Blocking extracellular Galectin-3 in patients with osteoarthritis

Alec R. Andrews a,1, Ana D. Fernandes b, Seth E. Brownmiller b, Yousif Hanna b, Mark C. Fisher b,1, Christene A. Huang a,c,*,1

a Center for Transplantation Sciences, Department of Surgery, Massachusetts General Hospital, Boston, MA, USA
b Department of Rheumatology, Massachusetts General Hospital, Boston, MA, USA
1 Department of Surgery, Division of Plastic and Reconstructive Surgery, Division of Transplant Surgery, University of Colorado School of Medicine, Aurora, CO, USA

ARTICLE INFO
Keywords: Osteoarthritis Galectin-3 Modified citrus pectin

ABSTRACT
Objective: This pilot clinical trial examined the efficacy of blocking extracellular Galectin-3 (Gal-3) with modified citrus pectin (MCP), in patients suffering from knee osteoarthritis (OA).
Methods: 50 patients were randomized in a 1:1 ratio to receive MCP or placebo at a dose of 4 g (5 capsules) twice daily for 12 weeks. Serum Gal-3 levels and OA severity were evaluated at baseline and 12 weeks. Gal-3 levels were detected by sandwich ELISA and OA severity was determined using WOMAC-knee, SF-36, and RAPID3 surveys during these visits. MCP tolerability was assessed by a basic metabolic panel during a week 6 follow up visit.
Results: Patients enrolled in both the MCP treatment and placebo groups shared similar baseline characteristics in OA severity, serum Gal-3 levels, and pain management. Improvement across all surveys was noted independent of supplement or placebo treatment. No significant change in Gal-3 levels were observed in either cohort over the 12-week study.
Conclusion: Treatment of knee OA with a 12-week course of MCP did not significantly improve disease burden compared to placebo.

1. Introduction

Knee osteoarthritis (OA) is a degenerative joint condition marked by cartilage loss, damage to supporting bone (including femur, patella, and tibia), and alterations in ligaments and vascular supply, culminating in a debilitating condition that inhibits proper movement. Repeated mechanical stress and injury contribute significantly to the onset of knee OA. However, clinical evidence suggests that chronic inflammation drives disease progression. [1,2]. Synovitis caused by lymphocyte infiltration of the synovial fluid is associated with advanced stages of OA. [3]. Accompanying synovial lymphocyte infiltration is an increase in inflammatory cytokines and proteolytic enzymes which are capable of damaging cartilage and remodeling the extracellular matrix, promoting the breakdown of knee joint components. [4,5].

Targeted therapies to resolve underlying inflammation are limited in OA, and current treatments such as NSAIDs and corticosteroids are ineffective in long term utilization due to associated side effects and a lack of compelling evidence supporting their benefit. [6]. Comorbidities commonly affecting the elderly population such as diabetes, cardiovascular disease, and gastrointestinal disorders further limit the pharmacologic options to address OA.

Galectin-3 (Gal-3) is a β-galactoside binding lectin with pleiotropic functions, both intra- and extracellularly. When Gal-3 is secreted into the extracellular environment by activated macrophages, induction of growth factor and pro-inflammatory signaling cascades initiate tissue fibrosis in chronic heart failure. [7]. Elevated serum Gal-3 levels correlate with numerous inflammatory and fibrotic conditions including cardiovascular disease, autoimmune diseases, cancer, and OA. [8-11]. Inhibitors of extracellular Gal-3, such as modified citrus pectin (MCP), have been proven to reduce inflammation in preclinical models. [12]. MCP is thought to disrupt the pro-inflammatory pathway through the binding of pectin-derived galactose chains to the carbohydrate recognition domain of Gal-3 in the extracellular space.

In this study, the efficacy of MCP was evaluated in a randomized,
A randomized, double blind, placebo controlled, clinical trial was conducted to assess the impact of MCP on knee OA symptoms. Patients were recruited from the rheumatology clinic and general population at Massachusetts General Hospital (MGH) through public posting of trial advertisement from January 2017 to February 2018. Eligibility criteria included patients between the age of 35–80 and those who experienced knee arthritis for at least 50% of the last month with a Kellgren and Lawrence (KL) Grade 2 or 3 classification of OA as determined by radiographic exam. Patients were omitted if they had active fibromyalgia, active inflammatory arthritis, no longer had native knee(s), or a KL Grade of 0, 1 or 4. Patient consent was obtained during the baseline visit after eligibility was confirmed. During this visit, study participants also completed WOMAC-Knee, SF-36, and RAPID3 surveys and a

| Table 1 |
|---------|
| Demographics, baseline and week 12 values. |
| MCP | Placebo |
| Age (years) | 62.2 | 63.7 | 0.6 |
| BMI (kg/m²) | 28.7 | 27.6 | 0.1 |
| Gender (% female) | 65.0 | 80.0 | 0.7 |
| NSAID use (%) | 60.0 | 50.0 | 1 |
| Tylenol use (%) | 20.0 | 10.0 | 1 |
| Neuropathic med use (%) | 10.0 | 10.0 | 1 |
| Supplement use (%) | 20.0 | 10.0 | 0.5 |
| RAPID3 (nu) | 14.1 | 13.9 | 0.8 |
| WOMAC-Knee (nu) | 32.1 | 34.8 | 0.5 |
| WOMAC-Pain (nu) | 7.1 | 7.7 | 0.6 |
| WOMAC-Stiffness (nu) | 2.9 | 3.8 | 0.7 |
| WOMAC-Function (nu) | 22.1 | 23.3 | 0.5 |
| Galectin-3 Level (ng/ml) | 6.3 | 6.5 | 0.8 |

| Week 12 |  |
|--------|--------|
| RAPID3 (nu) | 1.5 | 1.5 | 0.7 |
| WOMAC-Knee (nu) | 26.0 | 22.9 | 1 |
| WOMAC-Pain (nu) | 6.0 | 4.8 | 0.7 |
| WOMAC-Stiffness (nu) | 3.1 | 2.5 | 0.6 |
| WOMAC-Function (nu) | 16.9 | 15.5 | 1 |
| Galectin-3 Level (ng/ml) | 9.1 | 7.8 | 0.2 |
baseline serum was collected to assess Gal-3 level. MGH research pharmacy randomized 50 patients in blinded fashion to receive either MCP or placebo (25 participants in each group) at a dose of 4 g (5 capsules) twice daily for 12 weeks. A week 6 follow up visit was scheduled to assess adverse events and collect a basic metabolic panel. An additional 6-week course of treatment was dispensed to complete the study. Patients completed the study with a week 12 visit to assess efficacy as reported by WOMAC-Knee, SF-36, and RAPID3 surveys, in addition to a final Gal-3 serum level. Survey results are reported as normalized units (nu).

Chi-squared testing was performed to determine significance of categorical variables. Numerical variables were assessed by t-tests.

3. Results

A description of patient enrollment, group allocation, and the total number of study participants included in the final analysis are provided in Table 1. At baseline, there were no significant differences in age, gender, pain management, self-reported OA involvement, or Gal-3 levels between the MCP and placebo groups (Table 1).

Treatment with a 12-week course of MCP significantly improved OA symptoms as demonstrated by WOMAC, RAPID3 scoring (Table 2). However, a robust response with similar improvement occurred in the placebo group, SF-36 improvement was also noted in both groups (data not shown). Gal-3 levels slightly increased in both cohorts over the 12 weeks and no significant differences were detected when comparing MCP and placebo. Side effects were minimal, reported by 19% of patients in the MCP treatment arm and 14% of patients in the placebo arm. One case of hyperkalemia and one case of loose stool were potentially related to MCP treatment.

4. Discussion

The efficacy of MCP on OA symptoms was evaluated by changes in WOMAC-Knee and RAPID3 score. These patient reported outcome tools reliably gauge physiological burden of disease in OA patients, and their results are used by clinicians when prescribing OA treatment. [13]. Despite improvement across all surveys in the 12-week study, MCP treatment outcomes did not significantly differ from placebo. The small sample size in our pilot study possibly inflated the response to placebo, misrepresenting potential outcomes if this analysis was carried out in a larger population.

Baseline serum Gal-3 levels for the MCP and placebo group were 6.31 ng/ml and 6.51 ng/ml respectively. These values are considerably lower than those detected in patients with other fibrotic conditions. [14–16]. For example, patients who experienced an ST-elevation myocardial infarction (STEMI) 24–48 h before sample collection had an average serum Gal-3 level of 13.1 ng/ml (n = 96). [16]. The low baseline serum Gal-3 levels in this OA patient cohort likely reduced the therapeutic response to blocking Gal-3 function with MCP.

In conclusion, this study failed to show a benefit for knee OA with 12 weeks of supplementation with MCP compared to placebo.

Table 2

| MCP | Placebo | p-value |
|-----|---------|---------|
| Baseline | Week 12 | Δ | Baseline | Week 12 | Δ |
| RAPID3 (nu) | 14.1 | 1.5 | −12.5 | 13.9 | 1.5 | −12.5 | 0.7 |
| WOMAC-Knee (nu) | 32.1 | 26.0 | −6.1 | 34.8 | 22.9 | −11.9 | 1.0 |
| WOMAC-Pain (nu) | 7.1 | 6.0 | −1.1 | 7.7 | 4.8 | −2.9 | 0.7 |
| WOMAC-Stiffness (nu) | 2.9 | 3.1 | 0.2 | 3.8 | 2.5 | −1.2 | 0.6 |
| WOMAC-Function (nu) | 22.1 | 16.9 | −5.2 | 23.3 | 15.5 | −7.8 | 1.0 |
| Galectin-3 Level (ng/ml) | 6.3 | 9.1 | 2.8 | 6.5 | 7.8 | 1.3 | 0.2 |

Financial disclosure statement

The authors declare no conflicts of interest.

Trial Registration Number

NCT02800629

Acknowledgements

This study was supported through a pilot grant from the Harvard Football Players Health Study (FPHS). The FPHS is funded by a grant from the National Football League Players Association. The content is solely the responsibility of the authors and does not necessarily represent the official views of Harvard Medical School, Harvard University or its affiliated academic health care centers, the National Football League Players Association, or The Massachusetts General Hospital. The authors gratefully acknowledge EcoNugenics, Inc for the generous unrestricted research gift, including the MCP and matched placebo capsules used in this trial.

References

[1] C.L. Hill, D.J. Hunter, J. Niu, et al., Synovitis detected on magnetic resonance imaging and its relation to pain and cartilage loss in knee osteoarthritis, Ann. Rheum. Dis. 66 (2007) 1599–1603.
[2] M.J.M. Mineten, A. Blom, G.P. Snijders, et al., Galectin-3: a potential biomarker for monitoring disease activity in rheumatoid arthritis, J. Cell. Physiol. 210 (2006) 451–454.
[3] W.H. Robinson, C.M. Lepus, Q. Wang, et al., Low-grade inflammation as a key mediator of the pathogenesis of osteoarthritis, Nat. Rev. Rheumatol. 12 (2016) 580–592.
[4] J. Sellam, F. Berenbaum, The role of synovitis in pathophysiology and clinical symptoms of osteoarthritis, Nat. Rev. Rheumatol. 6 (2010) 625–635.
[5] J. Sokolove, C.M. Lepus, Role of inflammation in the pathogenesis of osteoarthritis: latest findings and interpretations, Ther Adv Musculoskelet Dis 5 (2013) 77–94.
[6] T.E. McAlindon, R.R. Bannuru, M.C. Sullivan, et al., OARSI guidelines for the non-surgical management of knee osteoarthritis, Osteoarthr. Cartil. 22 (2014) 363–388.
[7] U.C. Sharma, S. Pokharel, T.J. van Brakel, et al., Galectin-3 marks activated macrophages in failure-prone hypertrophied hearts and contributes to cardiac dysfunction, Circulation 110 (2004) 3121–3128.
[8] R.I. Lala, M. Puchita, D. Darabanti, L. Pilat, Galectin-3 in heart failure pathology—‘another brick in the wall’? Acta Cardiol. 70 (2015) 323–331.
[9] N.C. Henderson, T. Sethi, The regulation of inflammation by galectin-3, Immunol. Rev. 230 (2009) 160–171.
[10] V.L. Thijssen, R. Heuschen, J. Caers, A.W. Griffioen, Galectin expression in cancer diagnosis and prognosis: a systematic review, Biochim. Biophys. Acta 1855 (2015) 235–247.
[11] P. Reboul, J. Martel-Pelletier, J.P. Pelletier, Galectin-3 in osteoarthritis: when the fountain of youth doesn’t deliver its promises, Curr. Opin. Rheumatol. 16 (2004) 595–598.
[12] M. Kolatsi-Joannou, K.L. Price, P.J. Winyard, D.A. Long, Modified citrus pectin reduces galactin-3 expression and disease severity in experimental acute kidney injury, PLoS One 6 (2011), e18683.
[13] A. Lundgren-Nilsson, A. Dencker, A. Palstam, et al., Patient-reported outcome measures in osteoarthritis: a systematic search and review of their use and psychometric properties, RMD Open 4 (2018), e000715.
[14] S. Ohshima, S. Kuchen, C.A. Seemayer, et al., Galectin 3 and its binding protein in rheumatoid arthritis, Arthritis Rheum. 48 (2003) 2788–2795.
[15] W. Feng, X. Wu, S. Li, et al., Association of serum galectin-3 with acute exacerbation of chronic obstructive pulmonary disease, Med. Sci. Monit. 23 (2017) 4612–4618.
[16] W. Mosleh, M.R. Chaudhari, S. Sonkawade, et al., The therapeutic potential of blocking galectin-3 expression in acute myocardial infarction and mitigating inflammation of infarct region: a clinical outcome-based translational study, Biomark. Insights 13 (2018).