Original Article

Journal of Iranian Medical Council, Volume 9, Issue no. 1
http://dx.doi.org/10.18502/jimc.v9i1.20498

Efficacy of Ozone Therapy in Patients with Knee Osteoarthritis Referred to the Pain Clinic

Cyrus Emir Alavi[1,2], Shayan Samari[3], Sepideh Atefrad[3], Sara Sharifnia[3], Habib Eslami Kenarsari[3] and Mohaya Farzin[3]*

1. Neuroscience Research Center, Trauma Institute, Guilan University of Medical Sciences, Rasht, Iran
2. Department of Anesthesiology, School of Medicine, Guilan University of Medical Sciences, Rasht, Iran
3. Razi Clinical Research Development Unit, Guilan University of Medical Sciences, Rasht, Iran

*** Corresponding author**

Mohaya Farzin, PhD
Razi Clinical Research Development Unit, Guilan University of Medical Sciences, Rasht, Iran
Tel: +98 9112471138
Email: mf99155@yahoo.com

Received: 19 Nov 2024
Accepted: 5 Mar 2025

Citation to this article
Emir Alavi C, Samari Sh, Atefrad S, Sharifnia S, Eslami Kenarsari H, Farzin M. Efficacy of Ozone Therapy in Patients with Knee Osteoarthritis Referred to the Pain Clinic. *J Iran Med Counc.* 2026;9(1):246-52.

Abstract

Background: Knee Osteoarthritis (KOA) is a common degenerative joint disease that causes significant pain, disability, and impaired quality of life. Conventional therapies, including analgesics and physiotherapy, offer minimal alleviation. However, the emergence of ozone therapy, with its possible anti-inflammatory and regenerative properties, provides a promising and hopeful alternative.

Methods: The present study was conducted on 30 patients with KOA. They received weekly intra-articular ozone injections for 8 sessions for ozone injections, and their pain and functional outcomes were measured using the Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) Pain Inventory. In this study, SPSS was used, with a significance of $p< 0.05$. Baseline and post-treatment scores after one month were compared.

Results: The study involved 30 participants aged 71.80±8.38 *yrs*, with a majority of females (93. 3%). The WOMAC pain subscale was used to measure pain severity before and after ozone therapy. Significant improvements post-treatment, decreasing pain scores from 63.77±9.84 to 37.81±14.57 were observed.

Conclusion: Physical function and stiffness components also improved. The WOMAC Pain Questionnaire showed a significant reduction in pain intensity, indicating a substantial and reassuring improvement in pain management after ozone therapy. These results provide confidence and assurance regarding the possibility of ozone therapy. According to the existing results, the effectiveness of intra-articular ozone injection was significant. Therefore, ozone can be recommended as an efficient, durable treatment for mild or moderate KOA.

Keywords: Knee osteoarthritis, Ozone therapy, Pain

Introduction

Knee Osteoarthritis (KOA) is a degenerative joint disease that affects millions of people worldwide, especially those over 50 (1). It causes pain, stiffness, and reduced mobility due to the gradual deterioration of articular cartilage. The disease is a leading cause of disability worldwide, increasing due to aging populations and obesity. Addressing this degenerative joint condition is crucial for its adverse effects on quality of life (2-4).

Traditional treatment for KOA includes pharmacological interventions, such as Nonsteroidal Anti-Inflammatory Drugs (NSAIDs), physical therapy, and lifestyle modifications. Ozone therapy offers an alternative to traditional treatments for KOA, especially for patients who do not respond to or experience side effects from conventional options. However, these treatments often provide temporary relief and do not address the underlying degenerative process (5). As the global population ages, mild to moderate KOA increases, necessitating surgical interventions like total knee arthroplasty in advanced cases (6,7).

KOA symptoms are typically managed with NSAIDs, but these can be limited and have adverse effects. Ozone therapy, a minimally invasive treatment, offers a more effective and minimally invasive approach. Ozone improves oxygen delivery, reduces oxidative stress, and promotes tissue regeneration, offering a promising alternative (7,8). Knee arthroplasty, a surgical intervention for severe osteoarthritis, poses risks, particularly in older populations. Ozone, a molecule with anti-inflammatory, analgesic, and regenerative properties, is being utilized in ozone therapy, which is emerging as an alternative treatment for KOA (8).

Ozone injections have been investigated for their ability to reduce pain, improve joint function, and delay KOA progression. The mechanism is unclear, but it is believed to stimulate antioxidant defenses, reduce oxidative stress, and modulate joint inflammatory responses, providing a theoretical understanding of its possible efficacy (9,10).

Numerous studies have investigated ozone therapy for KAO. Raeissadat *et al* conducted a systematic review and meta-analysis highlighting the possible benefits of intra-articular ozone therapy in reducing pain and improving function (11). Similarly, Abreu Barbosa *et al* observed that ozone therapy led to significant functional improvements and symptom relief (12). This study demonstrated that ozone therapy activates antioxidant defenses, modulates inflammatory pathways, and promotes cartilage repair (13).

However, despite these promising findings, significant gaps remain in the existing literature. Most studies have been conducted in non-Iranian populations, leaving a lack of data on the regional efficacy of ozone therapy in Iran. Moreover, previous research focused on longer follow-up durations or larger cohorts, while the effects of short-term ozone therapy on smaller, specific samples remain less explored. This study addresses these gaps by providing data on short-term outcomes in an Iranian population.

Materials and Methods

Study design

This cross-sectional analytical study, conducted in 2023 at the Mehr Private Pain Clinic in Guilan Province, Iran, evaluated the effectiveness of ozone therapy in KOA.

Study population

The study specifically involved 30 patients diagnosed with grade 2 or 3 KOA, according to the Kellgren-Lawrence grading system. This system assesses the severity of osteoarthritis based on X-ray findings, making the findings directly applicable to this patient group. The baseline in this study refers to the time of the first injection. Assessments using pain, stiffness, and physical function were measured using the full Western Ontario and McMaster Universities Osteoarthritis Index scale (WOMAC) and they were conducted at this point to evaluate the patients' knee function before starting ozone therapy. The follow-up evaluations were carried out one month after completing the treatment.

Before injection: measurements were taken immediately before the first ozone injection to establish the values. One month after the infusion: outcome variables were assessed one month following the ozone therapy injection to evaluate its effectiveness.

The participants were divided into two age groups (≤72 *yrs* and >72 *yrs*) and two weight groups (≤78

kg and >78 *kg*). The cut-off values for age and weight were determined based on the **median values of the study population**. This approach ensured a balanced distribution of the participants in both categories, allowing for a more meaningful comparison of the outcomes. Previous studies on KOA have also used median-based stratification to analyze treatment effects across different subgroups."*

Exclusion criteria for oral painkillers and physical therapy

The exclusion criteria in the current study included factors that could interfere with assessing ozone therapy's efficacy. Patients who were actively using oral painkiller agents or undergoing physical therapy were excluded to ensure that the observed outcomes could be attributed solely to ozone therapy. This decision minimized confounding factors and strengthens the validity of the findings. This aspect will be clarified in the study to provide better transparency.

Method and sample size

The study was a Cohort analytical design conducted in 2023 at the Mehr Pain Clinic in Guilan Province. Convenience sampling was used to recruit 30 patients with grade 2 or 3 KOA based on a sample size calculation with a 5% error margin and 10% attrition rate, referencing Mishra SK's research.

Inclusion Criteria: Patients with grade 2 or 3 KOA, aged 45 to 65, with crepitation during joint movement and joint stiffness lasting morning stiffness instead. Exclusion Criteria: Patients with cancer, diabetes, osteoporosis, pregnancy, balance disorders, or severe joint space narrowing.

The patients received ozone therapy and were assessed using the modified WOMAC scale, which was administered one month before and after treatment to evaluate knee function and completed one month after treatment to assess pain severity (11). All the data (age, gender, weight, WOMAC scores) were analyzed using SPSS software (version 22). The participants were not financially burdened, and the university's ethics committee approved the study.

Ozone Preparation Details and Device Properties

The ozone used in this study was prepared using a medical-grade ozone generator (Ozomed® smartline machine (Kastner-Praxisbedarf GmbH, Medizintechnik, Germany)which ensured precise ozone concentration and purity. The ozone concentration was standardized at (Ozone therapy: 5 *ml* of ozone at a concentration of 30 *μ/ml* was injected into the joint along with half a cc of 2% lidocaine.). These characteristics are critical for maintaining the reproducibility and effectiveness of ozone injections, as supported by prior studies.

The choice of 8 sessions for ozone injections was based on prior clinical trials and evidence suggesting that this regimen balances effectiveness and patient compliance (11) and have shown significant improvements in pain and function with similar session numbers. This protocol was consistent with current recommendations for achieving optimal therapeutic outcomes. This rationale is expanded in the discussion section of the manuscript.

The pain clinic assesses KAO using the Kallgren-Lawrence classification. The afflicted joint is segmented into five regions (4-0). Classification criteria included the presence of osteophytes, joint space constriction, and subchondral sclerosis.

Due to the unavailability of the pre-treatment data, the improvement in patient outcomes was assessed based on post-treatment scores, compared with typical clinical expectations and historical data for the similar treatments.

Results

In this prospective study, 30 patients with KAO were referred to the Guilan Province Pain Clinic in 2023. The average age of the 30 patients was 72. Majority of the participants were female. The average weight of the patients was 78 *kg*. The lightest patient weighed 56 *kg*, and the heaviest patient weighed 96 *kg*. The patient's knee function was assessed using the WOMAC index before and one month after ozone therapy. The WOMAC score measures three subdomains: pain, stiffness, and physical function. The analysis indicated a significant improvement in all domains after treatment.

The age and weight cut-off points were selected based on the median values of the sample population (72 *yrs* for age and 78 *kg* for weight). This method was chosen to ensure that the participants were evenly distributed across the groups, reducing bias

and improving statistical reliability. Similar cut-off strategies have been employed in prior studies assessing osteoarthritis treatment outcomes. The initial mean total WOMAC score was 81.37±14.57. One month after ozone therapy, the mean WOMAC score significantly decreased to 39.67±14.25. The average pain score was 22.57±6.08 before treatment and declined to 10.93±4.83 following the ozone therapy. The mean stiffness score before treatment was 4.13±2.03, which reduced significantly to 2.00±1.46 after the therapy, indicating a notable improvement in joint stiffness. Regarding the physical function, the pre-treatment mean score was 41.47±7.79, which improved to 19.63±7.18 after the therapy (Table 1).

However, based on the post-treatment mean score of 37.57, there was a clear indication of pain relief compared to standard levels. A comparison of pain intensity after treatment showed a significant reduction in perceived pain. For patients younger than 72 *yrs*, the mean WOMAC score before ozone therapy was 79.20±15.49, which decreased to 35.00±9.57 one month after treatment. This change was statistically significant (p=0.001). For patients older than 72 *yrs*, the mean WOMAC score before therapy was 83.53±13.77, which dropped to 44.33±16.81 after one month of treatment. This difference was also statistically significant (p=0.001) (Table 2).

In the male patients, the mean WOMAC score before ozone therapy was 48.50±4.95, which decreased to 19.50±6.36 one month after treatment. However, this difference was not statistically significant (p=0.180). In the female patients, the mean WOMAC score before ozone therapy was 83.71±11.89, which reduced to 41.11±13.58 after one month. This change was statistically significant (p<0.001) (Table 3).

Or patients weighing less than 78 *kg*, the mean WOMAC score decreased from 83.07±13.39 before ozone therapy to 41.47±15.21 one month after the treatment, with a statistically significant difference (p=0.001). For patients weighing more than 78 *kg*,

Table 1. WOMAC knee function score and its subscales before and one month after ozone therapy

	WOMAC knee function score	Mean±S.D	Median (IQR)	Min	Max	p-value
Symptom score	Before ozone therapy	13.20±2.52	13(11.75,14.25)	8	19	p<0.001
	One month after ozone therapy	7.10±2.62	7(5,9)	3	13	
Stiffness score	Before ozone therapy	4.13±2.03	5(3,6)	0	6	p<0.001
	One month after ozone therapy	2.00±1.46	2(1.5,2)	0	5	
Pain score	Before ozone therapy	22.57±6.08	23.5(19,26.25)	9	35	p<0.001
	One month after ozone therapy	10.93±4.83	10(9,13.25)	2	23	
Daily life fnction score	Before ozone therapy	41.47±7.79	40.5(38,45.25)	22	56	p<0.001
	One month after ozone therapy	19.63±7.18	18(15.75,22.25)	7	38	
Total WOMAC score	Before ozone therapy	81.37±14.57	81(74.75,89.75)	45	111	p<0.001
	One month after ozone therapy	39.67±14.25	38(31.75,44)	15	78	

Table 2. WOMAC based on ege

	WOMAC knee function score by age	Mean±S.D	Median (IQR)	Min	Max	p-value
Younger than 72 *yrs*	Before ozone therapy	79.20±15.49	76(67,88)	52	110	p=0.001
	One month after ozone therapy	35.00±9.57	38(26,43)	15	49	
Older than 72 *yrs*	Before ozone therapy	83.53±13.77	84(79,92)	45	111	p=0.001
	One month after ozone therapy	44.33±16.81	40(34,50)	24	78	

Table 3. WOMAC based on gender

	WOMAC knee function score by gender	Mean±S.D	Median (IQR)	Min	Max	p-value
Male	Before ozone therapy	48.50±4.95	48.5(45,52)	45	52	p=0.180
	One month after ozone therapy	19.50±6.36	19.5(15,24)	15	24	
Female	Before ozone therapy	83.71±11.89	82.5(76.2,91.2)	61	111	p<0.001
	One month after ozone therapy	41.11±13.58	39(33.2,46)	25	78	

Table 4. WOMAC based on weight

	WOMAC knee function score by weight	Mean±S.D	Median (IQR)	Min	Max	p-value
Less than 78 *kg*	Before ozone therapy	83.07±13.39	81(74,92)	61	111	p=0.001
	One month after ozone therapy	41.47±15.21	38(28,48)	26	78	
More than 78 *kg*	Before ozone therapy	79.67±15.95	81(77,86)	45	110	p=0.001
	One month after ozone therapy	37.87±13.51	38(33,43)	15	75	

the mean WOMAC score dropped from 79.67±15.95 to 37.87±13.51, showing a statistically significant change (p=0.001) (Table 4).

The results demonstrate that ozone therapy effectively reduces joint stiffness. Ozone therapy significantly improves knee function and reduces pain in grade 2 and 3 osteoarthritis patients. The reduction in WOMAC scores was based on a single assessment using one month after injection. While this provides an initial picture of the patient's pain experience, it is essential to recognize that pain levels fluctuate. A more comprehensive assessment would require repeated assessments over time to confirm the sustained effectiveness of the treatment. In this case, the initial data show a significant reduction, but further studies with multiple assessments are recommended to assess the long-term benefits of the intervention more accurately.

Discussion

The improved WOMAC questionnaire evaluates knee functionality and the influence of osteoarthritis treatment on the quality of life. Results indicate a substantial enhancement in pain levels and knee functionality, consistent with other research highlighting the beneficial effects of ozone therapy (14-16).

Niknejad Hosseini validated the Persian version of this questionnaire's validity and reliability in a PhD project focused on the translation's efficacy in patients with KAO (17).

Raeissadat *et al* conducted a systematic review and meta-analysis that highlighted the possible benefits of intra-articular ozone therapy in patients with KAO (18). Moreover, the study by Abreu Barbosa demonstrated similar improvements in functional and symptomatic outcomes, further supporting the current study's conclusions (11,12).

Ozone therapy seems to exert its effects by activating antioxidant defenses and modulating inflammatory processes. Bocci *et al* discussed the physiological responses to ozone and noted its ability to increase oxygen metabolism and stimulate the release of growth factors involved in tissue repair. These mechanisms likely contribute to the improvements observed in patients with KOA, particularly in reducing oxidative stress and enhancing cartilage repair (13).

These findings are consistent with previous research, including Mishra, who observed similar benefits in KOA patients treated with intra-articular ozone (11). Moreover, studies such as that by Elvis and Ekta support the notion that ozone therapy modulates inflammatory pathways, enhancing oxygen metabolism and tissue repair (19).

Additionally, Bocci explained the dual role of ozone in promoting oxidative stress and inducing antioxidant defenses, which could explain its therapeutic effects in chronic degenerative conditions like osteoarthritis. This mechanism is further supported by its ability to increase levels of enzymes such as catalase and

Superoxide Dismutase (SOD), contributing to reduced oxidative damage in the joint (20).

Conclusion

According to the findings, intra-articular ozone injection significantly improved pain and knee function in patients with mild to moderate KAO during the one-month follow-up period. However, the absence of a control group and the limited follow-up duration are significant limitations. This study contributes to the existing literature by providing data on the short-term effects of ozone therapy in an Iranian population, where evidence has been limited. Future studies with larger sample sizes, control groups, and extended follow-up durations of one year or more to evaluate ozone therapy's long-term efficacy and safety are recommended.

Limitations

The most significant limitation is the absence of a control group, which restricts the ability to compare the effectiveness of ozone therapy with other treatments or a placebo. Additionally, due to the study design and data collection constraints, pre-treatment data were not available for all the patients, which may affect the accuracy of before-after comparisons. Another limitation is the sample size, which, despite being sufficient for initial analysis, may limit the generalizability of the findings to larger populations. Further studies with a randomized controlled trial (RCT) design and a larger sample size are recommended to validate these findings and provide stronger clinical evidence.

Future studies

Future studies should examine the long-term effects of ozone therapy through follow-up assessments to determine its lasting impact. Using more extensive and diverse sample sizes could improve the accuracy and generalizability of the findings. Studies comparing ozone therapy to other treatments can help assess its relative effectiveness.

Ethical approval

This study was conducted by the ethical principles outlined in the Declaration of Helsinki. The Ethics Committee of Guilan University of Medical Sciences (IR.GUMS.REC.1402.332) reviewed and approved the research protocol and procedures involving human participants. These ethical guidelines protect the study participants' rights, welfare, and well-being.

Funding

The author(s) received no specific funding for this work.

Acknowledgement

The authors would like to thank the Razi Clinical Research Development Unit.

Conflict of Interest

There was no conflict of interest in this manuscript.

References

1. Kang Y, Liu C, Ji Y, Zhang H, Wang Y, Bi W, et al. The burden of knee osteoarthritis worldwide, regionally, and nationally from 1990 to 2019, along with an analysis of cross-national inequalities. Arch Orthop Trauma Surg 2024;144(6):2731-43.

2. Hunter DJ, Bierma-Zeinstra S. Osteoarthritis. Lancet 2019;393(10182):1745-59.

3. Tehrani-Banihashemi A, Davatchi F, Jamshidi AR, Faezi T, Paragomi P, Barghamdi M. Prevalence of osteoarthritis in rural areas of Iran: a WHO-ILAR COPCORD study. Int J Rheum Dis 2014;17(4):384-8.

4. Zhang Y, Jordan JM. Epidemiology of osteoarthritis. Clin Geriatr Med 2010;26(3):355-69.

5. Lino VTS, Marinho DS, Rodrigues NCP, Andrade CAF. Efficacy and safety of ozone therapy for knee osteoarthritis: an umbrella review of systematic reviews. Front Physiol 2024;15:1348028.

6. Hochberg MC, Altman RD, April KT, Benkhalti M, Guyatt G, McGowan J, et al. American College of Rheumatology 2012 recommendations for the use of nonpharmacologic and pharmacologic therapies in osteoarthritis of the hand,

hip, and knee. Arthritis Care Res (Hoboken) 2012;64(4):465-74.

7. McAlindon TE, Bannuru RR, Sullivan MC, Arden NK, Berenbaum F, Bierma-Zeinstra SM, et al. OARSI guidelines for the non-surgical management of knee osteoarthritis. Osteoarthritis Cartilage 2014;22(3):363-88.

8. Losina E, Thornhill TS, Rome BN, Wright J, Katz JN. The dramatic increase in total knee replacement utilization rates in the United States cannot be fully explained by growth in population size and the obesity epidemic. J Bone Joint Surg Am 2012;94(3):201-7.

9. Veronese N, Cooper C, Reginster JY, Hochberg M, Branco J, Bruyère O, et al. Type 2 diabetes mellitus and osteoarthritis. Semin Arthritis Rheum 2019;49(1):9-19.

10. Teplyakova O, Vinnik Y, Drobushevskaya A, Malinovskaya N, Kirichenko A, Ponedelnik D. Ozone improved the wound healing in type 2 diabetics via down-regulation of IL- 8, 10 and induction of FGFR expression. Acta Biomed 2022;93(2):e2022060.

11. Mishra SK, Pramanik R, Das P, Das PP, Palit AK, Roy J, et al. Role of intra-articular ozone in osteo-arthritis of knee for functional and symptomatic improvement. Ind J Phys Med Rehabil 2011;22(2):65-9.

12. Abreu Barbosa P, Dias de Araújo LB, Lisboa Cordeiro AL. Effects of ozone therapy on joint rehabilitation in patients with osteoarthritis: a systematic review. Fisioterapia Brasil 2023;24(5).

13. Bocci V, Zanardia I, Valacchi G, Borrelli E, Travagli V. Validity of Oxygen-Ozone therapy as integrated medication form in chronic inflammatory diseases. Cardiovasc Hematol Disord Drug Targets 2015;15(2):127-38.

14. Xie F, Li SC, Goeree R, Tarride JE, O'Reilly D, Lo NN, et al. Validation of Chinese Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) in patients scheduled for total knee replacement. Qual Life Res 2008;17(4):595-601.

15. McConnell S, Kolopack P, Davis AM. The Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC): a review of its utility and measurement properties. Arthritis Rheum 2001;45(5):453-61.

16. Hawker G, Melfi C, Paul J, Green R, Bombardier C. Comparison of a generic (SF-36) and a disease specific (WOMAC) (Western Ontario and McMaster Universities Osteoarthritis Index) instrument in the measurement of outcomes after knee replacement surgery. J Rheumatol 1995;22(6):1193-6.

17. Eftekharsadat B, Niknejad-Hosseyni S, Babaei-Ghazani A, Toopchizadeh V, Sadeghi H. Reliability and validity of Persian version of Western Ontario and McMaster Universities Osteoarthritis index in knee osteoarthritis. J Analytic Res Clin Med 2015;3:170-7.

18. Raeissadat SA, Tabibian E, Rayegani SM, Rahimi-Dehgolan S, Babaei-Ghazani A. An investigation into the efficacy of intra-articular ozone (O(2)-O(3)) injection in patients with knee osteoarthritis: a systematic review and meta-analysis. J Pain Res 2018;11:2537-50.

19. Elvis A, Ekta J. Ozone therapy: a clinical review. J Nat Sci Biol Med 2011;2:66-70.

20. Bocci VA. Scientific and medical aspects of ozone therapy. State of the art. Arch Med Res 2006;37(4):425-35.