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ORIGINAL ARTICLE

Exploring the Relationship Between Primary Knee Osteoarthritis Severity and the Lipid Peroxidation Biomarker (MDA) in Synovial Fluid: A Preliminary Investigation

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ABSTRACT:
Background:Osteoarthritis (OA) is a gradually advancing and degenerative condition characterized by joint discomfort, tenderness, stiffness, locking, fluid buildup, diminished range of motion, swelling, crepitus, and impairment. The predominant clinical aspect of OA is the pain, which significantly influences functionality, mobility, overall quality of life, and often prompts individuals to seek medical guidance.**Methods**:A cross-sectional study was conducted in a hospital setting, focusing on patients with primary knee osteoarthritis. A total of 100 individuals, aged between 45 and 90 years, were randomly selected for the research, comprising 52 females and 48 males. The diagnosis of osteoarthritis was established using the American College of Rheumatology's Diagnostic criteria, while the severity of pain was assessed using a visual analogue scale. The Kellgren-Lawrence (K-L) radiographic assessment method was employed to grade knee osteoarthritis. The levels of Malondialdehyde (MDA) in the synovial fluid of all 100 individuals were determined using the Thiobarbituric acid technique. The study aimed to explore the relationship between the severity of knee osteoarthritis, oxidative stress markers, and synovial fluid MDA levels, investigating the potential link between oxidative stress-induced damage and the development of the disease.**Results**:The MDA values in synovial fluid for knee osteoarthritis grades 1, 2, 3, and 4 were recorded as 3.9±0.4, 4.3±0.5, 5.4±0.2, and 5.96±0.2, respectively. There was a statistically significant increase in MDA mean levels in synovial fluid with the progression of knee osteoarthritis severity, as determined by the Kellgren-Lawrence grading ($p < 0.001$).**Conclusion**:There was a positive association between Kellgren-Lawrence grading and synovial MDA levels. In patients with osteoarthritis, there was a notable elevation in free radical-induced lipid peroxidation, as indicated by the concentration of synovial fluid MDA, and this elevation correlated with the severity of osteoarthritis. These findings suggest the significance of oxidative stress in the etiopathogenesis of osteoarthritis, and imply that synovial MDA may serve as a potential biomarker for assessing the severity of the condition.
Keywords:osteoarthritis,peroxidation,synovial.

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INTRODUCTION

Osteoarthritis (OA) is a complex and widespread musculoskeletal condition, ranking as the 50th most common consequence of diseases and injuries on a global scale. It affects more than 4% of the world's population and is characterized by the gradual deterioration of joint cartilage, leading to changes in the underlying bone. This chronic condition imposes a significant burden on public health, contributing substantially to the nonfatal disease burden worldwide.[1,2]Knee osteoarthritis, constituting a substantial 85% of the most prevalent type of arthritis, holds a prominent position in the landscape of musculoskeletal disorders. Its prevalence is notably high, particularly among the elderly population in India, ranging from 22% to 39%. The knee, being a weight-bearing joint, is particularly susceptible to the degenerative changes associated with osteoarthritis, and this prevalence underscores its significance as a leading cause of disability, chronic pain, and overall morbidity.

The burden of knee osteoarthritis is amplified in the elderly, where the natural aging process may exacerbate joint degeneration. The impact on mobility, independence, and overall quality of life is profound, making it a primary contributor to disability in this demographic.[3] Furthermore, the prevalence and impact of osteoarthritis are not uniform across genders, with women facing a higher susceptibility. Approximately 45% of women over the age of 62 report symptomatic manifestations of knee osteoarthritis, and an astonishing 70% exhibit radiological evidence of the condition. This gender-specific predilection underscores the need for tailored approaches in understanding, preventing, and managing osteoarthritis.Osteoarthritis, with its intricate and multifactorial etiology, remains a disorder of unknown origin. This complexity makes it challenging to pinpoint specific causes, and the

condition manifests across various joints in the body.[4,5] Consequently, osteoarthritis stands as a common cause of disability, necessitating a comprehensive and integrated approach that encompasses medical interventions, lifestyle modifications, and public health initiatives.In conclusion, osteoarthritis, particularly knee osteoarthritis, is a pervasive and impactful health concern that warrants attention on both individual and societal levels. Understanding its prevalence, especially in the context of an aging population, and recognizing the gender-specific patterns of susceptibility are crucial for developing effective strategies aimed at prevention, early intervention, and improving the overall well-being of those affected by this debilitating condition.Osteoarthritis (OA) is a polygenic and complex illness with intricate origins. The development of OA involves the interplay of multiple genetic and environmental factors, which collectively contribute to its etiology. These factors are intricately connected to the activation of molecular pathways that play a pivotal role in the progression of articular damage, a hallmark feature of this degenerative condition.OA manifests as a degenerative joint disorder that leads to a spectrum of symptoms over time. These symptoms include pain, stiffness, effusion (accumulation of fluid in the joint), limited range of motion, swelling (oedema), crepitus (the sensation of grinding or popping in the joint), and gradual impairment of joint function. The cumulative impact of these symptoms significantly affects the quality of life for individuals grappling with OA, especially as the condition progresses.[6,7]Among the various clinical manifestations, osteoarthritic pain stands out as the most prominent and characteristic symptom. This pain, often described as a dull, aching discomfort, is a pervasive and challenging aspect of OA. It tends to worsen with joint use and is a key driver of the functional limitations experienced by individuals with OA. The nature and intensity of osteoarthritic pain can vary, influencing not only physical well-being but also mental and emotional aspects of an individual's health.The complex interplay of genetic predispositions and environmental factors in OA underscores the need for a comprehensive understanding of its pathogenesis. Molecular pathways involved in the advancement of articular damage provide valuable insights into potential targets for therapeutic interventions. Strategies aimed at alleviating symptoms, slowing disease progression, and improving overall joint function are critical in managing the multifaceted impact of osteoarthritis on affected individuals.In summary, osteoarthritis is a multifaceted condition influenced by both genetic and environmental factors. The activation of molecular pathways contributes to articular damage, resulting in a spectrum of symptoms that define the clinical presentation of OA. Osteoarthritic pain, a central and pervasive characteristic, underscores the importance of developing targeted interventions to enhance the quality of life for individuals navigating the challenges posed by this degenerative joint disorder.

Osteoarthritis (OA) is characterized by a spectrum of changes at the morphological, biochemical, molecular, and biomechanical levels, affecting both cells and the extracellular matrix (ECM). These changes culminate in a series of alterations, including softening, fibrillation, ulceration, and the eventual loss of articular cartilage. Synovial inflammation, sclerosis of subchondral bone, the formation of osteophytes, and subchondral cysts further contribute to the complexity of the condition.[8] Among these changes, the knee emerges as the most clinically significant site affected by osteoarthritis.This joint disorder represents a complex and heterogeneous form of joint deterioration. Current theories on the pathogenesis of OA propose a disruption in the homeostatic balance between the degradation and synthesis of bone and cartilage. Within this context, previous research has identified oxidative stress as a crucial factor in the development and progression of OA. Reactive oxygen species (ROS), including nitric oxide, superoxide anion, hydrogen peroxide, and hydroxy radical, play a central role in the pathogenic process.Oxidative stress involves highly reactive chemical compounds, such as ROS, that target molecules like proteins, lipids, and nucleic acids, causing cellular damage. This damage leads to structural and functional alterations in chondrocytes, the extracellular matrix, and overall tissue integrity, all of which contribute to the etiology of OA. Notably, oxidative damage occurs only when there is a disruption in the antioxidant system, and ROS generation surpasses the capacity of antioxidants to neutralize them.Halting the progression of osteoarthritis poses a significant challenge due to its multifaceted nature. Understanding the role of oxidative stress and the intricate interplay between reactive oxygen species and the antioxidant defense system is pivotal for developing targeted therapeutic strategies. Addressing the oxidative imbalance may provide avenues for interventions aimed at preserving joint structure and function in individuals affected by osteoarthritis.[9]In summary, osteoarthritis involves a cascade of changes encompassing cellular, molecular, and biomechanical levels, leading to the characteristic alterations in joint structure. The knee stands out as a particularly significant site of involvement. The disruption in the delicate balance between bone and cartilage homeostasis, coupled with oxidative stress, contributes to the intricate pathogenesis of OA, highlighting the need for targeted interventions to mitigate its impact on affected individuals.

MATERIALS AND METHODS

This study employs a hospital-based cross-sectional observational design, aiming to explore various aspects related to patients attending the Orthopaedics outpatient and inpatient department within a specified study period. The Institutional Ethics Committee has

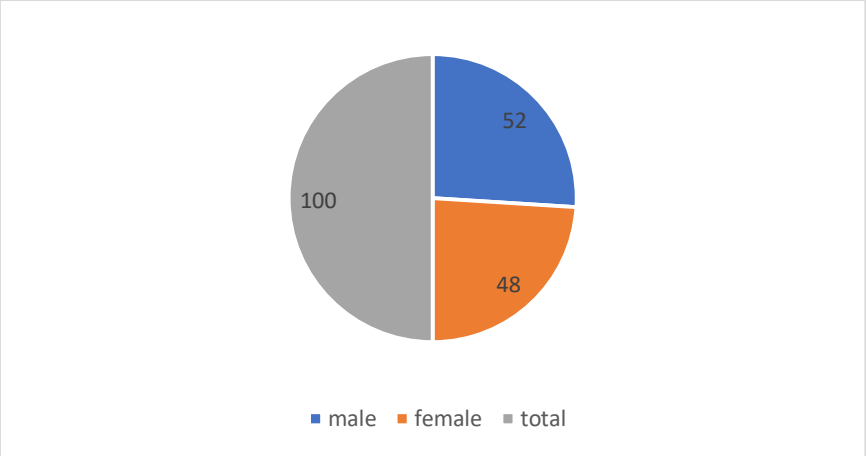
granted approval, underscoring the commitment to ethical standards, and informed written consent has been obtained from all participating patients, ensuring transparency and respect for individual autonomy.In terms of the sample size, the study initially enrolled 120 patients from the Orthopaedics department. However, 10 patients were subsequently excluded from the analysis. This exclusion involved 4 patients diagnosed with Rheumatoid arthritis, 4 with diabetes mellitus, and 2 with joint infections. The final cohort considered for analysis comprises the remaining eligible participants, reflecting a comprehensive representation of the patient population seeking orthopedic care at the hospital.The sampling method employed for participant selection is consecutive sampling, a systematic approach that enrolls individuals in sequence based on their arrival or availability. This method ensures an unbiased representation of patients seeking orthopedic care, contributing to the study's overall reliability and generalizability of findings. The combination of a robust study design, ethical considerations, and a well-defined sampling approach enhances the credibility and validity of the research outcomes.

This study has delineated specific inclusion and exclusion criteria to define the target population for comprehensive investigation. The inclusion criteria encompass patients aged 42 years and older exhibiting acute symptoms of osteoarthritis, particularly knee effusion. Additionally, the study includes individuals undergoing intra-articular pharmacological injection therapy, those scheduled for knee replacement procedures, and participants undergoing arthroscopic lavage for the management of their osteoarthritic symptoms. By focusing on this demographic and therapeutic interventions, the study aims to provide valuable insights into the acute manifestations and treatment modalities of osteoarthritis in an older population.

Conversely, the exclusion criteria are designed to maintain the study's specificity and ensure a more homogeneous participant group. Patients with a history of previous surgery on the same joint are excluded, as are those with inflammatory joint diseases. The criteria extend to individuals using steroids or other long-term medications and those experiencing pain resulting from a traumatic event. Furthermore, patients with systemic disorders, such as significant liver, renal, or heart diseases, which may contribute to elevated oxidative stress, are also excluded. These criteria collectively refine the study population, emphasizing a targeted investigation into acute osteoarthritis symptoms and related interventions while minimizing confounding variables from diverse medical backgrounds or treatments.

RESULTS

Figure1: Gender distribution of patients



The research included a total of 100 individuals, ranging in age from 45 to 90 years. The selection of participants was conducted through a random sampling process, ensuring a representative and unbiased sample for the study. Within this cohort, there were 52 females and 48 males, providing a gender-balanced representation. This demographic information offers a snapshot of the diverse age range and gender distribution within the study population, facilitating a more comprehensive analysis of the research outcomes. Random selection helps mitigate potential biases and ensures that the findings are more likely to be reflective of the broader population from which the sample was drawn.

Table 1: Mean value of age, MDA, VAS score & duration of disease according to K-L grading

Grading of Osteoarthritis	Case (n)	Age (years)	MDA (µml/L)	Vas score	Duration of disease (years)
Grade 1 (Dublous)	24	46.09	4.01	4.83	1.5
Grade 2 (Mild)	26	50.19	4.18	6.38	2.25
Grade 3 (Moderate)	30	60.3	5.4	7.21	5.67

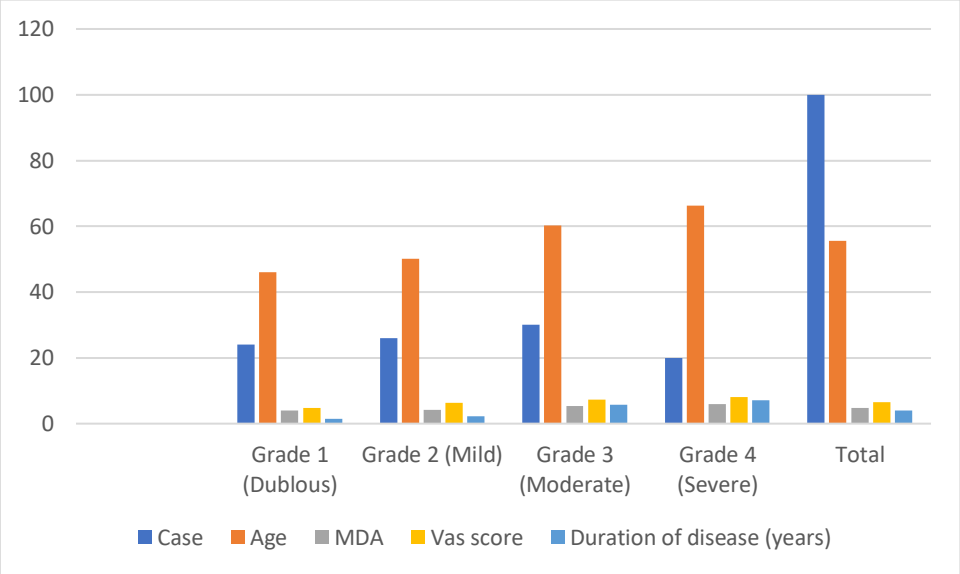
Grade 4 (Severe)	20	66.27	5.93	8	7.1
Total	100	55.57	4.86	6.6	4.07

The presented table outlines the grading of osteoarthritis cases, offering a detailed breakdown across different severity grades along with relevant demographic and clinical parameters. In Grade 1 (Dubious), characterized by a milder form of the condition, 24 cases were observed with an average age of 46.09 years, a mean MDA (markers of disease activity) of 4.01 µml/L, a Vas score indicating pain intensity at 4.83, and an average disease duration of 1.5 years. Grade 2 (Mild) comprised 26 cases, featuring an average age of 50.19 years, an MDA of 4.18 µml/L, a Vas score of 6.38, and a disease duration of 2.25 years.

Moving to Grade 3 (Moderate), which indicates a more advanced stage of osteoarthritis, there were 30 cases with an average age of 60.3 years, an elevated MDA of 5.4 µml/L, a Vas score of 7.21, and a longer average disease duration of 5.67 years. In Grade 4 (Severe), characterized by the most advanced stage of the condition, 20 cases were identified, featuring an average age of 66.27 years, a higher MDA of 5.93 µml/L, a Vas score at 8, and a more extended average disease duration of 7.1 years.

The cumulative data for all grades, summarized in the "Total" row, provide an overarching perspective on the entire cohort of 100 cases. The average age of 55.57 years, an MDA of 4.86 µml/L, a Vas score of 6.6, and an average disease duration of 4.07 years offer a composite overview of the collective demographic and clinical characteristics. This comprehensive analysis contributes valuable insights into the interplay between severity grades of osteoarthritis and associated parameters, facilitating a deeper understanding of the heterogeneity within this patient population.

Figure 2: Mean value of age, MDA, VAS score & duration of disease according to K-L grading



The study involved 100 subjects, who were categorized into four groups based on the Kellgren-Lawrence (K-L) grading system for primary knee Osteoarthrosis (KOA). The analysis revealed a notable association between K-L grading and several key parameters. As the K-L grading of osteoarthritis increased, there was a corresponding increase in the mean age of the subjects, highlighting a potential correlation between age and the severity of KOA.Additionally, parameters such as malondialdehyde (MDA) concentration, Visual Analog Scale (VAS) scores, and disease duration exhibited a consistent trend. Specifically, the mean MDA concentration (measured in µM/L) demonstrated a progressive rise with higher K-L grading. This suggests a potential link between oxidative stress, reflected by MDA levels, and the advancing severity of knee osteoarthritis, as indicated by the K-L grading.

The observed trends in VAS scores and disease duration align with the notion that as the severity of KOA increases, patients tend to experience higher levels of pain (reflected in VAS scores) and endure longer disease durations. These findings contribute valuable insights into the relationships between age, disease severity, and associated factors in primary knee Osteoarthrosis, offering a more nuanced understanding of the progression and impact of KOA.

DISCUSSION

The research in question employed the Kellgren-Lawrence (K-L) grading system as a pivotal tool for categorizing patients with knee osteoarthritis (KOA) based on the radiological severity of their condition. A

notable aspect that sets this study apart is its innovative use of synovial fluid from OA knees to conduct a detailed subgroup analysis of oxidative stress across various levels of knee osteoarthritis severity. This approach, to the best of our knowledge, represents a novel avenue of exploration, adding a distinctive dimension to the understanding of oxidative stress in the context of KOA.Primary knee osteoarthritis (KOA) emerges as a chronic condition with far-reaching implications, disrupting the delicate balance of cartilage metabolism. This disturbance leads to the gradual degradation of cartilage and, consequently, contributes to knee injury. The study underscores specific examples of reactive oxygen species (ROS), notably the involvement of superoxide anion, in mediating articular cartilage and joint destruction in the context of KOA.[10,11,12] Furthermore, the investigation highlights that oxidant levels within the synovial fluid of these patients are frequently found to be significantly elevated, indicating a potential role of oxidative stress in the pathogenesis of knee osteoarthritis.In the context of the current study, a particular focus was directed towards assessing the lipid peroxidation product, specifically the mean malondialdehyde (MDA) level. The study's findings revealed a noteworthy and statistically significant increase in the synovial fluid of patients diagnosed with Osteoarthritis of the Knee. This observation underscores the potential impact of oxidative stress on the biochemical milieu within the knee joint, particularly as it pertains to different severity levels of knee osteoarthritis. This novel exploration of oxidative stress markers in synovial fluid contributes substantially to our understanding of the biochemical intricacies associated with knee osteoarthritis, paving the way for further research and potential therapeutic insights.

The elevation in malondialdehyde (MDA) levels observed in the study points to an intensified generation of reactive oxygen species (ROS), implying an excess of oxidative damage within the studied patient group. ROS, including superoxide anion and hydrogen peroxide, are known for their ability to oxidize essential biomolecules, particularly membrane lipids. This oxidative stress-induced lipid peroxidation can have significant implications for cellular integrity and function.Consistent with the current findings, a study by Tanyawan S et al. corroborated the association between osteoarthritis (OA) and elevated plasma MDA levels when compared to a healthy control group. This supports the notion that the oxidative milieu is perturbed in OA, potentially contributing to the progressive degeneration of joint tissues.Shweta Dwivedi et al.'s research, encompassing both rheumatoid arthritis (RA) and osteoarthritis patients, further strengthens the link between oxidative stress and joint disorders.[13] The significantly higher serum MDA levels observed in these patient groups compared to controls underscore the systemic impact of oxidative stress in inflammatory joint conditions.The broader context of oxidative stress in joint disorders is elucidated by non-enzymatic interactions of oxygen with organic molecules, as highlighted in studies by Mezes M et al., Sarban S, Surapaneni KM et al., and Seven et al. These investigations consistently report higher MDA levels in both osteoarthritis and rheumatoid arthritis patients, emphasizing the shared mechanistic involvement of oxidative stress in these conditions.The present study's focus on synovial fluid from knee osteoarthritis patients reveals a substantial increase in mean MDA levels. Synovial fluid is integral to joint health, and elevated MDA concentrations in this fluid further emphasize the localized impact of oxidative stress on joint tissues. The cumulative evidence across various studies suggests that oxidative stress is not merely a consequence but a key contributor to the pathogenesis of osteoarthritis.[14]In conclusion, the expanding body of evidence supports the premise that oxidative stress, as indicated by increased MDA levels, plays a pivotal role in the development and progression of osteoarthritis. Understanding these molecular dynamics may pave the way for targeted interventions aimed at mitigating oxidative damage and, consequently, alleviating the burden of joint disorders on affected individuals.

The initiation of this study was prompted by a keen interest in delving into the intricate interplay between lipid peroxidation in synovial fluid and the severity of primary knee osteoarthritis. The overarching goal was to unravel the nuanced dynamics of oxidative stress in individuals presenting with varying degrees of primary knee osteoarthritis, with an emphasis on understanding its potential implications for disease progression and management strategies.The assessment of lipid peroxidation in synovial fluid, specifically through the measurement of malondialdehyde (MDA) levels, provided a quantitative means to explore the probable redox imbalance that contributes to oxidative damage within the joint environment.[15] The positive association observed between Kellgren-Lawrence grading—an established system for categorizing osteoarthritis severity—and synovial MDA levels highlighted a robust and positive correlation between the extent of lipid peroxidation and the severity of the disease process. This linkage not only validates the role of oxidative stress in osteoarthritis but also positions synovial MDA as a potential biomarker to gauge the severity of this debilitating joint condition.Understanding that oxidative stress is a pivotal factor in the etiopathogenesis of osteoarthritis, the study proposes the exploration of exogenous antioxidant supplementation as a viable strategy to mitigate oxidative damage. The potential of antioxidants to alleviate free radical-mediated musculoskeletal tissue degradation in osteoarthritis is underscored by the observed correlations. This insight is particularly noteworthy, suggesting that antioxidant

interventions may prove beneficial in the early stages of osteoarthritis or as individuals age, thereby serving as a preventive measure against disease progression.The robust findings of this study not only contribute to the scientific understanding of the oxidative mechanisms underlying osteoarthritis but also hold practical implications for clinical practice. Clinicians are encouraged to consider antioxidant treatment, especially in the early stages of osteoarthritis or as patients age, as a promising avenue to prevent or attenuate oxidative stress-induced musculoskeletal tissue degradation.[16] This proactive approach may not only impact osteoarthritis management but also has broader implications for addressing age-related diseases where oxidative stress plays a pivotal role in the pathophysiology. The study's comprehensive exploration bridges the gap between bench research and clinical applications, offering a potential paradigm shift in the approach to managing osteoarthritis and related conditions associated with oxidative stress.The implications of antioxidant therapy in the early stages of osteoarthritis extend beyond the immediate management of symptoms, potentially offering a strategy to slow the progression of the disease. The recognition of oxidative stress as a significant contributor to osteoarthritis pathogenesis underscores the importance of interventions that target this underlying mechanism. By incorporating antioxidant therapy at an early stage, there is potential not only to alleviate symptoms but also to modulate the fundamental processes that drive the degeneration of joint tissues.Looking ahead, future research in this area is poised to provide a more nuanced understanding of the utility of synovial fluid malondialdehyde (MDA) levels as an early marker for measuring oxidative stress in the knee joint.[17] The choice to focus on synovial fluid, as opposed to serum markers, reflects a recognition of the localized impact of oxidative stress within the joint environment. Synovial fluid serves as a direct and proximal indicator of the biochemical milieu within the knee joint, offering insights into the specific oxidative processes occurring at the site of pathology.The transition from serum markers to synovial fluid MDA levels as an early indicator holds promise for more accurate and targeted assessments of oxidative stress in osteoarthritis. As synovial fluid is in direct contact with joint tissues, its analysis provides a more specific reflection of the oxidative microenvironment. Utilizing synovial fluid MDA levels as an early marker has the potential to enhance diagnostic precision and inform therapeutic decisions at a stage where interventions may be more impactful.In summary, the prospect of antioxidant therapy in the early stages of osteoarthritis presents a proactive approach to disease management, aiming not only to alleviate symptoms but to impede the underlying progression. The evolving focus on synovial fluid MDA as an early marker reflects a growing appreciation for the localized nature of oxidative stress in joint disorders, paving the way for more targeted and effective interventions in the future. Continued research endeavors in this realm will contribute to refining our understanding of the interplay between oxidative stress and osteoarthritis, ultimately offering new avenues for therapeutic innovation and improved patient outcomes.

CONCLUSION

In patients with osteoarthritis (OA), the study revealed a noteworthy elevation in free radical-induced lipid peroxidation, as assessed by synovial fluid malondialdehyde (MDA) concentration. Importantly, this lipid peroxidation increased in tandem with the severity of osteoarthritis, suggesting a progressive impact of oxidative stress on joint tissues. This observation underscores the significance of oxidative stress in the etiopathogenesis of OA, positioning synovial MDA as a potential biomarker for determining the severity of the disease. The positive connection observed between Kellgren-Lawrence grading—a measure of osteoarthritis severity—and synovial MDA levels further supports the association between oxidative stress and disease progression.The potential of antioxidant supplementation in early osteoarthritis patients emerges as a promising avenue to slow the evolution of the illness. By enhancing the antioxidant status of the knee, such interventions aim to counteract free radical production and mitigate the destructive effects on cartilage. This proactive approach aligns with the growing recognition of the role of oxidative stress in driving the degenerative processes seen in OA.In summary, the study's findings suggest that oxidative stress is a pivotal player in the development and progression of osteoarthritis. Synovial MDA emerges as a potential biomarker for assessing disease severity, and antioxidant supplementation represents a promising strategy to impede the evolution of OA. The imperative for further research underscores the ongoing efforts to translate these insights into clinically effective interventions for individuals grappling with the challenges of osteoarthritis.

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