Mud-bath therapy and oral glucosamine sulfate in patients with knee osteoarthritis: a randomised, controlled, crossover study

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Abstract

Objective

To evaluate the efficacy and safety of combined treatment of mud-bath therapy and glucosamine crystalline sulfate (GlcN-S) in patients with knee osteoarthritis (OA).

Methods

This study was a randomised, controlled, crossover investigation. Patients were randomly assigned (1:1) by the investigators to two groups, named group 1 and 2. Group 1 included twenty-three patients receiving oral GlcN-S treatment from the beginning of the study (T0) to the end of the 3rd month of treatment (T3) and a combined treatment of both mud-bath therapy and GlcN-S from T3 to the end of the study at six months (T6).

Group 2 included twenty-two patients receiving a combined treatment of both mud-bath therapy and GlcN-S from T0 to T3 and that discontinued mud-bath therapy, receiving GlcN-S treatment alone, from T3 to T6.

Primary endpoints of the study consisted of evaluating OA severity and activity at baseline and at follow-up visits.

Results

All 45 patients, eligible for the study, completed the period of the crossover.

In group 1, no significant difference was shown in the comparison from T0 to T3, while from T3 to T6 most variables were significantly improved. In group 2, instead, the comparison between T0 and T3 showed a significant difference in different parameters. When comparing T3 and T6, despite an improvement of all the variables, no significant difference was shown.

Conclusion

The association of GlcN-S and mud-bath therapy has a positive and safe role in improving pain, function and quality of life in knee OA patients.

Key words

knee osteoarthritis, mud-bath therapy, glucosamine

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Introduction

Osteoarthritis (OA) represents the prevalent form of chronic joint disease, leading progressively to severe physical impairment, disability and reduced quality of life in adults from middle to elderly age and mainly in people over 65 years of age (1).

The progressive damage of cartilage is associated with bone remodelling and the formation of new bone tissue in the form of osteophytes and subchondral sclerosis (1). These findings can be variably associated with the concomitant presence of synovitis and inflammation of the whole joint's structures involved in the OA processes. The dreadful complication of severe OA is represented by massive joint damage and destruction leading to significant functional impairment and the need for articular prosthetic substitution (1, 2).

All joints may be affected by OA processes and the knee represents one of the most common localisation (1). diDDagnosis of knee OA relies on diagnostic criteria showing high specificity and sensitivity (3) and the Kellgren and Lawrence grading scale represents a radiological imaging classification ranging from grade 1 to grade 4, on the basis of presence and severity of narrowing of joint space, osteophytosis, sclerosis of knees and deformity of bone extremities (4).

Over the past years, along with treatment for pain relief, therapy with non-steroidal anti-inflammatory drugs (NSAIDs) has been often used in clinical practice for OA. However, long-term use of NSAIDs is avoided mainly due to their gastrointestinal, cardiovascular and kidney side effects (5, 6).

Glucosamine (GlcN), a natural amino sugar, has been increasingly used in the therapy of OA in different formulations (7, 8). Several data derived from randomised clinical trials (RCT) in OA have highlighted the safety and effectiveness of glucosamine crystalline sulfate (GlcN-S), due to its pharmacological, metabolic and mild anti-inflammatory properties and activities on the cartilage and chondrocytes (9-20). Recently, in addition to safety, GlcN-S has shown to be effective in the improvement of pain and functional impairment consequential to symptomatic OA (7).

Therapy of OA also includes non-pharmacologic modalities, such as spa therapy (5, 6). Mud packs and balneotherapy represent two spa therapy modalities that can be used alone or in combination. These are considered a potentially successful therapeutic option in knee OA patients improving function, perceived pain and quality of life (21-25).

In particular, a mud pack is a natural product providing a combination of mineral water with organic or inorganic material derived from geological and/or biological processes. It is used in the form of a mud wrap or bath and its main indication consists in reducing musculoskeletal pain and improving joint motion, muscle strength and functional mobility (21, 22). The effects of mud packs, as well balneotherapy on OA, are also strictly connected to specific physical-chemical characteristics (26, 27).

Until today, no RCT investigating combined treatment of mud-bath therapy and GlcN-S in patients with knee OA has been performed. Thus, the aim of this study was to evaluate efficacy and safety of this combined therapy.

Patients and methods

This study was a randomised, controlled, crossover investigation, with the aim of evaluating the short-term efficacy and safety of combined treatment providing mud-bath therapy in combination with oral GlcN-S in adults patients with knee OA.

Patients selection

Sixty consecutive patients (M/F: 26/34; mean age: 68.34±8.54 years) presenting bilateral knee pain were observed at the Orthopaedic Unit of Rizzoli Hospital, Ischia, Naples (Italy) during a eight-month period.

Inclusion criteria were represented by diagnosis of knee OA (3) and a baseline standing knee radiographs score ranging from 1 to 2 using the Kellgren method (4). Therefore patients with mild/moderate disease. The clinical visit was performed by one of rheumatologist. It was required that radiographs had been performed no longer than 3 months before the recruitment visit.

Exclusion criteria were represented by: concomitant occurrence of autoimmune

diseases and arthropathies other than OA, fibromyalgia, history of knee trauma and surgery, subjective intolerance or diseases contraindicating mud-bath therapy. Other exclusion criteria were previous use of knee intra joint hyaluronic acid and/or corticosteroid injection, use of chondroprotective agents, vitamin D, bisphosphonates, hormonal treatment, systemic corticosteroids. Another exclusion criteria was treatment with systemic and topic NSAIDs, and analgesic agents within the past week prior to the study and during the study period, and mud-bad therapy, physiotherapy and massages within the past 6 months prior to the study.

Design

Among the overall 60 patients observed, a total of 45 patients (M/F: 20/25; mean age: 65.44±7.48 years, range: 51-81 years) with a diagnosis of established knee OA according to the American College of Rheumatology Classification Criteria of OA (3) were included in the study for randomisation.

Patients were randomly assigned (1:1) by the investigators to two groups, named group 1 and 2. All subjects underwent follow-up visits every 3 months. Allocation to the group was alternative, starting with group 1, and performed at the time of recruitment visit by the reumatologist.

Group 1 included twenty-three patients receiving oral GlcN-S treatment from the beginning of study (T0) to the end of the 3rd month of treatment (T3) and a combined treatment of both mud-bath therapy and GlcN-S from T3 to the end of the study at six months (T6). Group 2 included twenty-two patients receiving a combined treatment of both mud-bath therapy and GlcN-S from T0 to T3 and that discontinued mud-bath therapy, receiving GlcN-S treatment alone, from T3 to T6 (Fig. 1).

Crystalline GlcN-S formulation at the dose of 1500 mg once daily (Rottapharm, S.p.A., Monza Italy) was administered orally once-daily in the form of packets of powder for oral solution.

On the island of Ischia (South of Italy), near the Gulf of Naples, several craters are located above huge layers of tuff and are characterised by a mild volcanic activity, conferring peculiar physicalchemical characteristics to the mud and water of this area. In particular, all the waters contain a significant amount of sodium (Na+) and chloride (Cl-) (28, 29). Besides, mineral water drawn from the locality "Ischia Porto-Lido" near the sea, presents saline-alkaline sulfate properties and characteristics due to underground leaked-in seawater. The average temperature is above 31°C at the source and fixed residue at a temperature of 180°C is of 6315 mg/L and concentrations of chloride and sodium ions are 3199 and 1689 mg/L, respectively (28, 29). Another element is represented by sulfate anions (SO4-) (239.6 mg/L) (28, 29).

Ischia mud is composed of volcanic-derived clay containing algae, microorganisms, and minerals mixed with local thermal water (28, 29). Mud-bath therapy was a combination of mud packs applied on both knees, up to a thickness of 10 cm at a temperature of around 47°C for 20 min and, when the mud was wiped off, patients were washed in mineral water, in a bathtub, at a temperature of around 38°C for more than 15 minutes. A total of 12 mud-bath applications, each biweekly, were carried out over a 3-month period in each group.

The primary endpoints of the study consisted of evaluating OA severity and activity at baseline and at follow-up visits by measurements of knee circumferential diameter and ranges of motion, and questionnaires on OA severity and activity, pain intensity, patient's health, functional status, therapy preferences, satisfaction and quality of life, anxiety and social dysfunction, at baseline and at follow-up visits.

The secondary endpoint verified the safety of mud-bath therapy and GlcN-S alone and/or in combination in patients with knee OA.

Clinical assessment

For each patient, data included personal history, physical examination with recording of measures of OA severity and activity: knee circumferential measurements, active knee extension range of motion (ROM) and active knee flexion

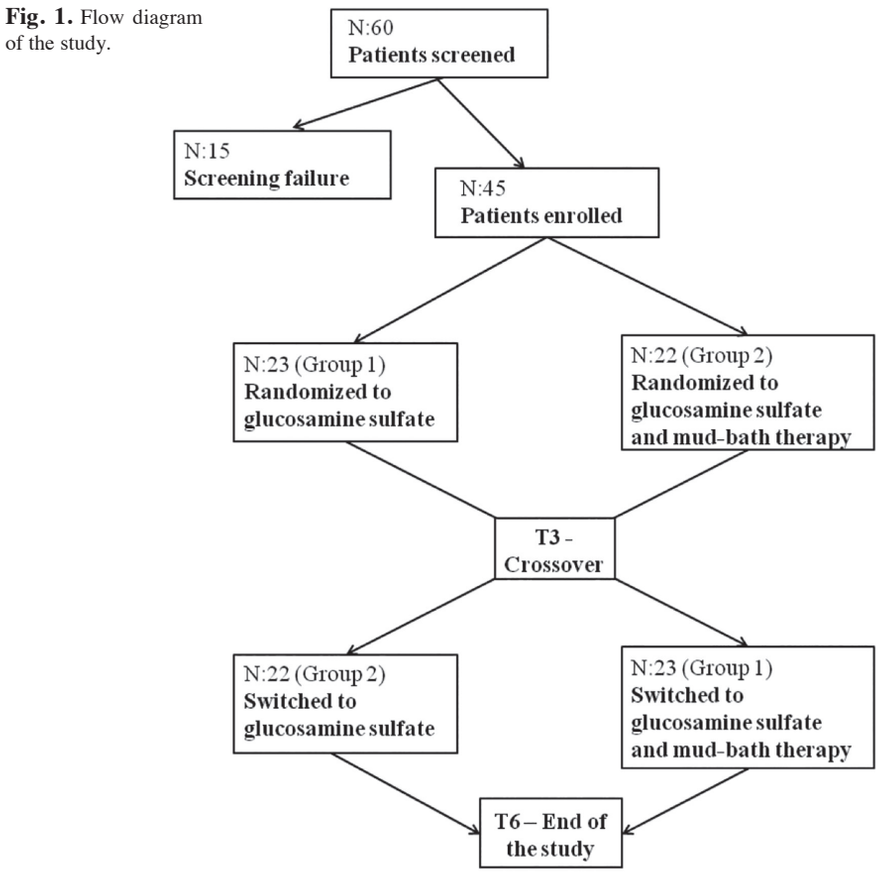

Fig. 1. Flow diagram of the study.

Table I. Characteristics of the study population.

Characteristics	Group 1	Group 2	*p*-value*
N	23	22	
F/M	10/12	10/13	0.878
mean age ± SD	64.38 ± 10.68	66.07 ± 9.51	0.688
BMI, mean ± SD	25.99 ± 6.58	27.88 ± 10.42	0.419
Disease duration of Knee OA, mean ± SD	11.54 ± 5.91	12.57 ± 6.21	0.754
Comorbidities, n° patients (%)			
Hypertension	6 (26.09)	5 (22.73)	0.793
Osteoporosis	7 (31.82)	8 (34.78)	0.753
Diabetes Mellitus	5 (21.74)	7 (31.82)	0.445
Dyslipidaemia	8 (34.78)	5 (22.73)	0.372
Hypo-, hyperthyroidism	3 (13.04)	5 (22.73)	0.396

*p value by sample t-test.
N: number; F: female; M: male; SD: standard deviation; BMI: Body Mass Index; OA: osteoarthritis.

ROM. In particular, knee circumferential measurements were performed in correspondence of midpatella area with an ordinary tape measure (30). Active knee extension and flexion range-of-motion of patients was measured by universal goniometer (31). Pain intensity, OA severity and activity were also evaluated by the use of the Western Ontario and McMaster Universities (WOMAC) Osteoarthritis Index, Lequesne algo-functional Index (32, 33), and visual analogue scale (VAS) (34). The Health Assessment Questionnaire (HAQ) was used for the evaluation of functional status (35, 36).

For the evaluation of somatic symptoms, anxiety, social dysfunction and depression, the 28-item General Health Questionnaire (GHQ-28) was used (37, 38).

For each considered variable, data were recorded and evaluated at enrolment and at each 3-month follow-up visit.

All the outcome measures were evaluated by a trained rheumatologist with expertise in osteoarthritis. In addition, the outcome assessor was blinded to treatment assignment.

The study protocol was approved by the Ethics Committee of the Local Health Unit (Naples, Italy) (protocol number: 12/CE). All patients signed and approved the informed consent.

Statistical analysis

Statistical analysis was performed using the SPSS software, v. 18 (SPSS inc, Chicago, Ill). Continuous data were expressed as means±SD, categorical variables as percentages. Continuous variables were compared by an independent sample *t*-test and by ANOVA analysis when needed. For values with a skewed distribution, the Mann-Whitney test was used to compare non-parametric continuous variables. The chi-square test was used to analyse categorical data and, when the minimum expected value for categorical data was <5, the Fisher exact test was used.

Results

All 45 patients, eligible for the study, completed the period of the crossover.

Demographic and clinical characteristics of two groups of patients are reported in Table I.

At baseline, no significant differences concerning the severity of knee osteoarthritis were present between group 1 and 2.

Results on indices of OA severity and activity showing active knee flexion ROM, active knee extension ROM and knee circumferential measurement are summarised in Table II.

Results on indices of pain intensity, OA severity and activity including WOMAC OA Index, Lequesne algo-functional index and VAS are summarised in Table III.

Results on indices of quality of life and psycho-social status: HAQ, GHQ-28 are summarised in Table IV.

In group 1, no significant difference was shown in the comparison from T0 to T3. When considering for group 1 the interval between T3 and T6, Flexion ROM (*p*:0,041), Extension ROM (*p*:0.0006), Lequesne algo-functional index (*p*=0.002), and HAQ (*p*=0.005) were significantly improved. In the comparison between T0 and T6 in group 1, a significant difference was recorded in the following parameters: Flexion ROM (*p*=0.049), Extension ROM (*p*:0.002), Lequesne algofunctional index (*p*:0.0007), VAS (*p*=0.017), HAQ (*p*=0.004) and GHQ-28 (*p*=0.004).

On the other hand, in group 2, the comparison between T0 and T3 showed already a significant difference in the

Table II. Indices of severity and osteoarthritis activity at baseline and at each follow-up three-month visit of the two groups after randomisation.

Variables		GROUP 1						GROUP 2					
		T0	T3	T3	T6	T0	T6	T0	T3	T3	T6	T0	T6
Active knee flexion ROM (grade)		122.5±27.4	122.6±25.4	122.6±25.4	136.8±21.3	122.5±27.4	136.8±21.3	128.9±13.6	137.2±11.2	137.2±11.2	138.6±9.1	128.9±13.6	138.6±9.1
	p	0.987		**0.041***		**0.049***		**0.036***		0.664		**0.009***	
Active knee extension ROM (grade)		3.5±2.3	3.2±2.4	3.2±2.4	5.5±1.9	3.5±2.3	5.5±1.9	3.5±2.1	5.4±2.5	5.4±2.5	5.8±1.9	3.5±2.1	5.8±1.9
	p	0.673		**0.0006***		**0.002***		**0.011***		0.534		**0.0005***	
Knee circumferential measurement (cm)		41.7±8.2	40.8±8.1	40.8±8.1	37.9±7.4	41.7±8.2	37.9±7.4	41.2±4.9	39.5±4	39.5±4	39±3.7	41.2±4.9	39±3.7
	p	0.691		0.199		0.093		0.239		0.674		0.115	

ROM: range-of-motion.

Table III. Indices of pain intensity and osteoarthritis severity at baseline and at each follow-up three-month visit of the two groups after randomisation.

		GROUP 1						GROUP 2					
Variables		T0	T3	T3	T6	T0	T6	T0	T3	T3	T6	T0	T6
WOMAC OA index		32.3±22.1	31.9±21.2	31.9±21.2	20.8±19.8	32.3±22.1	20.8±19.8	35±22.5	16±11.5	16±11.5	15.9±12.2	35±22.5	15.9±12.2
	p	0.951		0.067		0.064		**0.001***		0.967		**0.001***	
LEQUESNE algo functional index		11.2±5.3	10.4±4.8	10.4±4.8	6±4.7	11.2±5.3	6±4.7	11.3±4.4	5.5±4.2	5.5±4.2	7±4.5	11.3 ± 4.4	7 ± 4.5
	p	**0.569**		**0.002***		**0.0007***		0.0001*		0.255		**0.004***	
VAS		42.8±23.9	37.3±18.7	37.3±18.7	27±20.3	42.8±23.9	27±20.3	41.6±2	22.4±16.7	22.4±16.7	23.7±19.6	41.6±2	23.7±19.6
	p	0.379		0.073		**0.017***		**0.001***		0.82		**0.005***	

WOMAC: Western Ontario and McMaster Universities; VAS: visual analogue scales.

Table IV. Indices of quality of life and psycho-social status at baseline and at each follow-up three-month visit of the two groups after randomisation.

		GROUP 1						GROUP 2					
Variables		T0	T3	T3	T6	T0	T6	T0	T3	T3	T6	T0	T6
HAQ		0.8±0.63	0.8±0.6	0.8±0.6	0.3±0.4	0.8±0.6	0.3±0.4	0.7±0.6	0.3±0.4	0.3±0.4	0.2±0.3	0.7±0.6	0.2±0.3
	p	0.977		**0.005***		**0.004***		**0.007***		0.959		**0.003***	
GHQ28		2±0.5	1.7±0.5	1.7±0.5	1.6±0.5	2±0.5	1.6±0.5	1.9±0.5	1.5±0.4	1.5±0.40	1.6±0.5	1.9±0.5	1.6±0.5
	p	0.074		0.261		**0.004***		**0.007***		0.448		0.07	

HAQ: Health Assessment Questionnaire; GHQ-28: 28-item General Health Questionnaire.

following parameters: Flexion ROM (p=0.036), Extension ROM (p=0.011), WOMAC index (p=0.001), Lequesne algo-functional index (p=0.0001), VAS (p=0.001), HAQ (p=0.007) and GHQ-28 (p=0.007). When comparing T3 and T6 in group 2, despite improvement of all the variables, no significant difference was shown. In the comparison between T0 and T6 in group 2, a significant difference was shown in the following parameters: Flexion ROM (p=0.009), Extension ROM (p=0.0005), WOMAC (p:0.001), Lequesne algo-functional index (p=0.004) and VAS (p=0.005).

Knee diameter was the only variable which never showed a significant change in either group.

No adverse events were reported in either group.

When considering the entire study population, between T0 and T3, a significant improvement was found for the parameters WOMAC index (p=0.039), Lequesne algo-functional index (p=0.003), GHQ28 (p=0.002) and VAS (p=0.007). Between T3 and T6, the entire study population showed a significant improvement in Flexion ROM (p=0.043), Extension ROM (p=0.0043) and HAQ (p=0.023).

The comparison between T0 and T6 showed a significant improvement for the following parameters: Flexion ROM (p=0.004), Extension ROM (p=0.0001), Knee circumferential measurement (p=0.025), HAQ (p=0.0001), WOMAC index (p=0.0005), Lequesne algo-functional index (p=0.0001), GHQ28 (p=0.0008), and VAS (p=0.0003).

Discussion

The objective of this study was to determine the efficacy and safety of mud-bath therapy and GlcNS alone and/or in combination in patients with mild/moderate knee OA.

This work is the first attempt to evaluate this approach which may represent a relevant advance in knee OA patient management.

The results of the present randomised, controlled, crossover study showed that in group 1 which included twenty-three patients receiving GlcN-S treatment from T0 to T3 and a combined treatment of both mud-bath therapy in association with GlcN-S from T3 to T6, the only significant changes in the variables considered were found in this latter study period. In fact, a significant improvement of all the evaluated parameters was detected in the period between and T3 and T6, except for knee circumferential measurements, WOMAC, VAS and GHQ28.

In group 2 in which twenty-two patients received a combined treatment of both mud-bath therapy and GlcN-S from T0 to T3 and that discontinued mud-bath therapy receiving GlcN-S alone from T3 to T6, a significant improvement of all the evaluated parameters was verified in the period T0 to T3, except for knee circumferential measurements. This result remained stable at T6, with no further significant improvement.

In summary, group 2 showed globally better outcomes at the end of the first period and remained in a stable state during the following three months, while group 1 had best outcomes in the period T3 to T6. Therefore, the results indicate that greater improvement was obtained when patients were using GlcN-S in combination with mud-therapy as compared to therapy with GlcN alone.

Some of the variable modifications obtained are limited from a clinical point of view. However, they indicate a trend that deserves attention and further evaluation, mainly in a field as osteoarthritis with limited therapeutical options.

However, in Group 2, treatment with GlcN alone was able to maintain the positive results of the first phase of combined therapy. Instead, in Group 1, it did not correlate with significant improvement of outcomes. In addition, due to the spa therapy effects in osteoarthritis, lasting 4-6 months, the improvement observed in patients of group 2 from T3 to T6 could be at least partially related to the long-term effects of mud treatment.

At present, the literature data show efficacy and safety of GlcN in knee OA patient, even if most available RCTs demonstrate that GlcN was not more effective than placebo in OA (7, 8, 15-17, 20). The therapeutic effect of mud-bath therapy in knee OA has been evaluated in different studies, showing a significant improvement for patients that received mud therapy in comparison with the control group (39, 40). The data derived from the present RCT suggest that the association of GlcN and mud-bath therapy has a positive and safe role in improving pain, function and quality of life in knee OA patients. This effect could be related to the peculiar physico-chemical properties of Ischia spa therapy.

A major limitation of the study was the lack of blindness for the patient, which is particularly difficult to achieve given the characteristics of mud-bath therapy. In addition, GlcN-S may exert its best effect when continuously used for more than 6 months. Therefore, further studies should be planned with longer duration and follow-up.

In conclusion, the characteristics of Ischia natural mineral water and mud used in therapy represent an important therapeutic approach in OA patients, in combination with GlcN-S, and should be taken into account in further studies and in future re-evaluation of treatment guidelines for knee OA.

References

1. LOESER RF, GOLDRING SR, SCANZELLO CR, GOLDRING MB: Osteoarthritis: a disease of the joint as an organ. *Arthritis Rheum* 2012; 64: 1697-707.
2. GERYK LL, CARPENTER DM, BLALOCK SJ, DEVELLIS RF, JORDAN JM: The impact of co-morbidity on health-related quality of life in rheumatoid arthritis and osteoarthritis patients. *Clin Exp Rheumatol* 2015; 33: 366-74.
3. ALTMAN R, ASCH E, BLOCH D *et al*.: Development of criteria for the classification and reporting of osteoarthritis. Classification of osteoarthritis of the knee. Diagnostic and Therapeutic Criteria Committee of the American Rheumatism Association. *Arthritis Rheum* 1986; 29: 1039-49.
4. KELLGREN JH, LAWRENCE JS: Radiological assessment of osteo-arthrosis. *Ann Rheum Dis* 1957; 16: 494-502.
5. ALSHAMI AM: Knee osteoarthritis related pain: a narrative review of diagnosis and treatment. *Int J Health Sci* (Qassim) 2014; 8: 85-104.
6. JORDAN KM, ARDEN NK, DOHERTY M *et al*.; STANDING COMMITTEE FOR INTERNATIONAL CLINICAL STUDIES INCLUDING THERAPEUTIC TRIALS ESCISIT: EULAR Recommendations 2003: an evidence based approach to the management of knee osteoarthritis: Report of a Task Force of the Standing Committee for International Clinical Studies Including Therapeutic Trials (ESCISIT). *Ann Rheum Dis* 2003; 62: 1145-55.
7. TOWHEED TE, MAXWELL L, ANASTASSIADES TP *et al*.: Glucosamine therapy for treating osteoarthritis. *Cochrane Database Syst Rev* 2005; 2: CD002946.
8. TOWHEED TE, ANASTASSIADES T: Glucosamine therapy for osteoarthritis: an update. *J Rheumatol* 2007; 34: 1787-90.
9. PEAT G, MCCARNEY R, CROFT P: Knee pain and osteoarthritis in older adults: a review of community burden and current use of primary health care. *Ann Rheum Dis* 2001; 60: 91-7.
10. REGINSTER JY, DEROISY R, ROVATI LC *et al*.: Long-term effects of glucosamine sulphate on osteoarthritis progression: a randomised, placebo-controlled clinical trial. *Lancet* 2001; 357: 251-6.
11. PAVELKA K, GATTEROVA J, OLEJAROVA M, MACHACEK S, GIACOVELLI G, ROVATI LC: Glucosamine sulfate use and delay of progression of knee osteoarthritis: a 3-year, randomized, placebocontrolled, double-blind study. *Arch Intern Med* 2002; 162: 2113-23.
12. HERRERO-BEAUMONT G, IVORRA JA, DEL CARMEN TRABADO M *et al*.: Glucosamine sulfate in the treatment of knee osteoarthritis symptoms: a randomized, double blind, placebo-controlled study using acetaminophen as a side comparator. *Arthritis Rheum* 2007; 56: 555-67.
13. SAWITZKE AD, SHI H, FINCO MF *et al*.: Clinical efficacy and safety of glucosamine, chondroitin sulphate, their combination, celecoxib or placebo taken to treat osteoarthritis of the knee: 2-year results from GAIT. *Ann Rheum Dis* 2010; 69: 1459-64.
14. MCALINDON T: Why are clinical trials of glucosamine no longer uniformly positive? *Rheum Dis Clin North Am* 2003; 29: 789-801.
15. CIBERE J, KOPEC JA, THORNE A *et al*.: Randomized, double-blind, placebo-controlled glucosamine discontinuation trial in knee osteoarthritis. *Arthritis Rheum* 2004; 51: 738-45.
16. HOUPT JB, MCMILLAN R, WEIN C, PAGET-DELLIO SD: Effect of glucosamine hydrochloride in the treatment of pain of osteoarthritis of the knee. *J Rheumatol* 1999; 26: 2423-30.
17. HUGHES R, CARR A: A randomized, double-blind, placebo-controlled trial of glucosamine sulphate as an analgesic in osteoarthritis of the knee. *Rheumatology* (Oxford) 2002; 41: 279-84.
18. MCALINDON T, FORMICA M, LAVALLEY M, LEHMER M, KABBARA K: Effectiveness of glucosamine for symptoms of knee osteoarthritis: Results from an Internet-based randomized double-blind controlled trial. *Am J Med* 2004; 117: 643-9.
19. RINDONE JP, HILLER D, COLLACOTT E, NORDHAUGEN N, ARRIOLA G: Randomized, controlled trial of glucosamine for treating osteoarthritis of the knee. *West J Med* 2000; 172: 91-4.
20. CLEGG DO, REDA DJ, HARRIS CL *et al*.: Glucosamine, chondroitin sulfate, and the two in combination for painful knee osteoarthritis. *N Engl J Med* 2006; 354: 795-808.
21. TENTI S, CHELESCHI S, GALEAZZI M, FIORAVANTI A: Spa therapy: can be a valid option for treating knee osteoarthritis? *Int J Biometeorol* 2015; 59: 1133-43.
22. ESPEJO-ANTÚNEZ L, CARDERO-DURÁN MA, GARRIDO-ARDILA EM, TORRES-PILES S, CARO-PUÉRTOLAS B: Clinical effectiveness of mud pack therapy in knee osteoarthritis. *Rheumatology* (Oxford) 2013; 52: 659-68.
23. FIORAVANTI A, GIANNITTI C, BELLISAI B, IACOPONI F, GALEAZZI M: Efficacy of balneotherapy on pain, function and quality of life in patients with osteoarthritis of the knee. *Int J Biometeorol* 2012; 56: 583-90.
24. HARZY T, GHANI N, AKASBI N, BONO W, NEJJARI C: Short- and long-term therapeutic effects of thermal mineral waters in knee osteoarthritis: a systematic review of randomized controlled trials. *Clin Rheumatol* 2009; 28: 501-7.
25. FORESTIER R, FRANÇON A: Crenobalneotherapy for limb osteoarthritis: systematic literature review and methodological analysis. *Joint Bone Spine* 2008; 75: 138-48.
26. FIORAVANTI A, CANTARINI L, GUIDELLI GM, GALEAZZI M: Mechanisms of action of spa therapies in rheumatic diseases: what scientific evidence is there? *Rheumatol Int* 2011; 31: 1-8.
27. FIORAVANTI A, IACOPONI F, BELLISAI B, CANTARINI L, GALEAZZI M: Short- and long-term effects of spa therapy in knee osteoarthritis. *Am J Phys Med Rehabil* 2010; 89: 125-32.
28. CONSIGLI VP, MESSINA B: Hydrological research in Italy to-day. *Panminerva Med* 1961; 3: 474-6.
29. CARLÉ W: Die Mineral- und Thermalwässer am Golf von Neapel. *In*: *Geologische Rundschau*; 1965: 1261-313.
30. NICHOLAS JJ, TAYLOR FH, BUCKINGHAM RB, OTTONELLO D: Measurement of circumference of the knee with ordinary tape measure. *Ann Rheum Dis* 1976; 35: 282-4.
31. ROTHSTEIN JM, MILLER PJ, ROETTGER RF: Goniometric reliability in a clinical setting: Elbow and knee measurements. *Phys Ther* 1983; 63: 1611-5.
32. LEQUESNE M: Indices of severity and disease activity for osteoarthritis. *Semin Ar-*

thritis Rheum 1991; 6 (Suppl. 2): 48-54.

33. SALAFFI F, LEARDINI G, CANESI B *et al*.; GONORTHROSIS AND QUALITY OF LIFE ASSESSMENT (GOQOLA): Reliability and validity of the Western Ontario and McMaster Universities (WOMAC) Osteoarthritis Index in Italian patients with osteoarthritis of the knee. *Osteoarthritis Cartilage* 2003; 11: 551-60.
34. HAWKER GA, MIAN S, KENDZERSKA T, FRENCH M: Measures of adult pain: Visual Analog Scale for Pain (VAS Pain), Numeric Rating Scale for Pain (NRS Pain), McGill Pain Questionnaire (MPQ), Short-Form McGill Pain Questionnaire (SF-MPQ), Chronic Pain Grade Scale (CPGS), Short Form36 Bodily Pain Scale (SF-36 BPS), and Measure of Intermittent and Constant Osteoarthritis Pain (ICOAP). *Arthritis Care Res* (Hoboken) 2011; 63: S240-52.
35. FRIES JF, SPITZ PW, YOUNG DY: The dimensions of health outcomes: the health assessment questionnaire, disability and pain scales. *J Rheumatol* 1982; 9: 789-93.
36. RANZA R, MARCHESONI A, CALORI G *et al*.: The Italian version of the Functional Disability Index of the Health Assessment Questionnaire. A reliable instrument for multicenter studies on rheumatoid arthritis. *Clin Exp Rheumatol* 1993; 11: 123-8.
37. WERNEKE U, GOLDBERG DP, YALCIN I, USTÜN BT: The stability of the factor structure of the General Health Questionnaire. *Psychol Med* 2000; 30: 823-9.
38. FONTANESI F, GOBETTI C, ZIMMERMANN-TANSELLA C, TANSELLA M: Validation of the Italian version of the GHQ in a general practice setting. *Psychol Med* 1985; 15: 411-5.
39. FORESTIER R, DESFOUR H, TESSIER JM *et al*.: Spa therapy in the treatment of knee osteoarthritis: a large randomised multicentre trial. *Ann Rheum Dis* 2010; 69: 660-5.
40. FRAIOLI A, SERIO A, MENNUNI G *et al*.: A study on the efficacy of treatment with mud packs and baths with Sillene mineral water (Chianciano Spa Italy) in patients suffering from knee osteoarthritis. *Rheumatol Int* 2011; 31: 1333-40.