EXPERT REVIEWS

Cryotherapy in inflammatory rheumatic diseases: a systematic review

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The aim of this article was to review current evidence about cryotherapy in inflammatory rheumatic diseases (therapeutic and biological effects). For therapeutic effects, we performed a systematic review (PubMed, EMBASE, Cochrane Library, LILACS databases, unpublished data) and selected studies including non-operated and non-infected arthritic patients treated with local cryotherapy or whole-body cryotherapy. By pooling 6 studies including 257 rheumatoid arthritis (RA) patients, we showed a significant decrease in pain visual analogic scale (mm) and 28-joint disease activity score after chronic cryotherapy in RA patients. For molecular pathways, local cryotherapy induces an intrajoint temperature decrease, which might downregulate several mediators involved in joint inflammation and destruction (cytokines, cartilage-degrading enzymes, proangiogenic factors), but studies in RA are rare. Cryotherapy should be included in RA therapeutic strategies as an adjunct therapy, with potential corticosteroid and nonsteroidal anti-inflammatory drug dose-sparing effects. However, techniques and protocols should be more precisely defined in randomized controlled trials with stronger methodology.

Keywords: cryotherapy • cytokines • DAS28 • enzymes • pain VAS

Inflammatory joint diseases, such as rheumatoid arthritis (RA), represent a major public health concern, with both synovial inflammation causing joint destruction, pain and disability [1] and systemic inflammation thought to increase cardiovascular risk and mortality [2,3]. Recently, progress in immunology provided new therapeutic targets and new drugs such as biologic agents allowing to achieve clinical remission and prevent joint destruction [4,5]. These treatments, however, remain expensive, with rare but potentially life-threatening side effects such as infections [6]. Long-term nonsteroidal anti-inflammatory drugs (NSAIDs) and corticosteroids also have a well-known toxicity [7]. So the development of adjunct therapies in order to spare biologic and corticosteroid doses is a key focus in these diseases.

Cryotherapy is used empirically in a wide range of rheumatic diseases as a symptomatic treatment, with well-known analgesic, antiphlogistic, myorelaxing, vasoconstrictive, anti-inflammatory, enzyme-blocking and antioxidative effects [8–10]. It can be used not only in inflammatory joint diseases (such as crystal-induced arthritis, spondyloarthritis and RA [10]) but also in such painful rheumatic condition as osteoarthritis [8,9], fibromyalgia [11], shoulder capsulitis [12] and muscle damage [13]. Whole-body cryotherapy (WBC) also showed effects on bone biomarkers [14]. This adjunct treatment is cheap (at least for local cryotherapy [LC]) and generally well tolerated [15,16]. Technical modalities (local/general application, duration, number of sessions [17], and physical form) are very diverse and lack standardization [18]. This widespread use contrasts with a poor level of evidence [18]. Cryotherapy has been shown to decrease intra-articular temperature (T°C) in human knees to 30°C [19,20]. This intrajoint temperature is in the same range as therapeutic mild hypothermia used in several other medical fields. Mild hypothermia (28–34°C) has shown anti-inflammatory effects in healthy subjects [21,22] and in very diverse pathologies such as cerebral ischemia in humans [23] and murine models [23,24], traumatic tissue injury in murine models [25,26] and humans [27], hemorragic shock in rats [28], cardiac arrest in humans [29], coma in pigs [30],

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coronary artery or cardiopulmonary bypass in humans [31], aortic ischemia/reperfusion in mice [32], mechanical ventilation in rats [33,34], postexercise hyperthermia in humans [35], age-related macular degeneration in culture experiments using a retinal cell line [36] and pancreatitis in rats [37]. These studies showed potential effects on important molecular/cellular mechanisms involved in synovial inflammation and joint destruction such as proinflammatory cytokines [21,25,38], VEGF [36], enzymatic pathways (metalloproteinases [39,40], collagenase [41]), adhesion molecules (ICAM-1) and white blood cell infiltrate formation in rats [42] and humans [43], oxidative stress in rats [24] and humans [44], norepinephrine in humans [24,45,46]. This suggests potential therapeutic effects in inflammatory rheumatic diseases such as RA, as some of these molecular pathways are known to be related to pain, disease activity scores including 28-joint disease activity score (DAS28), biological inflammation and radiologic joint damage.

The aim of this article is to review data and evidence concerning cryotherapy's effects in inflammatory rheumatic diseases.

First, we performed a systematic review of the literature about cryotherapy's therapeutic effects in rheumatic inflammatory joint diseases. The primary endpoints were pain assessed by visual analogic scale (VAS) and DAS28. The secondary endpoints were tolerance and molecular anti-inflammatory effects of cryotherapy in these diseases.

Cryotherapy effects on pain & disease activity in rheumatoid arthritis: systematic review

Methods

We followed the PRISMA statement checklist for meta-analysis and systematic review quality criteria [47].

Searching

We used PubMed, EMBASE, LILACS and Cochrane library databases. Keywords 'cryotherapy,' 'cryotherapy arthritis,' 'cryotherapy inflammation,' 'cold,' 'cold arthritis,' 'cryostimulation' and 'WBC' were used alone and in combination. We considered articles with available abstracts in English, German, Spanish, French language and in referenced journals from their inception to March 2013. We also manually screened references cited in the selected articles, considered abstracts from rheumatology congresses (ACR, EULAR since 2001).

As concerns unpublished data, we considered the International Standard Randomised Controlled Trial Number Register [101], The National Institute of Health [102] and the WHO [103]. The screening was performed by two independent reviewers with discussion when needed in order to reach consensus.

Eligibility & study selection

Selection criteria for cryotherapy therapeutic effect evaluation were studies including inflammatory rheumatic disease patients (i.e., RA, microcrystals, peripheral spondyloarthritis) treated with LC or WBC, with endpoints evaluating pain and joint disease activity (pain VAS, erythrocyte sedimentation rate, C-reactive protein, DAS28 and Doppler activity). Articles about postoperative joint cryotherapy and infectious diseases were excluded. We selected original articles, abstracts, reviews and meta-analyses. Duplicates were removed.

Quality assessment

For cryotherapy therapeutic effects, we analyzed technical cryotherapy modalities in detail (physical form and device, duration, skin or joint temperature).

The methodology was also evaluated: study population, randomization, blinding, control groups (other therapeutic modalities such as pharmacological treatments, physical therapy, different cryotherapy techniques or placebo groups), withdrawal and dropout reporting, as well as potential confounders (corticosteroids, NSAIDs, biologics, physical exercise, kinesitherapy, BMI, considered joint) when assessed in the studies. Data extraction was performed by two independent reviewers.

Therefore, we assessed study quality based on specific validated scores depending on the study design. A JADAD 5-point scale was used for randomized controlled trials [48]. For non-randomized studies, we used the Newcastle–Ottawa Scale (NOS) system (0–9) [104]. Furthermore, a JADAD 11-point scale [48] was applied to all the selected studies whatever their design in order to compare them globally and to provide a general qualitative overview. Studies that scored six or higher using JADAD 11-point scale (3/5 with JADAD-5 and 5/9 with NOS) were considered to be of higher quality. This quality assessment was also performed by two independent reviewers.

Quantitative data synthesis

Most outcomes were continuous in nature (pain VAS, DAS28). When pooling data from different trials was possible, the principal measures of effect were means ± SD (weighted mean differences before/after cryotherapy or relative to control groups when possible). Heterogeneity was assessed graphically with 95% confidence intervals and statistically tested using Fisher's variance comparison tests. Heterogeneity threshold was calculated for each primary outcome ($F = 2.73$ or greater was significantly heterogeneous for pain VAS in patients treated with LC, $F = 2.7$ for pain VAS in WBC-treated patients, $F = 2.73$ for DAS28 in LC-treated patients and $F = 3.12$ in WBC-treated patients). We used a fixed effect model. Pooled means ± SD were compared before/after cryotherapy (within-group effect size; paired t tests; α risk 5%) and the mean differences before/after treatment were compared between cryotherapy-treated patients and control groups when available (between-group effect size; unpaired t tests; $\alpha = 5\%$; variance comparison using Fisher's test).

Data were analyzed using Statview© (SAS Institute Inc. Version 5.0) device. There were no *a priori* sensitivity and subgroup analyses. We also considered unpublished data in order to minimize publication bias.

Results

Flowchart

The Flowchart is shown in Figure 1.

Screening results

PubMed search (in English with abstracts) displayed 11,344 citations for 'cryotherapy' keyword on 4 March 2013. The keywords 'cryotherapy arthritis', 'cryotherapy inflammation', 'WBC', 'cold', 'cold arthritis', 'cold inflammation' and 'cryostimulation' showed 67, 346, 331, 108, 707, 733, 4355 and 31 results, respectively.

EMBASE database displayed 23,228 citations for 'cryotherapy' keyword, 22,632 results for 'cold', 1784 results for 'cold arthritis', 445 results for 'cryotherapy arthritis', 95 results for 'WBC', 17,445 results for 'cold inflammation', 3402 results for 'cryotherapy inflammation' and 32 results for 'cryostimulation'.

The LILACS database displayed 230 results for 'cryotherapy' keyword, 1 for 'cryotherapy arthritis', 7 for 'cryotherapy inflammation', 52 for 'WBC', 1479 results for 'cold' keyword, 6 for 'cold arthritis', 19 for 'cold inflammation' and 34 results for 'cryostimulation'.

In EULAR congress abstracts, we found 17 and 132 abstracts related to 'cryotherapy' and 'cold', respectively, since 2001 on EULAR website (ACR website: 21 abstracts related to 'cold' in 2006–2011, none related to 'cryotherapy').

The International Standard Randomised Controlled Trial Number Register website displayed 15 results for 'cryotherapy' keyword and 99 results for 'cold' keyword. The National Institutes of Health website displayed 188 results for 'cryotherapy' keyword (4 for 'cryotherapy arthritis', 5 for 'WBC') and 645 results for 'cold' keyword (16 for 'cold arthritis'). The WHO website displayed 1830 results for 'cryotherapy' keyword (56 for 'cryotherapy arthritis', 22 for 'whole body cryotherapy') and 3,020 results for 'cold' keyword (949 for 'cold arthritis').

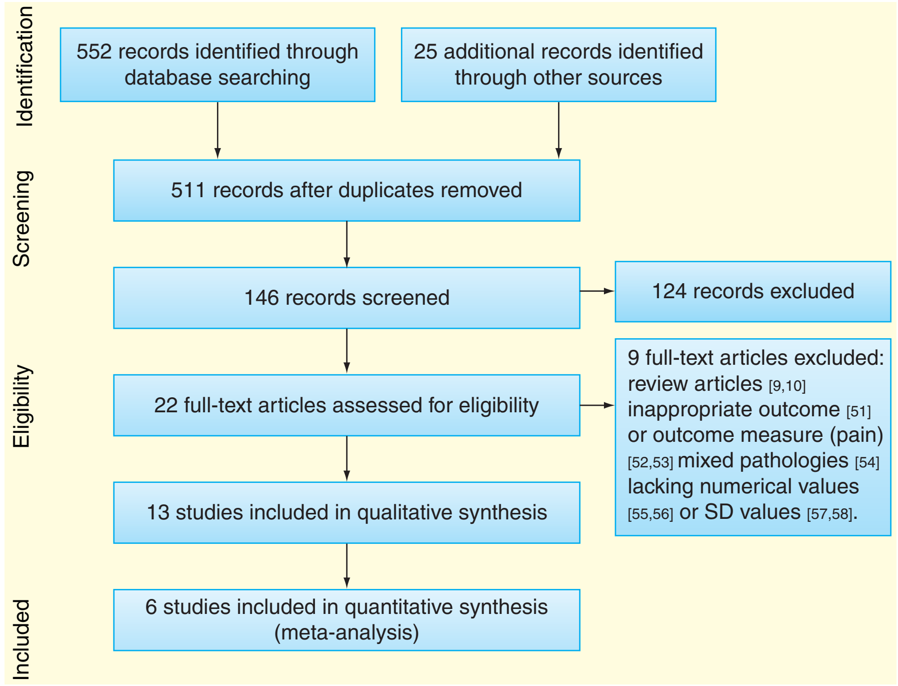

Figure 1. Study selection process.
DAS28: 28-joint Disease Activity Score (composite score including patient VAS for disease activity, acute-phase reactant (erythrocyte sedimentation rate or C-reactive protein), tender joint count and swollen joint score); LC: Local cryotherapy; RCT: Randomized controlled trial; SD: Standard deviation; VAS: Visual Analogic Scale; WBC: Whole-body cryotherapy.

Article selection process

First, articles were excluded on the basis of title and abstract: numerous records dealing with completely different scientific or medical fields such as dermatology, gynecology, urology, oncology, ophthalmology, infectious or lung diseases, chemistry, etc., very low temperature cell lysing-cryotherapy, local cryotherapy not applied to joints spine, etc. After duplicate removal, we found 511 records potentially dealing with cryotherapy in all types of joint diseases according to the titles and abstracts. After applying eligibility criteria, we screened 146 potentially relevant references in the field of therapeutic cryotherapy in inflammatory joint diseases.

Then, we excluded 124 articles for the following reasons: duplicates, articles related to postoperative cryotherapy, noninflammatory diseases, nonrheumatologic diseases, with inadequate outcomes or endpoints, lacking accuracy in cryotherapy technical description or numerical data reporting, with full text not available and insufficient data in the abstract.

A Cochrane meta-analysis including five RCT about cryotherapy in RA [49–53] was published in 2001 and updated in 2011 [18]. None of these articles were appropriate to be used in our meta-analysis, as summarized in Table 1. Two of the studies were performed in operated patients [49,50]. The outcomes [46,47] or outcome measures were inappropriate to our analysis [49,52,53]. Cold application was also probably insufficient in intensity [49,50], in duration [51] or periodicity [51–53]. Furthermore, hot packs used in three of the studies [50,52,53] could have proinflammatory properties [20] and therefore do not seem to be relevant treatments for control group.

The remaining articles (22 articles including 8 RCTs for therapeutic effects) were assessed for further evaluation on the basis of full-text article when available. Nine articles were further excluded [9,10,51–58].

The 13 remaining studies were potentially appropriate to be included in a meta-analysis. There were five RCTs, two nonrandomized controlled studies, three studies comparing several cryotherapy techniques in parallel treatment arms and three noncontrolled studies. Seven articles dealt with local cryotherapy [8,59–64], four with WBC [65–68] and two with both (Figure 1) [69,70].

Characteristics of the studies selected for quantitative analysis (cryotherapy therapeutic effects)

Study characteristics and quality assessment results are summarized in Table 2.

The endpoints were compared before and after cryotherapy.

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Table 1. Cochrane meta-analysis: review of the five randomized controlled trials about cryotherapy.

Pathology	Joint	Endpoints	Postoperative cryotherapy (yes/no)	JADAD score (/5)	Cryotherapy modality	Control group	Reason for exclusion	Ref.
24 RA (ARA criteria)	Knees	Joint circumference Infrared thermography	No	2/5 R1B0W1	Crushed ice in damp towels (10 min daily for 10 days)	Controlateral joint (no cryotherapy)	Endpoints Insufficient cold exposure (duration)?	[51]
5 RA; 83 OA	Knees	Pain (PCA use)	Yes	3/5 R2B0W1	Thermal pad (50°F vs 60°F vs 70°F); duration? periodicity?	None	Postoperative cryotherapy Insufficient cold exposure (temperature)?	[49]
14 chronic RA (definite or classic RA)	20 knees	Pain (none = 0–5 = severe) assessed by two observers at the same time Stiffness, range of movement, knee circumference, skin temperature, patient preference	No	1/5 R1B0W0	Ice packs in damp towels (20 min, once a day for 10 days)	Hot packs (cross-over)	Pain assessment	[52]
Patients hospitalized for surgical procedures to the hand	30 hands	Edema evolution over preoperative volume	Yes	2/5 (R1B0W1)	Cold water immersion (10°C for 4 min; twice a day for 1 day)	Hot packs (n = 15)	Postoperative Endpoints Insufficient cold exposure (temperature and duration)?	[50]
18 recent RA (<5 years)	Shoulders	Pain (Mc Gill questionnaire) Range of movement	No	1/5 (R1B0W0)	Ice (20 min) + exercises program	Hot packs (n = 9)	Pain assessment	[53]

This meta-analysis performed in 2001 and updated in 2011 mixed studies with cold or heat application. It showed no significant effect on pain (primary endpoint), joint swelling, medication intake, range of motion, grip strength or hand function. No harmful side effect was reported [18]. The five RCTs about cryotherapy had limitations: the studies showed a great heterogeneity as concerns cryotherapy methods, treated joints, outcomes, associated medications and physical exercise. The control groups were: hot packs in three studies and contralateral joint in one study. Heat application does not seem to be an appropriate control nor treatment group because it could increase joint inflammation and collagenase activity [20]. Cold exposure was probably insufficient in intensity and duration in some of the studies compared with more recent studies.
ARA: American Rheumatism Association; B: Blinding; OA: Osteoarthritis; PCA: Patient-controlled analgesia; R: Randomization; RA: Rheumatoid arthritis; W: Withdrawals (JADAD score).
Data taken from the articles cited in this table.

Table 2. Therapeutic effects of cryotherapy: articles included in the meta-analysis (n = 6).

Pathology/ joints (n)	LC/WBC (n)	Cryotherapy modalities	Control group (n)	Relevant endpoints (for meta-analysis) and evaluation times	JADAD 5/ NOS JADAD11	Bias/confounders	Ref.
RA (60 patients)	LC (n = 20) OR WBC (-60°C; n = 20) WBC (-110°C; n = 20)	Cold packs or cold air on 5 joints (-30°C; 10–30 or 1–5 min) OR WBC (-60°C OR -110°C; duration?) → Three-times a day; 7 days (20 applications)	None	Pain VAS DAS28 ESR CRP → Before; Day 7 'after the last cryotherapy' n = 20; 17; 17	R1B0W1 8/11	Associated kinesitherapy Corticosteroids (10/20; 14/20; 9/20); median dose 5 mg/day [2,5–15] NSAIDs: 16/20; 17/20; 18/20 DMARDs: 10/20; 9/20; 9/20 'cytostatics': 11/20; 14/20; 12/20 No change in pharmalogical treatment. BMI: 25.7 ± 4 vs 24.6 ± 3.6 vs 28.3 ± 5.9 Biologics, physical exercise, skin/ room T°C: NA	[70]
RA (ACR; n = 40 patients)	LC (2 modalities)	Cold air (-30°C; 3 min; n = 20) OR Liquid nitrogen vapors (-160°C; 3 min; n = 20) → Twice a day (knees in the morning, 4 h break, then hands) for 10 days	None	Pain VAS DAS28 → Before and after 10 days of treatment	S3C1O2 6/11	Associated kinesitherapy and physical exercise Corticosteroids 28/40 DMARDs 40/40 Biologics: none No change in pharmalogical treatment. BMI: 28.4 ± 4.5 and 28.2 ± 2.3 NSAIDs, skin/room T°C	[63]
Early RA (n = 36 patients)	LC (n = 20 patients)	Cold air (-60°C; 15 min; 10 sessions; hands, knees or ankles) Included in a Complex Rehabilitation Program (40 min exercise, 40 min occupational therapy + 'Drug therapy'). Total duration?	'Drug therapy' only (n = 16)	Pain VAS DAS28 → Before and after treatment (10 days?)	S3C1O1 3/11	Corticosteroids, NSAIDs, DMARDs, biologics, kinesitherapy, skin/room T°C, BMI: NA	[64]
RA (n = 48 patients), AS (n = 12)	WBC	WBC (-110°C for 3 min; twice a day) → Average number of sessions: 15.8 ± 8,37	None	Pain VAS DAS 28 (48 patients) BASDAI (12 patients) → Before and after treatment	S2C0O1 4/11	Associated kinesitherapy and physical exercise. No change in pharmalogical treatment. Corticosteroids, NSAIDs, kinesitherapy, physical exercise, skin temperature, BMI: NA	[66]

ACR: American College of Rheumatology (Diagnostic criteria for rheumatoid arthritis); B: Blinding; BASDAI: Bath Ankylosing spondylitis Disease Activity Index; C: Control groups; CRP: C-reactive protein; DMARD: Disease activity-modifying drug; ESR: Erythrocyte sedimentation rate; LC: Local Cryotherapy; NA: Not assessed; NSAID: Nonsteroidal anti-inflammatory drug; O: Outcome measurement (NOS score); Pain VAS: pain Visual Analogic; RA: Rheumatoid arthritis; RCTs: R: Randomization; S: Sampling; Scale DAS28: 28 joint-disease activity score (composite score including patient VAS for disease activity, acute-phase reactant (ESR or CRP), tender joint count and swollen joint score); T: Temperature; W: Withdrawals (JADAD score); WBC: Whole-body cryotherapy
Data taken from the cited articles.

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Table 2. Therapeutic effects of cryotherapy: articles included in the meta-analysis (n = 6) (cont.).

Pathology/joints (n)	LC/WBC (n)	Cryotherapy modalities	Control group (n)	Relevant endpoints (for meta-analysis) and evaluation times	JADAD 5/NOS JADAD11	Bias/confounders	Ref.
RA (ACR; n = 32 patients)	WBC (n = 15 patients)	WBC (-110°C for 3 min; once a day) + kinesitherapy → 'Complex therapy' for 8 days	Low frequency magnetic field (20–40 Hz; 5–7 mT; 20 min; n = 17 patients) + kinesitherapy	Pain VAS DAS28 → Before and after treatment (8 days)	S4C1O1 5/11	Associated kinesitherapy No change in pharmalogical treatment. Corticosteroids, NSAIDs, physical exercise, skin temperature, BMI: NA	[67]
RA (ACR; n = 41 patients)	WBC	WBC (-160°C; 3–5 min; twice a day (6 h interval) for 15 days) + active exercises (45 min)	None	Pain VAS → Before and after treatment (15 days)	S3C1O1 3/11	Associated kinesitherapy and physical exercise No change in pharmalogical treatment. Corticosteroids, NSAIDs, DMARDs, biologics, skin temperature, BMI: NA	[68]

ACR: American College of Rheumatology (Diagnostic criteria for rheumatoid arthritis); B: Blinding; BASDAI: Bath Ankylosing spondylitis Disease Activity Index; C: Control groups; CRP: C-reactive protein; DMARD: Disease activity-modifying drug; ESR: Erythrocyte sedimentation rate; LC: Local Cryotherapy; NA: Not assessed; NSAID: Nonsteroidal anti-inflammatory drug; O: Outcome measurement (NOS score); Pain VAS: pain Visual Analogic; RA: Rheumatoid arthritis; RCTs: R: Randomization; S: Sampling; Scale DAS28: 28 joint-disease activity score (composite score including patient VAS for disease activity, acute-phase reactant (ESR or CRP), tender joint count and swollen joint score); T: Temperature; W: Withdrawals (JADAD score); WBC: Whole-body cryotherapy
Data taken from the cited articles.

Cryotherapy modalities (technique, temperature, duration and periodicity) were heterogeneous. The main potential confounders were assessed when possible: Corticosteroid, NSAID, disease activity-modifying drug (DMARD), biologic intake, kinesitherapy, physical exercise, room temperature. No serious adverse event was reported in any of the studies.

We could only perform a pooled quantitative analysis for two endpoints: pain VAS and DAS28 in RA patients. For that purpose, six studies were included in the quantitative data analysis (Table 2) [64,65,67–69,71]. Reasons for excluding the seven other studies [8,59–62,65,69] were: one duplicate [69], impossibility to combine data for power Doppler hypersignal endpoint due to different evaluation scores [59–61], as for gout patients: too different designs [8,62], one study mixed patients suffering from heterogeneous rheumatic diseases (inflammatory as well as noninflammatory) [65]. Straub and Hirvonen's studies were considered as duplicates as they were performed, at least partly, in the same patient cohort [69,70]. For WBC, we only considered -110°C-treated patients in Hirvonen's study [70].

Results: study quality assessment

Six studies including 257 RA patients were appropriate to be included in quantitative data synthesis. There was one RCT with 40 patients meeting the inclusion criteria [70], two controlled trials [63,64], two studies comparing parallel cryotherapy treatment groups [67,68] and one noncontrolled study [66].

The RCT scored 2 out of 5 (JADAD5 score) [70]. As for the five noncontrolled studies, the mean NOS quality score was 5 ± 1.2 [63,64,66–68]. Overall, the mean JADAD11 score for the six selected studies was 4.8 ± 1.9. The quality scores for each study are displayed in Table 2.

Results: heterogeneity assessment

We could only perform quantitative analysis for two endpoints: pain VAS (mm) and DAS28 in RA patients after chronic application (7–15 days) (Figures 2 & 3) [63,64,66–68,70].

There was no significant heterogeneity between studies for pain VAS and DAS28 in LC or WBC-treated patients, as shown in Figures 2 & 3, displaying means and 95% confidence intervals. Fisher's tests showed F0 = 1.48; p: [0.2; 0.3] for pain VAS after local cryotherapy (Figure 2A), F0 = 1.44; p: [0.2; 0.3] for DAS28 after local cryotherapy (Figure 2B), F0 = 1.07; p: [0.3; 0.5] for pain VAS after WBC (Figure 3A), F0 = 0.47; p: [0.5; 0.9] for DAS28 after WBC (Figure 3B).

Paired t-tests were used to assess pain VAS and DAS28 evolution after cryotherapy.

As concerns local cryotherapy, the mean number of cold applications was 17.1 (ranging from 10 to 20), mean temperature of -70.3°C (-30 to -160) applied for 11.5 min (3–30).

As for WBC and pain VAS (mm), the mean number of applications was 20.2 (8–30) at a mean temperature of -126.5°C (-110 to -160) during 3.2 min (2–5).

Considering WBC and DAS28, the mean number of applications was 14.4 (8–20) at a temperature of -110°C during 2.8 min (2–3).

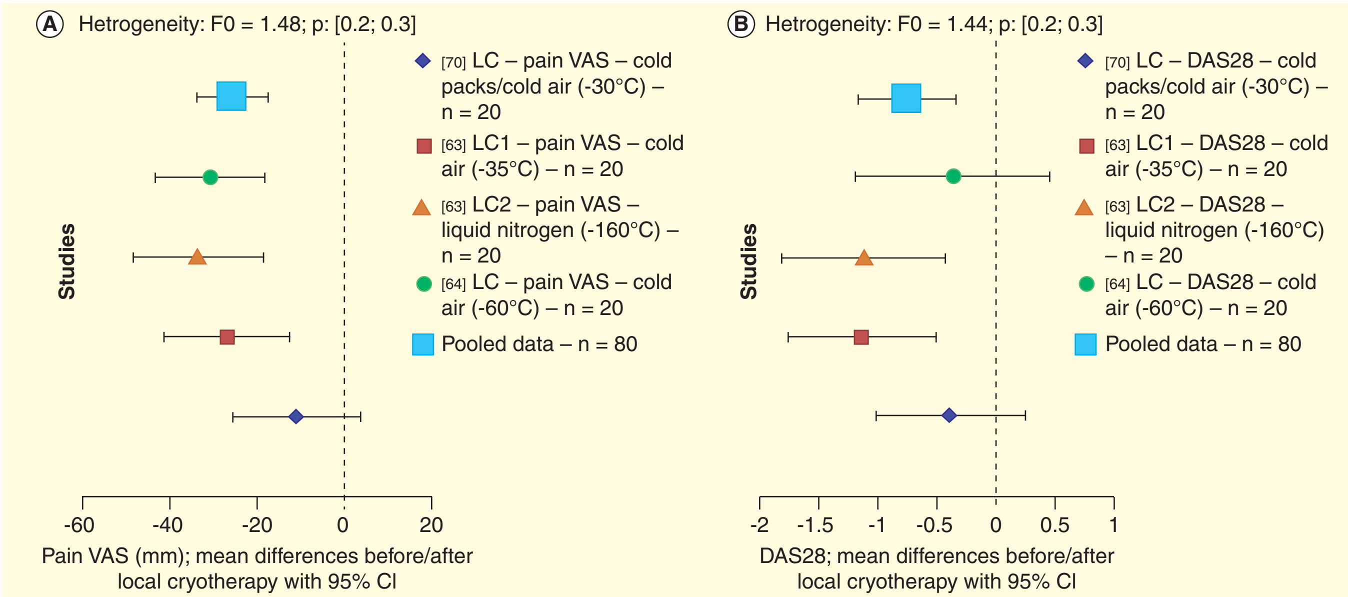

Figure 2. Effects of local cryotherapy on pain. VAS **(A)** and DAS28 **(B).**
DAS28: 28-joint disease Activity Score (composite score including patient VAS for disease activity, acute-phase reactant (erythrocyte sedimentation rate or C-reactive protein), tender joint count and swollen joint score); LC: Local cryotherapy; VAS: Visual Analogue Scale. Data taken from the cited articles.

Results: primary outcomes (pain VAS & DAS28)

As concerns pain, LC (cold packs, cold air, liquid nitrogen applied on 1–5 joints) significantly reduced pain VAS (mm) in 80 RA patients originating from three studies [63,64,70], with 20 of these patients were included in a RCT [70]. Mean pain VAS decreased from 59.10 ± 25.86 (95% CI: 42.17–75.63) to 33.55 ± 20.77 (95% CI: 26.07–56.33) after LC ($p < 0.000002$). WBC also significantly decreased pain VAS in 124 RA patients from 4 studies [66–68,70] (20 patients originated from a RCT [66]. Mean pain VAS decreased from

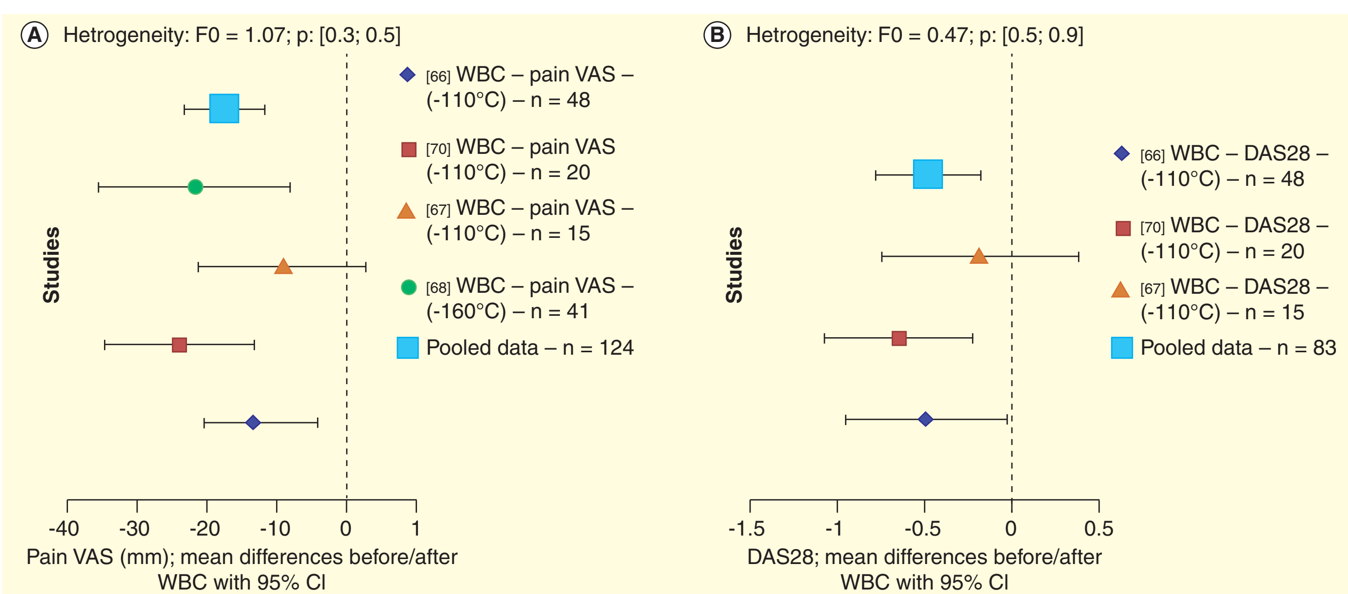

Figure 3. Effects of whole-body cryotherapy on pain. VAS **(A)** and DAS28 **(B)**. Mean differences in pain VAS (mm) or DAS28 before/after LC or WBC are represented for each of the six studies included in the meta-analysis [63,64,66–68,70], with 95% confidence intervals. Heterogeneity was also tested using Fisher's test (F0 and p-values are shown on the graphs). Design of the studies: randomized controlled trial [70], controlled trials [63,64], parallel cryotherapy treatment groups [67,68] and noncontrolled study [66].
DAS28: 28-joint disease Activity Score (composite score including patient VAS for disease activity, acute-phase reactant (erythrocyte sedimentation rate or C-reactive protein), tender joint count and swollen joint score); LC: Local cryotherapy; VAS: Visual Analogue Scale; WBC: Whole-body cryotherapy.

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Table 3. Local cryotherapy techniques.

Local cryotherapy	Physical form	Temperature	Pressure	Duration	Skin temperature	Ref.
Ice bags	Ice cubes, mixture of water and crushed Fee	0°C	Straps for compression	10–30 min 30 min	13–15°C in 15–30 min 1 G°C (minimal value)	[72] [20]
Cold packs prerefrigerated gels	Joint-shaped, flexibility (CryoCuff®; Polar Care®) Gel-filled cold pack (TMP Tiishaus® 12X29 cm	-15°C	+	10–30 min; three-times a day for 7 days 20 min; 5/day 20 min	 22–24°C 5, 5°C	[69] [82] [58,59]
Gas (thermal shock)	Cold air (filtered ambient air: no consumables) Cryo 5®: 40001/min	-30°C -20 to -30°C	0	5 min 10–30 min; 3/day; 7 days 3 min 3 min	9.7°C in 5 min (data not available) 23.1°C after 1 min 6°C	[19] [68] [15] [58]
	Liquid Nitrogen vapors (Medivent®)	-160°C	0	6.5 min 3 min	9.8°C (minimal value) 17,9°C after 1 min	[20] [15]
	CO_2 microcristals (Cryotron®)	-78°C	50 bars (2–75 bars)	45 s–2 min (2/day); flare duration 90s (3/day)	7.3°C 2°C in 20–30S 12°C	[73] [8]

53.15 ± 20.45 (95% CI: 49.55–56.75) at baseline to 35.64 ± 26.69 mm (95% CI: 30.94–40.34) after WBC ($p < 0.000002$).

As regards disease activity, LC significantly reduced DAS28 in 80 RA patients from 3 studies [63,64,70], with 20 patients included in a RCT [70]. Mean DAS28 decreased from 5.45 ± 1.37 (95% CI: 5.14–5.75) at baseline to 4.69 ± 1.16 (95% CI: 4.44–4.95) after LC ($p < 0.0001$), which could suggest a systemic effect of LC that was applied on several joints (4–6) in these patients. WBC also significantly reduced DAS28 in 83 RA patients from 3 studies [66,67,70], including 20 patients originating from a RCT [70]. Mean DAS28 decreased from 4.27 ± 0.83 (95% CI: 4.02–4.52) at baseline to 3.79 ± 0.81 (95% CI: 3.56–4.02) after WBC ($p < 0.002$).

Results: secondary outcomes (tolerance & physiological effects)

As concerns tolerance, no major adverse effect was reported in any of the screened studies. Cryotherapy is overall a well-tolerated treatment [8,9] compared with other adjunct therapies in RA such as corticosteroids and NSAIDs. The contraindications are patients with systemic lupus erythematosus, vasculitis, cryoglobulinemia, cold hypersensitivity, allergy or urticarial, cold-induced bronchospasm, Raynaud's phenomenon, acrocyanosis, sickle cell anemia, skin circulation disorders, paroxysmal cold hemoglobinuria, heart arrhythmia, symptomatic cardiovascular or lung disease, uncontrolled hypertension, advanced diabetes mellitus and cutaneous hypoesthesia. It should be avoided in patients with scleroderma, spinal cord injury or poor circulation (risk of skin lesions such as frostbite, chilblains or necrosis). Beyond a certain application duration threshold (for instance, 20–30 min for cold packs, 2 min for CO_2 cryotherapy at -78°C as indicated in manufacturers' instructions for use), cryotherapy can be painful and proinflammatory. Anyway, specific instructions for use should be read carefully before using any cryotherapy device, especially as concerns maximal recommended application duration. During CO_2 cryotherapy, skin temperature must be kept above 2°C, gas blow must be performed at 10–15 cm from skin surface (4–6 cm for cold air) [15], the application area must be swept and ice crystal formation on skin surface must be avoided (frostbite, chilblain and burn prevention). Cold packs must not be in direct contact with the skin. Cryotherapy can also induce nerve lesions (it must be used with caution in the vicinity of superficial nerves) and slow wound healing. The main cryotherapy techniques are summarized in Table 3 (local cryotherapy) and Table 4 (whole-body cryotherapy).

As for cryotherapy, physiological effects in RA, LC may reduce joint temperature to about 30°C in healthy as well as arthritic human knees for 2 h [19,20].

Studies in animal models and other medical fields suggest that mild hypothermia (with local and/or core body temperatures around 30°C) may inhibit white blood cell infiltrate formation [42], proinflammatory cytokine gene transcription [23,30], enzymatic pathways such as collagenases [41], metalloproteinases [39,40], proangiogenic agents such as VEGF [36]. In RA, cryotherapy might decrease proinflammatory cytokine and proteolytic enzyme levels, but studies are rare. LC significantly decreased serum TNF-α and tended to decrease serum IL-6 levels in 40 RA patients [63]. LC and WBC tended to decrease serum IL-6 levels in 59 RA patients [69]. WBC significantly decreased serum histamine levels in 20 RA patients [71]. In experiments using RA synovial collagenase cultured with human collagen fibrils, the authors showed a four-time decreased collagen lysis at 33 versus 36°C [41]. In arthritic zymosan-injected

Table 4. Ice-water and whole-body cryotherapy techniques.

Technique	Physical form	Temperature	Pressure	Duration	Skin temperature	Ref.
Ice-water immersion						
Ice water		0–20°C	+	0–2°C for 20 s (three-times a week for 12 weeks)		[22,45]
Whole-body cryotherapy						
Cryogenic chambers	Dehydrated air (Criostream®) Cold air cooled by liquid nitrogen (Zimmer® KR2005N®)	-60°C to 140°C < (1 or 2 acclimation chambers)	0	2–3 min 2 min (three-times/ week; 12 weeks)	12–16°C (110*0)	[45]
				2 min; three-times a day; 7 days		[68]
		-130°C		3 min/day–10 days	11°C (forearm)	[81]

Data were taken from the cited articles.

rabbits, ice chip application caused a nonsignificant decrease in cell infiltration and synovial hyperplasia [72]. These results hold strong therapeutic promises in RA. However, studies about cryotherapy's molecular effects in RA are scarce and heterogeneous, so we could not perform any quantitative data analysis.

Discussion

Pooling 6 studies including 257 RA patients, we show that chronic local or WBC (14–20 applications) significantly decreases pain VAS (mm) and DAS28 (within-group effect size).

As concerns control groups, 16 patients were treated with 'drug therapy' and compared with LC-treated patients [64] and 17 patients exposed to magnetic fields were compared with WBC-treated patients [67]. These control groups were poorly described, and the studies were not randomized, so we could not perform any comparison with pooled mean differences in cryotherapy-treated patients nor calculate any between-group effect size. We excluded control groups with heat application that has proinflammatory effects [20]. It is of course difficult to create placebo groups for cryotherapy. All the patients in the selected studies received associated pharmacological treatment. This drug therapy intake (NSAIDs, corticosteroids, DMARDs and biologics) was not precisely described in four out of six studies. However, RA treatment is quite standardized and pharmacological treatment protocols (drugs and doses) remained stable before and throughout the studies, so the variations in pain VAS and DAS28 scores are likely to reflect cryotherapy's effects as an adjunct therapy.

We pooled patients treated with different cryotherapy techniques, because group sizes were not sufficient for separate analyzes, and because no significant difference for considered endpoints was found between these techniques in studies using parallel treatment arms. Notably, we could not perform any subgroup analysis comparing cold packs (cooling) to gaseous cryostimulation in LC-treated patients due to insufficient sample sizes [63,70]. Cryotherapy protocols were quite heterogeneous (duration, intensity, considered joints, physical agents, temperature, duration and periodicity) as summarized in Table 2. The overall quality scores of the selected studies were quite low, but they reflect currently available evidence about cryotherapy. Studies were mainly limited by a lack of randomization and valid control groups. It is obviously difficult to find appropriate placebo groups for cryotherapy. Dropouts and withdrawals were also poorly reported. However, as cryotherapy is a very well-tolerated treatment, and as no major side effect was reported in any of the selected studies, the amount of missing data is likely to be very low.

Importantly, despite various cryotherapy modalities and potential confounders, the six selected studies showed very homogeneous results (Figure 2).

Unlike Welsh's Cochrane meta-analysis, we excluded articles dealing with postoperative cryotherapy, as surgery by itself might interfere with joint inflammation (Tables 1 & 2).

Expert commentary & five-year view

Clinical practice and physiological rationale strongly suggest a potential interest of cryotherapy as an adjunct therapy in rheumatic inflammatory diseases.

Cryotherapy applied locally on an inflamed joint allows to reach a 30°C intra-articular temperature plateau, with a possibly 2–3 h remanent local hypothermia [19,20]. Studies conducted in other medical fields suggest that it might therefore downregulate such proangiogenic and proinflammatory pathways as VEGF, proinflammatory cytokines and enzymatic activities involved in synovial microvascular hyperplasia, joint inflammation and destruction (Figure 4).

Synovial and systemic endothelial dysfunction in RA induce pain, joint inflammation and destruction and increased cardiovascular morbidity and mortality. Cryotherapy, by upregulating noradrenalin pathway, could downregulate IL-6 and i-NOS pathways, which are known to be involved in endothelial dysfunction an inflammation [3]. Further studies are needed to establish these molecular effects of cryotherapy specifically in RA. Studies in animal models such as collagen-induced arthritis or adjuvant-induced arthritis will certainly lead to a better description of cryotherapy effects on these promising molecular

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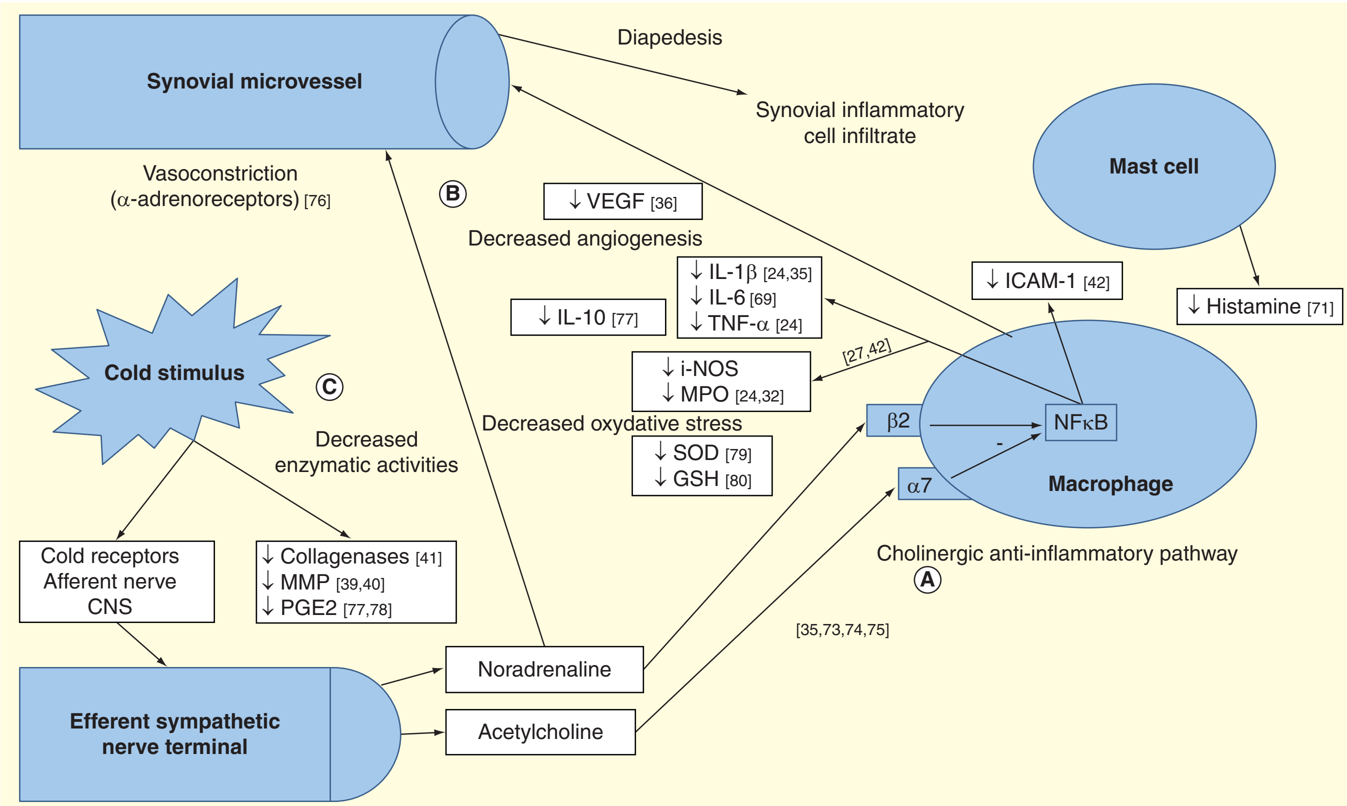

Figure 4. Molecular pathways involved in cryotherapy (proposed model). In rheumatoid arthritis, local and systemic inflammation promote neoangiogenesis which in turn favors inflammatory cell infiltrate and proinflammatory cytokine release. **(A)** After cold stimulation, the autonomic nervous system is activated [73] and efferent sympathetic neurons release acetylcholine that binds a7nAchR receptor and noradrenaline that binds β2-adrenoceptor. These ligand–receptor interactions may then inhibit the NFκB pathway and subsequently downregulate proinflammatory cytokine, oxidative stress agent and adhesion molecule gene transcription [23,35,38,73–75]. **(B)** Noradrenaline also induces vasoconstriction through α-adrenoceptor binding on the vascular wall [76], which could contribute to limit inflammation. Cryotherapy might also downregulate the expression of proangiogenic factors such as VEGF [36]. **(C)** Cryotherapy might also downregulate important enzymatic pathways involved in joint inflammation and destruction [39,59,77,78]. Citations refer to studies conducted in humans [23,35,39,41,69,71,73,76–79], human cell [41] or cell line [36] cultures, rats [24,38,40,42,80], mice [23,32], dogs [76] and two review articles [74,75].
ICAM-1: Intercellular adhesion molecule-1; i-NOS: Inducible NO-synthase; MMP: Metalloproteinase; PGE2: Prostaglandin E2; VAS: Visual Analogic Scale; WBC: Whole-body cryotherapy.
Data taken from the articles cited below and in the figure.

targets in the field of rheumatology, as already the case in neurology for instance, with well-known therapeutic effects of mild hypothermia after brain ischemia [23,24].

We could show a significant decrease in pain VAS (mm) and DAS28 in RA patients after chronic LC as well as WBC (within-group effect size). This result was remarkably constant among the six selected studies (Figure 2). However, we could not calculate any between-group effectsize because available control groups were small and methodologically unsatisfying. Randomized trials with valid control groups and stronger methodology are required in order to measure this effect size more accurately.

In light of the results of this systematic review and considering a solid biological rationale, cryotherapy deserves to be evaluated as a full therapeutic option in patients without any corticosteroid, NSAID, DMARD, biologic or physical therapy.

Short-term cryotherapy effects should also be addressed. LC applied once to an inflamed joint has been shown to decrease synovial power Doppler hypersignal in RA, which is a good reflect of synovial neoangiogenesis and inflammation [60,61]. Our team is currently studying the effects of two local cryotherapy applications on synovial power Doppler hypersignal as well as synovial fluid cytokine and VEGF levels in arthritic patients.

In order to conduct these important studies, a better standardization of cryotherapy techniques will be required (Table 3). Optimal cryotherapy protocols need to be precisely defined (physical agent, temperature and duration periodicity). It is notably important to determine, for each cryotherapy technique, the therapeutic range and the cold intensity threshold beyond which it may become proinflammatory [10,20,59,69]. Gaseous LC might induce a more pronounced and acute decrease in tissue temperature (thermal shock) and cold packs a deeper and more prolonged cooling. WBC is still expensive, but new techniques using filtered and cooled ambient air without any consumable will probably be cheaper and require less room space, allowing a more widespread use.

These studies will help to define cryotherapy's role in treatment strategies in RA and other joint inflammatory diseases, most probably as an adjunct therapy to DMARDs and targeted biologic treatments, along with corticosteroids and NSAIDs. Corticosteroid and NSAID toxicity represent a major public health concern, with numerous, well-known, side effects and complications. Cryotherapy used as an adjuvant therapy and applied using standardized and optimized protocols could help to spare corticosteroid and NSAID doses in these patients, and subsequently decrease cardiovascular, infectious, gastrointestinal morbidity and mortality. This treatment option may be of special interest in an increasing number of patients with NSAID and/or corticosteroid contraindications (cardiovascular diseases, diabetes, kidney deficiency, and so on). This dose-sparing effect should also be addressed and measured specifically in randomized controlled trials.

Local cryotherapy is a cheap and very well-tolerated therapeutic option, which can be easily performed at patient's home. In the future, it could contribute to reduce the economic burden and iatrogenicity related to the treatment of arthritic patients, especially for the elderly.

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Key issues

- Molecular pathways targeted by cryotherapy (proinflammatory cytokines, VEGF, cartilage-degrading enzymes) suggest interesting anti-inflammatory properties in rheumatic inflammatory diseases, which should be further investigated.
- Cryotherapy could be an interesting adjunct therapy in these diseases with a better safety profile as compared with corticosteroids and NSAIDs.
- By pooling six studies, we show that chronic local cryotherapy and WBC significantly reduce pain visual analogic scale and 28-joint disease activity score in rheumatoid arthritis (within-group effect size). However, methodological issues and a lack of control groups prevent from calculating any between-group effect size.

References

Papers of special note have been highlighted as:
• of interest
•• of considerable interest

1 de Punder YM, van Riel PL. Rheumatoid arthritis: understanding joint damage and physical disability in RA. *Nat. Rev. Rheumatol.* 7(5), 260–261 (2011).

2 Semb AG, Kvien TK, DeMicco DA *et al.* Effect of intensive lipid-lowering therapy on cardiovascular outcome in patients with and those without inflammatory joint disease. *Arthritis Rheum.* 64(9), 2836–2846 (2012).

3 Prati C, Berthelot A, Wendling D, Demougeot C. Endothelial dysfunction in rat adjuvant-induced arthritis: up-regulation of the vascular arginase pathway. *Arthritis Rheum.* 63(8), 2309–2317 (2011).

4 Schipper LG, Vermeer M, Kuper HH *et al.* A tight control treatment strategy aiming for remission in early rheumatoid arthritis is more effective than usual care treatment in daily clinical practice: a study of two cohorts in the Dutch Rheumatoid Arthritis Monitoring registry. *Ann. Rheum. Dis.* 71(6), 845–850 (2012).

5 Kavanaugh A, Fleischmann RM, Emery P *et al.* Clinical, functional and radiographic consequences of achieving stable low disease activity and remission with adalimumab plus methotrexate or methotrexate alone in early rheumatoid arthritis: 26-week results from the randomised, controlled OPTIMA study. *Ann. Rheum. Dis.* 72(1), 64–71 (2013).

6 Woodrick RS, Ruderman EM. Safety of biologic therapy in rheumatoid arthritis. *Nat. Rev. Rheumatol.* 7(11), 639–652 (2011).

7 Atzeni F, Turiel M, Caporali R *et al.* The effect of pharmacological therapy on the cardiovascular system of patients with systemic rheumatic diseases. *Autoimmun. Rev.* 9(12), 835–839 (2010).

8 Chatap G, De Sousa A, Giraud K, Vincent JP. Pain in the elderly: prospective study of hyperbaric CO2 cryotherapy (neurocryostimulation). *Joint Bone Spine* 74(6), 617–621 (2007).

9 Demoulin C, Vanderthommen M. Cryotherapy in rheumatic diseases. *Joint Bone Spine* 79(2), 117–118 (2012).

10 Oosterveld FG, Rasker JJ. Treating arthritis with locally applied heat or cold. *Semin. Arthritis Rheum.* 24(2), 82–90 (1994).

11 Bettoni L, Bonomi FG, Zani V *et al.* Effects of 15 consecutive cryotherapy sessions on the clinical output of fibromyalgic patients. *Clin. Rheumatol.* 32(9), 1337–1345 (2013).

12 Ma SY, Je HD, Jeong JH, Kim HY, Kim HD. Effects of whole-body cryotherapy in the management of adhesive capsulitis of the shoulder. *Arch. Phys. Med. Rehabil.* 94(1), 9–16 (2013).

13 Hausswirth C, Louis J, Bieuzen F *et al.* Effects of whole-body cryotherapy vs. far-infrared vs. Passive modalities on recovery from exercise-induced muscle damage in highly-trained runners. *PLoS ONE* 6(12), e27749 (2011).

14 Galliera E, Dogliotti G, Melegati G, Corsi Romanelli MM, Cabitza P, Banfi G. Bone remodelling biomarkers after whole body cryotherapy (WBC) in elite rugby players. *Injury* 44(8), 1117–1121 (2013).

15 Korman P, Straburzynska-Lupa A, Romanowski W, Trafarski A. Temperature changes in rheumatoid hand treated with nitrogen vapors and cold air. *Rheumatol. Int.* 32(10), 2987–2992 (2012).

16 Nadler SF, Weingand K, Kruse RJ. The physiologic basis and clinical applications of cryotherapy and thermotherapy for the pain practitioner. *Pain Physician* 7(3), 395–399 (2004).

doi: 10.1586/1744666X.2014.870036

17 Lubkowska A, Dołęgowska B, Szyguła Z. Whole-body cryostimulation–potential beneficial treatment for improving antioxidant capacity in healthy men–significance of the number of sessions. *PLoS ONE* 7(10), e46352 (2012).

18 Welch V, Brosseau L, Shea B, McGowan J, Wells G, Tugwell P. Thermotherapy for treating rheumatoid arthritis. *Cochrane Database Syst. Rev.* (2), CD002826 (2011).

19 Kim YH, Baek SS, Choi KS, Lee SG, Park SB. The effect of cold air application on intra-articular and skin temperatures in the knee. *Yonsei Med. J.* 43(5), 621–626 (2002).

20 Oosterveld FG, Rasker JJ. Effects of local heat and cold treatment on surface and articular temperature of arthritic knees. *Arthritis Rheum.* 37(11), 1578–1582 (1994).

- **The only study monitoring intrajoint temperature in arthritic patients treated with local cryotherapy. Local cryotheray induces an intrajoint temperature plateau (about 30°C for 2 h).**

21 Lubkowska A, Szygula Z, Chlubek D, Banfi G. The effect of prolonged whole-body cryostimulation treatment with different amounts of sessions on chosen pro- and anti-inflammatory cytokines levels in healthy men. *Scand. J. Clin. Lab. Invest.* 71(5), 419–425 (2011).

22 Dugue B, Leppanen E. Adaptation related to cytokines in man: effects of regular swimming in ice-cold water. *Clin. Physiol.* 20(2), 114–121 (2000).

23 Yenari MA, Han HS. Influence of hypothermia on post-ischemic inflammation: role of nuclear factor kappa B (NFkappaB). *Neurochem. Int.* 49(2), 164–169 (2006).

24 Zhang H, Zhou M, Zhang J, Mei Y, Sun S, Tong E. Therapeutic effect of post-ischemic hypothermia duration on cerebral ischemic injury. *Neurol. Res.* 30(4), 332–336 (2008).

25 Hildebrand F, van Griensven M, Giannoudis P *et al.* Effects of hypothermia and re-warming on the inflammatory response in a murine multiple hit model of trauma. *Cytokine* 31(5), 382–393 (2005).

26 Truettner JS, Suzuki T, Dietrich WD. The effect of therapeutic hypothermia on the expression of inflammatory response genes following moderate traumatic brain injury in the rat. *Brain Res. Mol. Brain Res.* 138(2), 124–134 (2005).

27 Nandate K, Vuylsteke A, Crosbie AE, Messahel S, Oduro-Dominah A, Menon DK. Cerebrovascular cytokine responses during coronary artery bypass surgery: specific production of interleukin-8 and its attenuation by hypothermic cardiopulmonary bypass. *Anesth. Analg.* 89(4), 823–828 (1999).

28 Gundersen Y, Vaagenes P, Pharo A, Valo ET, Opstad PK. Moderate hypothermia blunts the inflammatory response and reduces organ injury after acute haemorrhage. *Acta Anaesthesiol. Scand.* 45(8), 994–1001 (2001).

29 Hastbacka J, Tiainen M, Hynninen M *et al.* Serum matrix metalloproteinases in patients resuscitated from cardiac arrest. The association with therapeutic hypothermia. *Resuscitation* 83(2), 197–201 (2012).

30 Meybohm P, Gruenewald M, Zacharowski KD *et al.* Mild hypothermia alone or in combination with anesthetic post-conditioning reduces expression of inflammatory cytokines in the cerebral cortex of pigs after cardiopulmonary resuscitation. *Crit. Care* 14(1), R21 (2010).

31 Tavares-Murta BM, Cordeiro AO, Murta EF, Cunha Fde Q, Bisinotto FM. Effect of myocardial protection and perfusion temperature on production of cytokines and nitric oxide during cardiopulmonary bypass. *Acta Cir. Bras.* 22(4), 243–250 (2007).

32 Kang J, Albadawi H, Casey PJ *et al.* The effects of systemic hypothermia on a murine model of thoracic aortic ischemia reperfusion. *J. Vasc. Surg.* 52(2), 435–443 (2010).

33 Hofstetter C, Boost KA, Flondor M *et al.* Anti-inflammatory effects of sevoflurane and mild hypothermia in endotoxemic rats. *Acta Anaesthesiol. Scand.* 51(7), 893–899 (2007).

34 Morita Y, Oda S, Sadahiro T *et al.* The effects of body temperature control on cytokine production in a rat model of ventilator-induced lung injury. *Cytokine* 47(1), 48–55 (2009).

35 Pournot H, Bieuzen F, Louis J *et al.* Time-course of changes in inflammatory response after whole-body cryotherapy multi exposures following severe exercise. *PLoS ONE* 6(7), e22748 (2011).

36 Coassin M, Duncan KG, Bailey KR, Singh A, Schwartz DM. Hypothermia reduces secretion of vascular endothelial growth factor by cultured retinal pigment epithelial cells. *Br. J. Ophthalmol.* 94(12), 1678–1683 (2010).

- **An experimental study in ophtalmology suggesting that cryotherapy might downregulate VEGF production.**

37 Fujimoto K, Fujita M, Tsuruta R *et al.* Early induction of moderate hypothermia suppresses systemic inflammatory cytokines and intracellular adhesion molecule-1 in rats with caerulein-induced pancreatitis and endotoxemia. *Pancreas* 37(2), 176–181 (2008).

38 Webster CM, Kelly S, Koike MA, Chock VY, Giffard RG, Yenari MA. Inflammation and NFkappaB activation is decreased by hypothermia following global cerebral ischemia. *Neurobiol. Dis.* 33(2), 301–312 (2009).

39 Suehiro E, Fujisawa H, Akimura T *et al.* Increased matrix metalloproteinase-9 in blood in association with activation of interleukin-6 after traumatic brain injury: influence of hypothermic therapy. *J. Neurotrauma.* 21(12), 1706–1711 (2004).

40 Truettner JS, Alonso OF, Dalton Dietrich W. Influence of therapeutic hypothermia on matrix metalloproteinase activity after traumatic brain injury in rats. *J. Cereb. Blood Flow Metab.* 25(11), 1505–1516 (2005).

41 Harris ED, Jr., McCroskery PA. The influence of temperature and fibril stability on degradation of cartilage collagen by rheumatoid synovial collagenase. *N. Engl. J. Med.* 290(1), 1–6 (1974).

- **An experimental study suggesting that cryotherapy might downregulate collagenase activity in RA.**

42 Cao J, Xu J, Li W, Liu J. Influence of selective brain cooling on the expression of ICAM-1 mRNA and infiltration of PMNLs and monocytes/macrophages in rats suffering from global brain ischemia/reperfusion injury. *Biosci. Trends.* 2(6), 241–244 (2008).

43 Pournot H, Bieuzen F, Duffield R, Lepretre PM, Cozzolino C, Hausswirth C. Short term effects of various water immersions on recovery from exhaustive intermittent exercise. *Eur. J. Appl. Physiol.* 111(7), 1287–1295 (2011).

44 Dugue B, Smolander J, Westerlund T *et al.* Acute and long-term effects of winter swimming and whole-body cryotherapy on plasma antioxidative capacity in healthy women. *Scand. J. Clin. Lab. Invest.* 65(5), 395–402 (2005).

45 Leppaluoto J, Westerlund T, Huttunen P *et al.* Effects of long-term whole-body cold exposures on plasma concentrations of ACTH, beta-endorphin, cortisol, catecholamines and cytokines in healthy females. *Scand. J. Clin. Lab. Invest.* 68(2), 145–153 (2008).

46 Brenner IK, Castellani JW, Gabaree C *et al.* Immune changes in humans during cold exposure: effects of prior heating and exercise. *J. Appl. Physiol.* 87(2), 699–710 (1999).

47 Moher D, Liberati A, Tetzlaff J, Altman DG, the PRISMA Group. Preferred Reporting Items for Systematic Reviews ans Meta-Analyses: The PRISMA Statement. *PLoS Med.* 6(7), e1000097 (2009).

48 Jadad AR, Moore RA, Carroll D *et al.* Assessing the quality of reports of randomized clinical trials: is blinding necessary? *Control Clin. Trials* 17(1), 1–12 (1996).

49 Ivey M, Johnston RV, Uchida T. Cryotherapy for postoperative pain relief following knee arthroplasty. *J. Arthroplasty.* 9(3), 285–290 (1994).

50 Rembe EC. Use of cryotherapy on the postsurgical rheumatoid hand. *Phys. Ther.* 50(1), 19–23 (1970).

51 Bulstrode S CA, Harrison RA. a controlled trial to study the effects of therapy on joint inflammation in chronic arthritis. *Physiother. Pract.* 2, 104–108 (1986).

52 Kirk JA, Kersley GD. Heat and cold in the physical treatment of rheumatoid arthritis of the knee. A controlled clinical trial. *Ann. Phys. Med.* 9(7), 270–274 (1968).

53 Williams J, Harvey J, Tannenbaum H. Use of superficial heat versus ice for the rheumatoid arthritis shoulder: a pilot study. *Physiother. Can.* 38(1), 8–13 (1986).

54 Metzger D, Zwingmann C, Protz W, Jackel WH. [Whole-body cryotherapy in rehabilitation of patients with rheumatoid diseases–pilot study]. *Rehabilitation (Stuttg)* 39(2), 93–100 (2000).

55 Kelly S, Taylor P, Pitzalis C. Three dimensional power Doppler signal is more sensitive to change than two dimensional signal under a physiological stimulus. *Arthritis Rheum.* 58, S467 (2008).

56 Wojtecka-Lukasik E, Ksieszopolska-Orlowska K, Burakowski T *et al.* Effect of cryotherapy on adjuvant arthritis in the rat. *Ann. Rheum. Dis.* (Suppl.), AB0050 (2002).

57 Knorre BV, Keitel W. Comparative study of therapy: ultrasound, cryotherapy and intra-articular corticosonoids to treat alterations of the shoulder joint due to inflammation. *Z. Physiother.* 42, 221–225 (1990).

58 Jonderko G, Szopinski J, Galaszek M, Galaszek Z. Influence of heat and cold air therapy in sequence on the pain threshold as well as on the subjective pain feeling in rheumatoid arthritis. *Z. Phys. Med. Baln. Med. Klim.* 19, 49–53 (1990).

59 Albrecht K, Albert C, Lange U, Muller-Ladner U, Strunk J. Different effects of local cryogel and cold air physical therapy in wrist rheumatoid arthritis visualised by power Doppler ultrasound. *Ann. Rheum. Dis.* 68(7), 1234–1235 (2009).

60 Strunk J, Strube K, Klingenberger P, Muller-Ladner U, Lange U. Two- and three-dimensional Doppler sonographic evaluation of the effect of local cryotherapy on synovial perfusion in wrist arthritis. *Rheumatology (Oxford)* 45(5), 637–640 (2006).

61 Ellegaard K, Torp-Pedersen S, Henriksen M, Lund H, Danneskiold-Samsoe B, Bliddal H. Influence of recent exercise and skin temperature on ultrasound Doppler measurements in patients with rheumatoid arthritis–an intervention study. *Rheumatology (Oxford)* 48(12), 1520–1523 (2009).

• **A randomized controlled trial with crossover design showing wrist power Doppler hypersignal decrease after a single cold pack application.**

62 Schlesinger N, Detry MA, Holland BK *et al.* Local ice therapy during bouts of acute gouty arthritis. *J. Rheumatol.* 29(2), 331–334 (2002).

• **A randomized controlled trial in gout showing greater pain VAS decrease in patients treated by corticosteroids + colchicine + ice packs versus corticosteroids + colchicine.**

63 Jastrzabek R, Straburzynska-Lupa A, Rutkowski R, Romanowski W. Effects of different local cryotherapies on systemic levels of TNF-alpha, IL-6, and clinical parameters in active rheumatoid arthritis. *Rheumatol. Int.* 33, 2053–2060 (2013).

•• **A controlled trial comparing two cryotherapy modalities (cold air versus liquid nitrogen, 20 applications over 10 days, two groups of 20 patients) and showing significant decreases in pain VAS, DAS28 and serum TNF-α.**

64 Orlova EV, Karateev DE. Efficacy of complex rehabilitation of patients with early rheumatoid arthritis. *Ann. Rheum. Dis.* 71(Suppl. 3), 466 (2012).

65 Lange U, Uhlemann C, Muller-Ladner U. [Serial whole-body cryotherapy in the criostream for inflammatory rheumatic diseases. A pilot study]. *Med. Klin. (Munich)* 103(6), 383–388 (2008).

66 Braun KP, Brookman-Amissah S, Geissler K, Ast D, May M, Ernst H. [Whole-body cryotherapy in patients with inflammatory rheumatic disease. A prospective study]. *Med. Klin. (Munich)* 104(3), 192–196 (2009).

67 Gizinska M, Straburzynska-Lupa A, Romanowski W *et al.* The influence of whole-body cryotherapy (-110°C) and kinesitherapy on selected functional parameters and DAS28 in female patients with rheumatoid arthritis. *Ann. Rheum. Dis.* 69(Suppl. 3), 719 (2010).

68 Samborski W, Lisinski P, Sobieska M. An effectiveness of kinesis therapy preceded by cryotherapy in rheumatoid arthritis patients with coxitis. *Ann. Rheum. Dis.* 65(Suppl. II), 613 (2006).

69 Straub RH, Pongratz G, Hirvonen H, Pohjolainen T, Mikkelsson M, Leirisalo-Repo M. Acute cold stress in rheumatoid arthritis inadequately activates stress responses and induces an increase of interleukin 6. *Ann. Rheum. Dis.* 68(4), 572–578 (2009).

• **A randomized controlled trial showing a decrease in serum IL-6 levels in RA patients without corticosteroids after chronic cryotherapy (LC applied to five joints simultaneously or WBC (-60°C) both performed two- or three-times a day for 7 days). Conversely, -110°C WBC following the same application protocol increased serum IL-6 levels. This suggests potential systemic anti-inflammatory effects of cryotherapy and a possible pro-inflammatory effect of cryotherapy beyond a cold stimulation intensity threshold.**

70 Hirvonen HE, Mikkelsson MK, Kautiainen H, Pohjolainen TH, Leirisalo-Repo M. Effectiveness of different cryotherapies on pain and disease activity in active rheumatoid arthritis. A randomised single blinded controlled trial. *Clin. Exp. Rheumatol.* 24(3), 295–301 (2006).

• **A randomized controlled trial showing significant decreases in pain VAS and DAS28 in RA patients treated with LC as well as WBC (performed two- or three-times a day for 7 days)**

71 Wojtecka-Lukasik E, Ksiezopolska-Orlowska K, Gaszewska E *et al.* Cryotherapy decreases histamine levels in the blood of patients with rheumatoid arthritis. *Inflamm. Res.* 59(Suppl. 2), S253–S255 (2010).

72 Weinberger A, Giler S, Nyska A. Treatment of inflammatory synovitis with ice

 doi: 10.1586/1744666X.2014.870036

application. *Arthritis Rheum.* 38(Suppl.), S242, 532 (1995).

73 Mourot L, Cluzeau C, Regnard J. Hyperbaric gaseous cryotherapy: effects on skin temperature and systemic vasoconstriction. *Arch. Phys. Med. Rehabil.* 88, 1339–1343 (2007).

74 Pavlov VA, Tracey KJ. The cholinergic anti-inflammatory pathway. *Brain Behav. Immun.* 19, 493–499 (2005).

75 Tracey KJ. Reflex control of immunity. *Nat. Rev. Immunol.* 9, 418–428 (2009).

76 Shepherd JT, Rusch NJ, Vanhoutte PM. Effect of cold on the blood vessel wall. *Gen. Pharmacol.* 14, 61–64 (1983).

77 Banfi G, Melegati G, Barassi A *et al.* Effects of whole-body cryotherapy on serum mediators of inflammation and serum muscle enzymes in athletes. *J. Therm. Biol.* 34, 55–59 (2009).

78 Stalman A, Berglund L, Dungnerc E *et al.* Temperature-sensitive release of prostaglandin e2 and diminished energy requirements in synovial tissue with postoperative cryotherapy: a prospective randomized study after knee arthroscopy. *J. Bone Joint Surg. Am.* 93, 1961–1968 (2011).

79 Miller E, Markiewicz L, Saluk J *et al.* Effect of short-term cryostimulation on antioxidative status and its clinical applications in humans. *Eur. J. Appl. Physiol.* 112, 1645–1652 (2012).

80 Zhang H, Zhang JJ, Mei YW *et al.* Effects of immediate and delayed mild hypothermia on endogenous antioxidant enzymes and energy metabolites following global cerebral ischemia. *Chin. Med. J.* 124, 2764–2766 (2011).

81 Demoulin C, Brouwers M, Darot S *et al.* Comparison of gaseous cryotherapy with more traditional forms of cryotherapy following total knee arthroplasty. *Ann. Phys. Rehabil. Med.* 55, 229–240 (2012).

82 Banfi G, Lombardi G, Colombini A *et al.* Whole-body cryotherapy in athletes. *Sports Med.* 40, 509–517 (2010).

Websites

101 International Standard Randomised Controlled Trial Number Register. www.controlled-trials.com/isrctn

102 ClinicalTrials.gov. www.clinicaltrials.gov

103 WHO. International Clinical Trials Registry Platform. www.who.int/ictrp/search/en

104 Wells G, Shea B. Data extraction for nonrandomised systematic reviews. University of Ottowa, Ottowa. www.ohri.ca/programs/clinical_epidemiology/oxford.htm