2017 *	NRS	48/48 children attending a secondary school. 11.2 ± 0.5 years	Salivary cortisol. Before, during, and after intervention (at 8:30, 10:30, and 12:30 a.m.) three times over the school year (in fall, spring, and summer).	One full school day (8:45 a.m.–1:05 p.m.) per week (over 1 school year) spent in a forest.	Intervention: 37 children	Comparison/indoor lessons at school: 11 children
Horiuchi et al. 2013	NRS	48/42 healthy young volunteers and elderly participants, 21 males and 27 females. Group 1, 21.5 ± 2.8 years; group 2, 59.2 ± 6.2 years	Salivary cortisol. Before and after intervention (unspecified time of day).	Walking at a comfortable pace in a forest for about 3 h and a total distance of 6 km. Drinking beverages with alcohol or caffeine was not allowed.	Intervention: 24 young participants	Intervention: 18 elderly participants
Jia et al. 2016 *	RTC	20/18 patients with chronic obstructive pulmonary disease Group 1 range, 67–77 years; group 2 range, 61–79 years	Serum cortisol. Before and after intervention (before breakfast, at 7:00 a.m., in different days).	Walking in a forest at an unhurried pace for about 1.5 h, with a 20-min rest during the walk, both in the morning and in the afternoon.	Intervention: 10 patients	Comparison/walking in urban area: 8 patients
Jung et al. 2015	NRS	39/38 workers at high risk of stress and burnout, all females. Group 1, 29.4 ± 8.9 years; group 2, 36.5 ± 12.2 years	Salivary cortisol. In the morning (at around 10 a.m.) at days 1 and 3.	Walking and meditating in a forest (2 to 3 h every day for 2 days), as well as being involved into a psychological program characterized by music and cognitive-behavioral therapy for 3 days at a dedicated camp.	Intervention: 18 participants selected among people who used to visit forests less than once a month	Control/no intervention: 20 participants selected among people who used to visit forests at least once a month
Kim et al. 2009 *	NRS	73/63 patients with major depressive disorder, 9 males and 54 females. Group 1, 43.4 ± 12.1 years; group 2, 44.3 ± 13.5 years; group 3, 48.8 ± 9.6 years	Salivary cortisol. Before and after intervention, at 10 a.m.	Cognitive behavior therapy-based psychotherapy applied in a forest environment for 4 weeks with weekly 3-h sessions, from 10 a.m. to 1 p.m. on Wednesdays.	Intervention: 23 participants	Comparison/indoor psychotherapy program: 19 participants
Kobayashi et al. 2017 *	RCT (crossover)	408/348 healthy volunteers, all males. 21.7 ± 1.6 years	Salivary cortisol. After intervention, between 9:30 and 12:00 a.m.	Watching a forest without walking for 15 min.	Intervention: 348 participants	Comparison/watching an urban area: 348 participants
Komori et al. 2017	RCT (crossover)	22/22 healthy volunteers, all males. 31.5 ± 5.6 years	Salivary cortisol. Before and after intervention, between 12 a.m. and 4 p.m.	Walking in a forest for 2 h following a 4-km-long path.	Intervention: 22 participants	Comparison/walking in an urban area: 22 participants
Lee et al. 2009	RCT (crossover)	12/12 healthy volunteers (university students), all males. 21.3 ± 1.1 years	Salivary cortisol. In the morning (6:15–7:15 a.m.), before (2:10–3:00 p.m.) and after (2:30–15:20 p.m.) intervention, and in the evening (6–7 p.m.).	Watching a forest without walking for 15 min after a 15-min resting period on a chair. Smoking or drinking beverages with alcohol was not allowed. Caffeine intake was kept under control.	Intervention: 12 participants	Comparison/watching an urban area: 12 participants
Lee et al. 2011 *	RCT (crossover)	12/12 healthy volunteers (students), all males. 21.2 ± 0.9 years	Salivary cortisol. In the morning, 15 min before, during, and 15 min after intervention.	Watching a forest without walking for 15 min after a 15-min resting period on a chair. Smoking or	Intervention: 12 participants	Comparison/watching an urban area: 12 participants

Table 1 (continued)

				drinking beverages with alcohol or caffeine was not allowed.		
Mao et al. 2012b *	RCT	20/20 healthy volunteers (students), all males. 20.8 ± 0.5 years	Serum cortisol. In the morning (before breakfast) both before and after intervention.	Walking at an unhurried pace for about 1.5 h in a forest, with a 10-min rest during the walk, both in the morning and in the afternoon.	Intervention: 10 participants	Comparison/walking in an urban area: 10 participants
Ochiai et al. 2015a *	NRS (uncontrolled trial)	17/17 healthy or sub-healthy (6 patients with treated hypertension) volunteers, all females. 62.2 ± 9.4 years	Salivary cortisol. In the afternoon (between 3:28 and 3:35 p.m.) on the day before intervention, and between 2:57 and 3:05 p.m. after intervention.	Walking, resting, deep breathing, having lunch, and taking a lecture in a forest. The entire program lasted 4 h and 41 min.	Intervention: 17 participants	
Ochiai et al. 2015b *	NRS (uncontrolled trial)	9/9 sub-healthy participants with high-normal blood pressure, all males. 56.0 ± 13.0 years	Serum cortisol. In the afternoon (between 3:14 and 3:35 p.m.) on the day before intervention and after intervention.	Walking, resting, deep breathing, having lunch, and taking a tourist train in a forest. The entire program lasted 4 h and 35 min.	Intervention: 9 participants	
Olafsdottir et al. 2018	RCT	90/64 healthy subjects (students), unspecified gender data for cortisol outcome. 24.4 ± 2.6 years	Salivary cortisol. In the afternoon (immediately before and after intervention, and 20–30 min after it, following the application of a cold stimulus).	Walking for 40 ± 5 min in a forest along a specific path. Smoking, drinking, and eating were not allowed. The same experiment was performed during an exam and a no-exam period.	Intervention: 20 participants	Comparison 1/indoor walking on a treadmill: 23 participants
Park et al. 2007 *	RCT (crossover)	12/12 for walking and 9 for watching healthy volunteers (students), all males. 22.8 ± 1.4 years	Salivary cortisol. In the morning (6:15–7:15 a.m.), before (10:40–11:30 a.m.), and after (11:00–11:50 a.m.) forest walking, before (2:10–3 p.m.) and after (2:30–3:20 p.m.) forest watching, and in the evening (6–7 p.m.).	Walking in a forest for 20 min in the morning then watching a forest (without walking) for 20 min in the afternoon (same day).	Intervention: 12/9 participants	Comparison/walking in an urban area then watching it: 12/9 participants
Park et al. 2008 *	RCT (crossover)	12/12 healthy volunteers (students), all males. 21.3 ± 1.1 years	Salivary cortisol. In the morning (6:30–7:30 a.m.), before (1:50–4 p.m.) and after (2:10–4:20 p.m.) intervention, and in the evening (6:20–7:20 p.m.).	Watching a forest without walking for 15 min.	Intervention: 12 participants	Comparison/watching an urban area: 12 participants
Park et al. 2009 *	RCT (crossover)	280/260 for watching, 74 for walking healthy volunteers (students), all males. 21.7 ± 1.5 years	Salivary cortisol. Early in the morning (before breakfast), before and after forest watching, and before and after forest walking.	Watching a forest for 14 ± 2 min, then walking in the forest for 16 ± 5 min.	Intervention: 260/74 participants	Comparison/watching an urban area then walking in it: 260/74 participants
Sung et al. 2012 *	NRS (non-randomized controlled trial)	56/56 patients with hypertension, 22 males and 34 females. Group 1, 66 ± 7 years; group 2: 63 ± 11 years	Salivary cortisol. At initial visits (before intervention) and at 8-week final visits (after intervention).	Cognitive behavior therapy-based program in a forest environment (3-day program similar to that one described in the study by Kim et al. 2009) .	Intervention: 28 patients	Control/no intervention: 28 patients
Toda et al. 2013 *	NRS (non-randomized crossover trial)	20/20 healthy volunteers, all males. 67.6 ± 2.8 years	Salivary cortisol. Immediately before (10:15 a.m.) and after (11:00 a.m.) intervention, and 20 min (11:20 a.m.) and 40 min after walking finished (11:40 a.m.).	Walking in a forest following a 1000-m-long path for around 45 min.	Intervention: 20 participants	Control/no intervention: 20 participants
Triguero-Mas et al. 2017 *	RCT (crossover)	26/26 healthy subjects, 11 males and 15 females. 44.3 ± 26.2 years	Salivary cortisol. In the morning, at baseline (9:00 a.m.), before and after the 30-min intervention, before and after the	Spending 30 min in a forest, then resting (marker sampling), then spending 180 min in the same	Intervention: 26 participants	Comparison 1/spending time on the beach: 26 participants

Table 1 (continued)

			180-min intervention, and in the evening (6:00 p.m.).	environment, presumably walking (no specific activity was prescribed by researchers). Caffeine was not allowed.		
Tsunetsugu et al. 2007 *	RCT (crossover)	12/9 for walking and 11 for watching healthy volunteers (students), all males. 21 0 ± 1 0 years	Salivary cortisol. In the morning before breakfast (06:15–07:15 a.m.), before walking (10:40–11:30 a.m.), after walking (11:00–11:50 a.m.), before watching (2:10–3:00 p.m.), after watching (2:30–3:20 p.m.), in the evening before dinner (6:00–7:00 p.m.).	Walking in a forest for 15 min in the morning then watching the forest (without walking) for 15 min in the afternoon (same day).	Intervention: 9/11 participants	Comparison/watching an urban area, then walking in it: 9/11 participants
Tyrväinen et al. 2014	RCT (crossover)	95/77 healthy volunteers, 6 males and 71 females. 47.6 ± 8.7 years	Salivary cortisol. Before watching the forest/city park/urban area, between watching and walking in the same place, after walking.	Watching a forest for 15 min then walking in it for 30 min, covering a distance of around 2 km.	Intervention: 77 participants	Comparison 1/watching a city park, then walking in it: 77 participants
Yu et al. 2016	NRS (uncontrolled trial)	24/24 postmenopausal women (12 of them with chronic illnesses), all females. Range, 50–79 years	Salivary cortisol. Always at 11 a.m., before and after intervention, as well as 2 and 4 weeks after it.	Watching a forest, as well as walking and meditating in it, for 3 days at a dedicated camp.	Intervention: 24 participants	

Reference	Group 3	Significant differences in cortisol levels within intervention group (pre-post) ($p < 0.05$)	Significant differences in cortisol levels between groups at baseline ($p < 0.05$)	Significant differences in cortisol levels between groups after intervention ($p < 0.05$)	Quality of evidence (NIH)
Dettweiler et al. 2017 *		Yes—decrease	NR	Yes (intervention < comparison)	+
Horiuchi et al. 2013		Yes—decrease (young participants), No (elderly participants)	Yes (young > elderly participants)	No	+
Jia et al. 2016 *		No	No	Yes (intervention < comparison)	++
Jung et al. 2015		Yes—increase	No	Yes (intervention > control)	++
Kim et al. 2009 *	Control/no intervention: 21 participants	Yes—decrease	No	Yes (intervention < comparison and control)	++
Kobayashi et al. 2017 *		–	–	Yes (intervention < comparison)	+++
Komori et al. 2017		Yes—decrease	NR	No	++
Lee et al. 2009		No	Yes (intervention < comparison)	Yes (intervention < comparison)	+++
Lee et al. 2011 *		No	Yes (intervention < comparison)	No	++
Mao et al. 2012b *		NR	No	Yes (intervention < comparison)	+++
Ochiai et al. 2015a *		Yes—decrease	–	–	+
Ochiai et al. 2015b *		Yes—decrease	–	–	++
Olafsdottir et al. 2018	Comparison 2/indoor nature video watching: 21 participants	Yes—decrease (both during the exam and during the no-exam period)	No	Yes (intervention < comparison 2) during the exam period, no during the no-exam period	++

Table 1 (continued)

Park et al. 2007 *		NR	Yes (both before walking and watching, intervention < comparison)	No (after walking), Yes (after watching, intervention < comparison)	++
Park et al. 2008 *		NR	No	Yes (intervention < comparison)	++
Park et al. 2009 *		NR	NR	Yes (both after watching and after walking, intervention < comparison)	++
Sung et al. 2012 *		Yes—decrease	No	Yes (intervention < control)	++
Toda et al. 2013 *		Yes—decrease	No	Yes (intervention < control)	+
Triguero-Mas et al. 2017 *	Comparison 2/spending time in an urban environment: 26 participants	Yes—decrease	No	Yes (intervention < comparison 2)	+++
Tsunetsugu et al. 2007 *		Yes—decrease (pre-post walking), no (pre-post watching)	No	Yes (both after watching and after walking, intervention < comparison)	++
Tyrväinen et al. 2014	Comparison 2/watching an urban area, then walking in it: 77 participants	Yes—decrease	No	No	+++
Yu et al. 2016		No (immediately after intervention), yes (2 and 4 weeks after intervention)	/	/	+++

NR, not reported; *NRS*, non-randomized study; *RCT*, randomized controlled trial

Quality of evidence from included studies (assessed with dedicated NIH tools): +++ = good quality; ++ = fair quality; + = poor quality

Articles are alphabetically listed according to the first author's surname. Studies described in articles indexed in Medline or Embase are marked with an * in the first column. Data regarding participants' age have been approximated to the first digit after decimal points until consensus was reached. Assessed domains were selection bias (random sequence generation and allocation concealment), reporting bias (selective reporting), performance bias (blinding of participants and personnel), detection bias (blinding of outcome assessment), attrition bias (incomplete outcome data), and other biases (other sources of bias). Performance bias was not considered a key domain because forest bathing necessarily requires the subject's participation and direct involvement. Studies were considered at high overall risk of bias when there was a high risk of bias in at least one key domain or unclear risk of bias in at least two key domains. Studies were considered at unclear overall risk of bias if only one key domain had unclear risk of bias. If all the key domains had a low risk of bias, the overall risk of bias was reported to be low too.

Results of this additional quality assessment were reported in a table (Supplementary Table B).

Summary measures

Mean difference (MD) was used as a measure of effect size in the quantitative synthesis. Given that studies included in the meta-analysis were selected in such a way as to be as homogeneous as possible, a fixed-effect model was adopted not to overweight data from studies with a very small sample size, thus better estimating sub-group and overall size effects.

Synthesis of results

Results were summarized in two tables (Table 1 reporting characteristics and quality of included trials and Table 2 reporting information about study forest type and location) and then discussed to obtain a qualitative synthesis. For the sake of completeness of our review, another table (Supplementary Table A) was created and filled with data of studies reported in conference proceedings or described in articles with no English/French/Spanish/Italian full text but with at least an English abstract or summary. Afterwards, a quantitative synthesis with selected RCTs was performed. The software used to perform the meta-analyses was "Review Manager" (RevMan, version 5.3).

Pre-post effect size analysis was excluded due to possible biased outcomes (Cuijpers et al. 2017) and only controlled studies were included in the quantitative synthesis. On the basis of available data and considering the aim of this work, it was decided to perform two main meta-analyses (Figs. 2 and 3) which pooled concentration levels (measured in μg/dl) of salivary cortisol before and after forest bathing (walking in a forest or simply watching it) compared with the same hormone levels before and after visiting an urban area. When necessary, cortisol level concentrations were converted from nmol/l to μg/dl. Two additional meta-analyses (Supplementary Figures A and B) were performed with available data. The first one (Supplementary Figure A) pooled concentration levels of salivary cortisol early in the morning (at waking up) of the day of intervention or comparison. The other one (Supplementary Figure B) pooled pre-post differences in concentration levels of salivary cortisol of intervention (forest bathing) groups compared with the same data regarding comparison (urban visiting) groups. A sensitivity analysis was performed to understand whether changing the degree of correlation between pre- and post-intervention cortisol levels could affect the overall results. Then, results were critically appraised and discussed.

I^2 was used as a measure of consistency. I^2 values of 25%, 50%, and 75% were interpreted as representing small, moderate, and high levels of heterogeneity (Higgins et al. 2003).

Risk of bias across studies

Publication bias of studies included in the main meta-analyses (Figs. 2 and 3) was visually assessed with funnel plots (Fig. 4).

Additional analyses

When possible, a sub-group analysis was performed to better estimate the effect size of the two main components of forest bathing, namely forest watching and walking, compared with the same activities performed in an urban environment.

Results

Databases were searched as reported above and 971 articles were retrieved. After article screening and selection, twenty-two (22) studies were included in the systematic review (Dettweiler et al. 2017; Horiuchi et al. 2013; Jia et al. 2016; Jung et al. 2015; Kim et al. 2009; Kobayashi et al. 2017; Komori et al. 2017; Lee et al. 2009; Lee et al. 2011; Mao et al. 2012b; Olafsdottir et al., 2018; Ochiai et al. 2015a; Ochiai et al. 2015b; Park et al. 2007; Park et al. 2008; Park et al. 2010; Sung et al. 2012; Toda et al. 2013; Triguero-Mas et al. 2017; Tsunetsugu et al. 2007; Tyrväinen et al. 2014; Yu et al. 2016) and eight (8) studies in the meta-analysis (Kobayashi et al. 2017; Komori et al. 2017; Lee et al. 2009; Lee et al. 2011; Park et al. 2007; Park et al. 2008; Park et al. 2010; Tsunetsugu et al. 2007). Data regarding article screening and selection process were summarized in a dedicated flowchart (Fig. 1).

Characteristics of included studies were reported in a table (Table 1). Among them, it was possible to identify three (3) uncontrolled studies (Ochiai et al. 2015a, b; Yu et al. 2016) and nineteen (19) controlled trials (Dettweiler et al. 2017; Horiuchi et al. 2013; Jia et al. 2016; Jung et al. 2015; Kim et al. 2009; Kobayashi et al. 2017; Komori et al. 2017; Lee et al. 2009; Lee et al. 2011; Mao et al. 2012b; Olafsdottir et al.,

Table 2 Forest type, location, and characteristics of included studies

Reference	Design	Location	Forest type and characteristics
Dettweiler et al. 2017	NRS	Germany	A German forest near Heidelberg (no details reported).
Horiuchi et al. 2013	NRS	Japan	A Japanese forest (no details reported).
Jia et al. 2016	RCT	China	A forest with the following predominant species: *Ormosia hosiei*, *Cinnamomum camphora*, *Magnolia officinalis* subsp. *biloba*, and *Nyssa sinensis*.
Jung et al. 2015	NRS	South Korea	Saneum recreational forest, with many fir trees (Seo et al. 2015).
Kim et al. 2009	NRS	South Korea	The Hong-Reung arboretum, a forest garden with acicular trees, broadleaf trees, shrubs, herb gardens, and alpine plants.
Kobayashi et al. 2017	RCT	Japan	34 forests across Japan.
Komori et al. 2017	RCT	Japan	Kumano-Kodo forest, a natural place of sacred pilgrimage in Japan.
Lee et al. 2009	RCT	Japan	A forest dominated by *Pinus densiflora Sieb. et Zucc.*, *Quercus serrata Thunb.* and *Castanea crenata Sieb. et Zucc.*
Lee et al. 2011	RCT	Japan	A forest characterized by broadleaved deciduous trees.
Mao et al. 2012b	RCT	China	Wuchao mountain forest with the following predominant species: *Ormosia hosiei*, *Diospyros glaucifolia*, *Chamaecyparis pisifera*, *Zelkova schneideriana*, *Machilus pauhoi*, *Cladrastis platycarpa*, *Manglietia yuyuanensis*, *Cinnamomum camphora*, *Ormosia hosiei*, *Magnolia officinalis subsp. biloba*, and *Nyssa sinensis*.
Ochiai et al. 2015a	NRS	Japan	Akasawa Natural Recreation Forest in Nagano Prefecture composed of 300–350-year-old Kiso cypresses (*Chamaecyparis obtusa*) (Zhang et al. 2015).
Ochiai et al. 2015b	NRS	Japan	Akasawa Natural Recreation Forest in Nagano Prefecture composed of 300–350-year-old Kiso cypresses (*Chamaecyparis obtusa*) (Zhang et al. 2015).
Olafsdottir et al. 2018	RCT	Iceland	A forest, near Reykjavík, with 26 different tree species predominated with the Sitka Spruce (*Picea sitchensis*).
Park et al. 2007	RCT	Japan	Seiwa Prefectural Forest Park with oak trees (*Quercus acutissima Carruth.*, *Quercus serrata Thunb.*).
Park et al. 2008	RCT	Japan	A mixed forest in Shinano Town.
Park et al. 2010	RCT	Japan	24 forests across Japan.
Sung et al. 2012	NRS	South Korea	Two recreation forest sites: Hoengseong and Saneum. Saneum forest mainly consists of Korean pine (*Pinus koraiensis*) and other broadleaf trees mixed. Hoengseong forest is mainly composed of Japanese Larch (*Larix kaempferi*) and other broadleaf trees mixed.
Toda et al. 2013	NRS	Japan	A forest on Ikoma Mountain at the edge of Osaka Prefecture with deciduous trees (Muramatsu et al. 2000).
Triguero-Mas et al. 2017	RCT	Spain	The Collserola Natural Park, near Barcelona, dominated by Aleppo pine (*Pinus halepensis*), and oaks (*Quercus ilex* and *Quercus cerrioides*) with dense forest floor.
Tsunetsugu et al. 2007	RCT	Japan	A deciduous broadleaf forest mainly consisting of old-growth beech.
Tyrväinen et al. 2014	RCT	Finland	Keskuspuisto forest, near Helsinki, with conifers and other trees (Ojala et al. 2018).
Yu et al. 2016	NRS	South Korea	Chungbuk University's experimental forest at Woraksan National Park with many pine trees.

RCT, randomized controlled trial; *NRS*, non-randomized study

Studies are alphabetically sorted according to the first author's surname

2018; Park et al. 2007; Park et al. 2008; Park et al. 2010; Sung et al. 2012; Toda et al. 2013; Triguero-Mas et al. 2017; Tsunetsugu et al. 2007; Tyrväinen et al. 2014).

Eleven (11) trials had a crossover design (Kobayashi et al. 2017; Komori et al. 2017; Lee et al. 2009; Lee et al. 2011; Park et al. 2007; Park et al. 2008; Park et al. 2009; Toda et al. 2013; Triguero-Mas et al. 2017; Tsunetsugu et al. 2007; Tyrväinen et al. 2014) and nine (9) studies lacked any form of randomization (Dettweiler et al. 2017; Horiuchi et al. 2013; Jung et al. 2015; Kim et al. 2009; Ochiai et al. 2015a; Ochiai et al. 2015b; Sung et al. 2012; Toda et al. 2013; Yu et al. 2016). The number of patients varied across studies, ranging from 9 to 348 (median = 23). In all included studies, except one (Dettweiler et al. 2017), participants were adults, mostly healthy volunteers, as shown in Table 1. In only three (3) studies, the participants' levels of serum cortisol were measured (Jia et al. 2016; Mao et al. 2012b; Ochiai et al. 2015b), while in the other trials, levels of salivary cortisol were assessed. The length of intervention (forest bathing) varied across studies, ranging from 15 min (Kobayashi et al. 2017; Park et al. 2008) to half a day (Dettweiler et al. 2017). Moreover, in four (4) studies, intervention did not comprise any walking activity in the forest, but only watching the natural environment while resting (Kobayashi et al. 2017; Lee et al. 2009; Lee et al. 2011; Park et al. 2008), and in one study, participants were free to choose how to spend their time in the forest, presumably walking most of the time, although full rest or strenuous physical activity was not allowed (Triguero-Mas et al. 2017).

When considering results of the included controlled studies, fourteen (14) (Dettweiler et al. 2017; Jia et al. 2016; Kim et al. 2009; Kobayashi et al. 2017; Lee et al. 2009; Mao et al. 2012b; Olafsdottir et al., 2018; Park et al. 2007; Park et al. 2008; Park et al. 2009; Sung et al. 2012; Toda et al. 2013; Triguero-Mas et al. 2017; Tsunetsugu et al. 2007) of them reported significant differences ($p < 0.05$) in cortisol concentrations between groups after intervention, with lower levels in the forest group. In four (4) studies, no significant differences between groups were found after intervention (Horiuchi et al. 2013; Komori et al. 2017; Lee et al. 2011; Tyrväinen et al. 2014), while in one study, levels of cortisol were significantly higher in the forest group after intervention (Jung et al. 2015). When considering included studies with no control/comparison group, pre-post analysis revealed a significant decrease in cortisol levels (Ochiai et al. 2015a, b; Yu et al. 2016), although, in one of them, the reduction of these hormone levels was not reported immediately after intervention, but only 2 to 4 weeks after it, in the follow-up period (Yu et al. 2016).

All studies were performed in Asia (Japan, China, South Korea) or Europe (Germany, Iceland, Finland, Spain) and forests were mostly composed of broadleaf deciduous and/or evergreen coniferous trees, as shown in Table 2.

According to an overall evaluation based on the NIH tool, the quality of evidence from the included studies was rated as good for six (6) studies (Kobayashi et al. 2017; Lee et al. 2009; Mao et al. 2012b; Triguero-Mas et al. 2017; Tyrväinen et al. 2014; Yu et al. 2016), fair for twelve (12) studies (Jia et al. 2016; Jung et al. 2015; Kim et al. 2009; Komori et al. 2017; Lee et al. 2011; Ochiai et al. 2015b; Olafsdottir et al., 2018; Park et al. 2007; Park et al. 2008; Park et al. 2009; Sung et al. 2012; Tsunetsugu et al. 2007), and poor for four (4) studies Table 1 (Dettweiler et al. 2017; Horiuchi et al. 2013; Ochiai et al. 2015a; Toda et al. 2013). Overall risk of bias (assessed with the Cochrane tool) of trials included in the quantitative synthesis was unclear for three (3) studies (Kobayashi et al. 2017; Lee et al. 2009; Lee et al. 2011) and high for five (5) studies (Supplementary Table B) (Komori et al. 2017; Park et al. 2007; Park et al. 2008; Park et al. 2009; Tsunetsugu et al. 2007). The most important sources of bias were identified as unclear information about allocation concealment and attrition bias (Supplementary Table B).

The main results of the quantitative synthesis showed that salivary cortisol levels were significantly lower in the forest bathing groups compared with the urban groups both before (MD = − 0.08 μg/dl [95% CI − 0.11 to − 0.05 μg/dl]; $p < 0.01$) and after intervention (MD = − 0.05 μg/dl [95% CI − 0.06 to − 0.04 μg/dl]; $p < 0.01$). A moderate-to-low level of heterogeneity across studies was found in the first meta-analysis (I^2 = 46%; Fig. 2), while a high level of heterogeneity was found in the second meta-analysis ($I^2 = 88\%$; Fig. 3). When considering the sub-group analysis, salivary cortisol levels were significantly lower in groups assigned to forest watching compared with groups assigned to urban watching both before (MD = − 0.10 μg/dl [95% CI − 0.13 to − 0.07 μg/dl]; $p < 0.01$) and after intervention (MD = − 0.05 μg/dl [95% CI − 0.07 to − 0.04 μg/dl]; $p < 0.01$). Furthermore, salivary cortisol levels were lower, although not significantly, in groups assigned to forest walking compared with groups assigned to urban walking before intervention (MD = − 0.05 μg/dl [95% CI − 0.09 to 0.00 μg/dl]; $p = 0.06$), whereas a significant difference was found after intervention (MD = − 0.04 μg/dl [95% CI – 0.07 to − 0.01 μg/dl]; $p < 0.01$).

Moreover, cortisol levels early in the morning (at waking up) of the day of intervention were significantly lower in the forest groups compared with the urban groups (MD = − 0.15 μg/dl [95% CI – 0.23 to − 0.08 μg/dl]; $p < 0.01$), although the level of heterogeneity was high ($I^2 = 68\%$) (Supplementary Figure A). Finally, the last analysis showed that no significant differences were found in pre-post variations in cortisol levels of the forest groups compared with the same data regarding comparison urban groups (if $r = 0.5$, so MD = − 0.02 μg/dl [95% CI – 0.05 to 0.01 μg/dl]; $p = 0.20$) (Supplementary Figure B). Despite changes in the degree of correlation ($r = 0.1$ or $r = 0.9$) applied for the sensitivity analysis, the overall mean difference remained non-significant (Supplementary Figure B). However, when considering the sub-group analysis, results showed a significant tendency to favor forest watching if compared with urban watching in terms of pre-post differences in cortisol levels (if $r = 0.5$, so MD = − 0.05 μg/dl [95% CI – 0.08 to − 0.01 μg/dl]; $p < 0.01$). The sensitivity analysis showed that this tendency had a significance of $p = 0.04$ if $r = 0.1$, and of $p < 0.01$ if $r = 0.9$.

Asymmetry in funnel plots (Fig. 4) suggested a potential risk of publication bias with a possible over-representation of trials with low precision and with results in favor of forest bathing.

Discussion

Eleven (11) included trials had a crossover design with participants serving as their own controls (Kobayashi et al. 2017; Komori et al. 2017; Lee et al. 2009; Lee et al. 2011; Park et al. 2007; Park et al. 2008; Park et al. 2009; Toda et al. 2013; Triguero-Mas et al. 2017; Tsunetsugu et al. 2007; Tyrväinen et al. 2014). This particular study design is usually adopted when a long-term follow-up is not required and when investigated conditions are unlikely to significantly change over

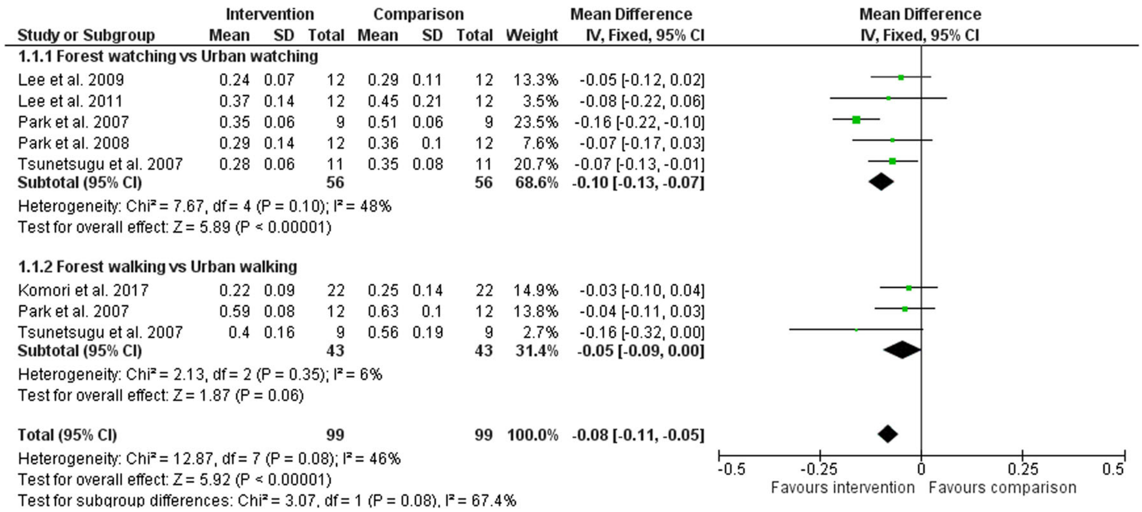

Fig. 2 Forest plot referred to the meta-analysis about cortisol levels *before* intervention (forest bathing) versus comparison (urban visiting). Description: concentration levels (measured in μg/dl) of salivary cortisol before forest bathing (walking in a forest or simply watching it) compared with the same hormone levels before visiting an urban area. Two sub-groups are described, each of them characterized by a different component of studied intervention. In each sub-group, studies are listed according to the first author's surname. Means and standard deviations are reported in columns and a fixed-effect model was adopted to estimate sub-group and overall size effects

time (Higgins 2015). In the included trials, the main focus was on measuring short-term variations of levels of a stress hormone (cortisol) which is expected to show a predictable circadian cycle (Tsigos et al. 2016). Investigators might have adopted a crossover design in order to obtain significant results with numerically limited study samples. However, the order and the carry-over effects cannot be fully excluded, the former indicating the influence on results of the order of administration of intervention and control, while the latter referring to the lasting effects on a long-term of one treatment on outcomes, thus introducing a confounding factor in the evaluation of the effects of the second treatment (Freeman 1989). In one study, although uncontrolled and involving a limited number of heterogeneous participants, it was suggested that forest bathing can significantly influence cortisol levels 2 to 4 weeks after intervention (Yu et al. 2016). Evidence from studies investigating the influence of forest bathing on the immune function underscored that the effects of this practice may last from 7 (Li et al. 2008a; Li et al. 2008b; Li et al. 2010) up to more than 30 days (Li 2010) after intervention. These considerations need to be taken into account when appraising evidence from included trials, whose

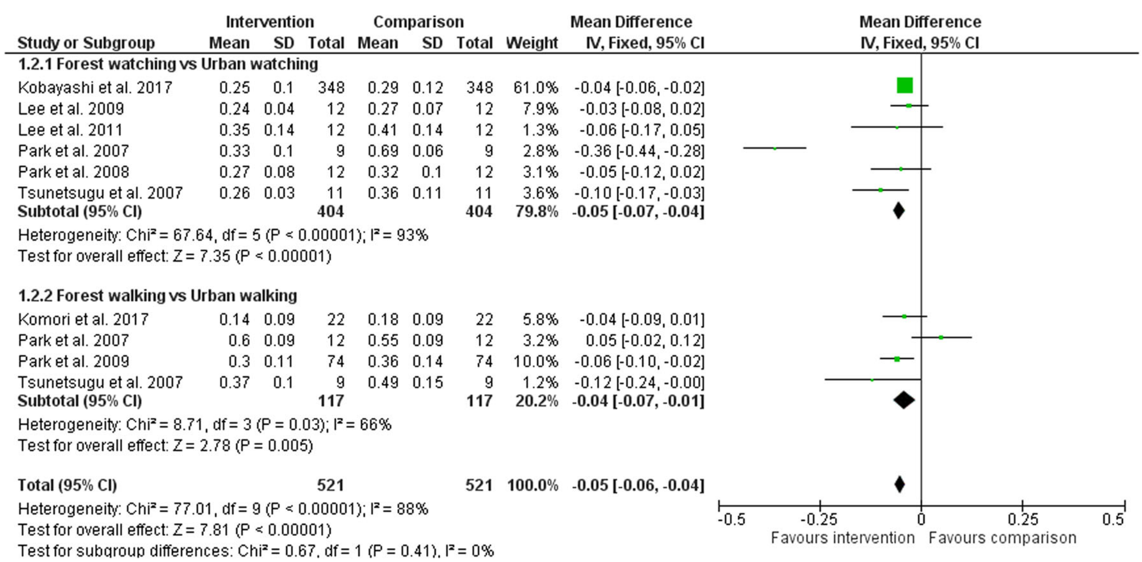

Fig. 3 Forest plot referred to the meta-analysis about cortisol levels *after* intervention (forest bathing) versus comparison (urban visiting). Description: concentration levels (measured in μg/dl) of salivary cortisol after forest bathing (walking in a forest or simply watching it) compared with the same hormone levels after visiting an urban area. Two sub-groups are described, each of them characterized by a different component of studied intervention. In each sub-group, studies are listed according to the first author's surname. Means and standard deviations are reported in columns and a fixed-effect model was adopted to estimate sub-group and overall size effects

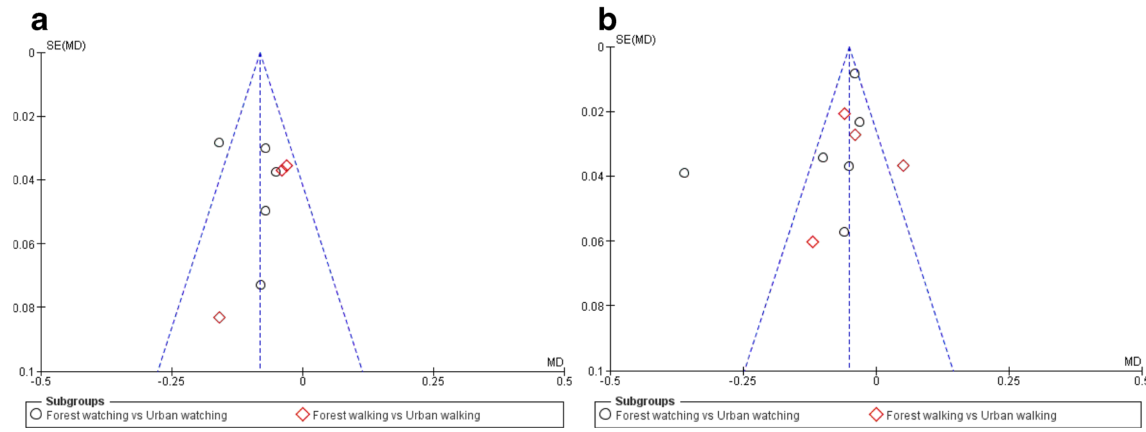

Fig. 4 Funnel plots for a visual assessment of publication bias. Description: funnel plot A (on the left) refers to the meta-analysis displayed in Fig. 2 about cortisol levels *before* intervention (forest bathing) versus comparison (urban visiting). Funnel plot B (on the right) refers to the meta-analysis displayed in Fig. 3 about cortisol levels *after* intervention (forest bathing) versus comparison (urban visiting)

results in favor of "Shinrin-yoku" might actually underestimate the effects of this practice on stress hormone levels, especially when forest bathing was administered as the first intervention.

Overall, all but two included studies (Jung et al. 2015; Lee et al. 2011) indicated that cortisol levels were either significantly diminished within the intervention (forest) group, or significantly lower after intervention in the forest group if compared with control/comparison group values. Further evidence from studies reported in conference proceedings or with Japanese full text seems to confirm these findings (Kondo and Takeda, 2017; Tsunetsugu and Miyazaki, 2007; Kondo et al. 2011; Song et al. 2015), with reported beneficial effects even for particular study populations usually dealing with performance stress, such as competitive athletes (Kondo and Takeda, 2017). Findings of the trial conducted by Olafsdottir et al. (2018) suggested that the cortisol-lowering effect of forest bathing may be more pronounced among subjects facing a stressful period (students during an exam period), thus underscoring, along with evidence from the abovementioned study with athletes (Kondo and Takeda, 2017), that "Shinrin-yoku" may be useful for the management of both mental and physical performance stress. On the contrary, the study by Jung et al. (2015) reported a significant increase in cortisol levels within the intervention group after forest bathing. Several factors might have contributed to this outlying result, for example the inclusion of a relatively limited number of study participants with impaired baseline levels of stress (workers at high risk of burnout, all females) and the administration of a multi-component intervention comprising even music listening and a cognitive-behavioral therapy for 3 days at a dedicated camp. Moreover, at baseline, a significant difference between groups was reported in terms of depression as a Worker's Stress Response Inventory (WSRI) item, which implied a cortisol-disrupting underlying condition in the forest group (Burke et al. 2005; Jung et al. 2015). In addition to this, given that study participants assigned to the forest group were recruited among people who used to visit forests less than once in a month, the stress induced by performing several activities in a wild and unknown environment might have counteracted the anti-stress effects of forest bathing. In the study by Lee et al. (2011), no significant variations within or between groups were detected after intervention. In this trial, a very limited number of study participants were recruited ($n = 12$, all males) and this might have biased results. However, a positive anticipatory effect of forest bathing on cortisol levels was found. In fact, a comparison of this stress hormone levels between groups before intervention showed that values were significantly lower in the forest group immediately before intervention and early in the morning (Lee et al. 2011).

The quantitative synthesis showed that forest bathing, in particular forest watching, was associated with significantly lower cortisol levels both before and after this practice if compared with visiting an urban area (Figs. 2 and 3). Therefore, it can be assumed that an important anti-stress component of forest bathing was its "anticipatory effect" (i.e., a placebo effect) on levels of cortisol (Kaptchuk et al. 2008). Study participants who were informed that they were going to practice "Shinrin-yoku" reported significantly lower cortisol levels if compared with volunteers who were sent to visit an urban area. An interesting study by Berk et al. (2008) involving fasting healthy males undergoing a humor/laughter experience demonstrated that "the perception of anticipating a positive eu-stress event decreases stress hormones cortisol and catecholamines, concomitant with positive mood state changes." Forest bathing is considered an anti-stress practice and planning to visit a forest seems to positively influence cortisol levels, even before physically interacting with it; therefore, watching a forest, and possibly even the sole mental visualization of a forest, may have a role in triggering anticipated placebo effects.

When considering the sub-group analysis, it can be observed that cortisol levels were significantly lower in intervention groups after forest watching if compared with the same values after urban watching (Fig. 3). Evidence from neurophysiological investigations measuring regional hemoglobin saturation in the brain demonstrated that the prefrontal cortex activity of volunteers assigned to a forest area was more "stabilized" than that of participants assigned to an urban area, and this correlated with subjective relaxation (Joung et al. 2015). It was also demonstrated that viewing a real forest may have a positive effect on cerebral activity and psychological responses, while staying in a forest and breathing its air without having the opportunity to visually interacting with it did not exert the same beneficial effects (Horiuchi et al. 2014). Globally, these results may have interesting implications for the development of new strategies for stress management and for prevention of stress-related conditions.

If we still consider the sub-group analysis, it can be noted that cortisol levels were significantly lower in the intervention groups after forest walking if compared with the same values after urban walking (Fig. 3). It is possible to hypothesize that performing a light physical activity in a green environment could have contributed to the reduction of stress levels, as demonstrated by other studies (Marselle et al. 2013), thus diminishing the participants' cortisol levels on a short term.

The analysis also showed that no significant differences were found in pre-post variations of cortisol levels of the forest groups compared with the same data regarding comparison urban groups in terms of overall effect size (Supplementary Figure B). These results, in line with the findings of the study by Lee et al. (2011), seem to indicate that, when comparing the effects of forest bathing versus urban visiting, the anticipated placebo effect related to planning and visualizing the intervention may play a more important role in influencing cortisol levels rather than the actual experience of "Shinrin-yoku." It has to be acknowledged that some missing and un-retrievable data might have biased this result and that, when considering the sub-group analysis, the tendency to favor forest watching if compared with urban watching in terms of pre-post differences of cortisol levels indicated a significant effect of the contemplative part of this meditative practice, despite any anticipated placebo effect.

Studies on the effects of "Shinrin-yoku" on the immune function showed that visiting a forest can induce a significant increase in the number and activity of natural killer (NK) cells, and follow-up data demonstrated that the improved NK cell activity lasted for more than 30 days after intervention (Li et al. 2007; Li 2010). Moreover, forest bathing was associated with significantly higher granulysin-, perforin-, and granzyme A/B–expressing lymphocytes in the blood, and a significantly lower concentration of adrenaline in the urine (Li 2010). On the other hand, visiting an urban area did not increase the number and activity of NK cells, nor the level of intracellular granulysin, perforin, and granzymes A/B (Li 2010). Similar results were obtained with both male and female study participants, regardless of their gender (Li et al. 2008a; Li et al. 2010). The authors concluded that phytoncides (volatile antimicrobial substances released by plants) like alpha- and beta-pinene detected in the forest air might have partially contributed to the effects of forest bathing on the immune function (Li et al. 2006; Li et al. 2008a; Li et al. 2008b; Li et al. 2010). In a controlled experiment with healthy female volunteers, it was demonstrated that smelling air saturated with alpha-pinene could induce a physiological relaxation, and study participants reported a significant increase of the overall mean high-frequency component of heart rate variability during olfactory stimulation (associated with parasympathetic nervous activity) and a significant decrease of their heart rate (Ikei et al. 2016). In a study involving young students, the specific forest composition combined with characteristics of the study sample also showed different effects on perceived anxiety alleviation, with a significant stress relief activity of birch (*Betula platyphylla Suk.*) forests on students of both genders, a more pronounced activity of oak (*Ouereus mongolica Fisch. ex Ledeb*) forests on females, and a limited effect of maple (*Ater triflorum Komarov*) forests on all participants (Guan et al. 2017). In a study by Nam and Uhm (2008) involving 60 participants, inhalation of phytoncides was associated with a significant decrease in serum cortisol levels. Inhaled compounds were obtained from conifers like pines (*Pinus sylvestris*) and cypresses (*Cupressus sempervirens*) (Nam and Uhm, 2008). However, in another trial conducted by Kondo et al. (2011), serum cortisol levels of study participants significantly decreased after forest bathing in sub-groups wearing smell-blocking masks or no mask, while they did not significantly variate in the sub-group wearing visual-blocking masks. These data, although limited, seem to indicate that watching the forest may influence cortisol levels more than breathing its air. Overall, evidence from all these studies suggests that phytoncides can possibly have a role in the effects of forest bathing on stress relief and relaxation, although their specific effect on cortisol levels deserves to be further investigated.

In general, "Shinrin-yoku" as a whole appears as a beneficial practice with the potential to decrease stress levels. Although phytoncides and other volatile compounds can play a role in the overall effects of "Shinrin-yoku," watching the forest and contemplating the natural environment seem the most important components capable of significantly influencing cortisol levels. This seems to be in line with the hypothesis of the role of visualization in triggering the above discussed cortisol-lowering placebo effect. In fact, cortisol levels were measured immediately before forest bathing, in the natural environment, and results underscore the importance of simply visualizing the forest in influencing these hormone levels. Moreover, the main results of the quantitative synthesis showed that salivary cortisol levels were significantly lower in the forest bathing groups compared with the urban groups before intervention, with a moderate-to-low level of heterogeneity across studies found in this analysis ($I^2 = 46\%$; Fig. 2). This provides further confirmation to the hypothesis that, possibly, the anticipated placebo effect could be one of the most determinant components in obtaining a significant decrease in cortisol levels. It is possible that the activity of forest bathing on cortisol levels may be due to psycho-biological phenomena related to the effects on the individual of the forest archetype or, more in general, of the visualization of nature scenes. In fact, it has been observed that nature scenes can determine a higher esthetic judgment compared with scenes with human subjects (Di Dio et al. 2016), and this greater esthetic experience may suggest that, during the mental visualization of a forest, the hypothalamus-hypophysis-adrenal axis can be influenced, which would deserve further investigations. Furthermore, participants of both groups possibly planned in advance and visualized the activity before intervention or comparison, but cortisol levels were significantly lower in the intervention (forest) group, thus suggesting that also the content (forest or urban) of the possible visualization and planning can have an impact on the studied outcome. Given that a pilot study has already demonstrated that a virtual reality forest can facilitate the recovery from stress (Annerstedt et al. 2013), digital computer-generated environments could be created and used to harness the abovementioned beneficial effects even on patients who cannot physically reach a real forest because of individual disabilities or other health problems. Further studies would be desirable to better understand how these restorative natural environments can be sustainably exploited in future healthcare strategies (Depledge et al. 2011).

Limitations

Included studies were heterogeneous in terms of characteristics of study population and intervention (walking in a forest, simply watching it, or both, even combined with other treatments such as psychotherapy). Study designs were various, and some trials lacked randomization or control. Most studies involved a very limited number of participants, with an extreme sex ratio (e.g., all males) and belonging to similar age ranges, thus resulting in possibly biased results characterized by an underestimated variance. No study involved participants belonging to particular categories such as pregnant women or very old people (aged 80+). The overall quality of included studies evaluated with the NIH tools was "good" (highest quality on 3-point rating scale) only for five (5) studies (Kobayashi et al. 2017; Lee et al. 2009; Mao et al. 2012b; Tyrväinen et al. 2014; Yu et al. 2016), while the others scored less. Several included trials lacked any information about the methods of randomization and blinding assignment to intervention or control/comparison. Moreover, studies conducted and published before 2009 did not report enough data to precisely estimate dropout rates. Finally, given that limited evidence exists on the topic and that symmetry was not found in all funnel plots (Fig. 4), publication bias cannot be excluded.

Conclusions

In conclusion, our findings suggest that forest bathing has the potential to significantly influence cortisol levels on a short term in such a way as to reduce stress and anticipated placebo effects can play an important role in it. Due to limited available data, further research is advised to better understand the effects of forest bathing on stress hormones.

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