Journal of Anesthesiology & Clinical Research
https://hmpublisher.com/index.php/JACR/index
Vol 3 Issue 1 2022

Potential of Activated Platelet-Rich Plasma Against Osteoarthritis: In Vivo Study

Rachmat Hidayat[1#], Patricia Wulandari[2]

[1]Department of Biology, Faculty of Medicine, Universitas Sriwijaya, Palembang, Indonesia

[2]Cattleya Mental Health Center, Palembang, Indonesia

***Corresponding author:**
Rachmat Hidayat
Department of Biology,
Faculty of Medicine,
Universitas Sriwijaya,
Palembang, Indonesia
Email:
dr.rachmat.hidayat@gmail.com

ABSTRACT

Introduction: Osteoarthritis (OA) is a disorder that causes a decrease in the quality of life in elderly patients. The current treatment is only symptomatic in reducing inflammation. This study is one of the exploratory studies to examine the potential of platelet-rich plasma (PRP) in optimizing the improvement of OA patients through the inhibition of inflammatory signals in joint tissue in vivo. **Methods:** This study is an experimental study with a post-test-only approach with a control group design. A total of 30 rats (*Rattus norvegicus*) Wistar strain was included in this study and met the inclusion criteria in the form of the male gender, weight between 150-200 grams, and age 8-10 weeks. The rats were divided into 3 groups, namely the group that was not induced by OA and not given PRP (P1), the OA group and given 50 uL saline injection (P2), the OA group and given PRP 50 uL (P3), the treatment was carried out once a week for 4 weeks. **Results:** The results showed that the P3 group that was treated with platelet-rich plasma showed a significant decrease in interleukin-1ß levels when compared to the P2 group that was induced by OA but was only treated with saline ($p<0.05$). **Conclusion:** Platelet-rich plasma has the potential as a biological agent against osteoarthritis in an in vivo study.

Keywords: osteoarthritis, in vivo study, platelet-rich plasma, interleukin, experimental study.

Introduction

Osteoarthritis (OA) is a joint movement disorder, especially in the knee joint, caused by chronic inflammation in the joints.[1-3] The chronic inflammation that occurs in OA is caused by loss of joint cushion (joint cartilage), which causes trauma when the joint is moved. Trauma that occurs chronically triggers chronic inflammation that triggers complaints of pain and difficulty moving joints.[4] OA disorders are common in elderly patients, along with joint cartilage degeneration. According to epidemiological data, there has been a threefold increase in OA patients in the last 10 years. This is due to the increasing life expectancy, where more and more patients are more than 60 years old. This increase in life expectancy should also be accompanied by an increase in the quality of life of elderly patients.[5]

OA is one of the disorders that cause a decrease in the quality of life of sufferers, where the majority of patients are elderly. The current treatment is only symptomatic in reducing inflammation. The use of non-steroidal anti-inflammatory drugs that are very massive and used in the long term, on the one hand, triggers a series of unwanted side effects in the form of gastrointestinal disorders and triggers various other metabolic and endocrine disorders. This triggers the exploration of new therapeutic modalities for the development of definitive therapeutic modalities. Management of OA should not only refer to the chemical aspect in the form of drugs, but it is necessary to develop stem cell-based biological agents. One of the promising modalities to be developed is platelet-rich plasma.[6]

Platelet-rich plasma is one of the modalities of biological agents obtained from the patient's own blood. Platelet-rich plasma is the result of processing the patient's blood, where platelets are isolated from plasma through a structured separation process. Platelets are blood components that are responsible for repairing tissue damage. This makes platelets rich in growth factors that are important in repairing damaged tissue. Proper isolation and activation of platelets are important factors in optimizing the growth factors present in platelet-rich plasma.[7,8] This study is one of the exploratory studies to examine the potential of platelet-rich plasma in optimizing the improvement of OA patients through the inhibition of inflammatory signals in joint tissue in vivo.

Methods

This study is an experimental study with a post-test-only approach with a control group design. A total of 30 rats (*Rattus norvegicus*) Wistar strain was included in this study and met the inclusion criteria in the form of the male gender, weighing between 150-200 grams, and of

age 8-10 weeks first, rats were acclimatized for 7 days, then divided into 3 groups (P1, P2, and P3) randomly, where each group consisted of 10 rats. The P1 group was a group of mice that were not induced by OA and were not treated with platelet-rich plasma; The P2 group was a group of rats that were induced by OA by intra-articular injection of monosodium iodoacetate (MIA, 2 mg/50 uL) and administered an intra-articular saline injection of 50 uL; A P3 group is a group of rats that were induced by OA by intra-articular injection of monosodium iodoacetate (MIA, 2 mg/50 uL) and given an intra-articular injection of 50 uL of platelet-rich plasma, the treatment was administered once a week for 4 weeks. This study has been approved by the CMHC-Science and Research Center research ethics commission, number No.33/CMHC/KEPK/2021.

Platelet-rich plasma was obtained by first taking 3 mL of rat blood, then the process of isolation of platelet-rich plasma was carried out by mixing with 0.5% citrate buffer and centrifuged at 1200 rpm for 15 minutes. Next, the platelets were isolated and activated by adding 1% thrombin. The process of making platelet-rich plasma is carried out at the Eureka Research Laboratory, Palembang, Indonesia. Induction of OA was carried out by first anesthesia in rats using ketamine (dose of 0.015 mg/gBW) intramuscularly and chlorate (dose of 0.0025 mg/gBW) subcutaneously. Monosodium iodoacetate (MIA) was injected intra-articularly genu dextra region of experimental rats. Mice were monitored daily for signs of distress and signs of infection. Evacuation of genu dextra was carried out by anesthesia first in mice, then perfusion was carried out, and genu dextra was taken. The genu dextra rat tissue was put into a closed microtube container containing 0.9% NaCl liquid, one container for one sample. The samples were temporarily stored in a cooler bag (temperature $\leq$ 20ºC) and immediately stored in the freezer (temperature -20ºC). Analysis of IL-1B levels was carried out using the enzyme-linked immunosorbent assay (ELISA) method according to the manufacturer's instructions (CloudClone®).

After the data is collected, data cleaning, coding, and tabulation are carried out. All results were assessed by means ± standard deviation accompanied by a normality test (Shapiro-Wilk) and data homogeneity test (Levene Statistic). The test used in this study is one-way ANOVA. The results are said to be meaningful if $p \leq 0.05$. Data analysis was performed using SPSS version 25 for Windows.

Results and Discussion

Table 1 shows the assessment of inflammatory markers (IL-1βlevels). The higher levels of IL-1β indicate inflammation in the synovial tissue, as occurs in OA conditions. The P3 group that received the platelet-rich plasma treatment showed a significant decrease in IL-1β levels when compared to the P2 group that was induced by OA but was only treated with saline ($p<0.05$).

Table 1. Comparison of IL-1β levels between groups

Group	IL-1β levels (ρg/mL) Mean ± SD	value*
P1	23.56 ± 1.87	0.002
P2	245.87 ± 12.32	
P3	55.64 ± 2.43	

*one-way ANOVA, $p<0.05$

OA was caused by chronic inflammation due to trauma initiated by degeneration of joint cartilage.[9-11] Chronic trauma causes inflammation that results in the activation of various pro-inflammatory cytokines, namely interleukin-1ß (IL-1β), interleukin-6 (IL-6), and tumor necrosis factor (TNF)-α.[12] Chronic activation of the IL-1ß cytokine results in the inability to activate the anti-inflammatory cytokine TGF-β.[13] This causes no repair of cartilage tissue. Even the chronic inflammatory process causes osteoclast activation and further degrades bones and joints. The platelet-rich plasma that is rich in growth factors shows the potential in suppressing the activation of the inflammatory cytokine IL-1β.[14] The ability of platelet-rich plasma to suppress IL-1β suggests the potential of this biological agent in reducing chronic inflammation and preventing increasingly severe cartilage and bone damage.14 Of course, these results show the promising potential of platelet-rich plasma as a biological agent modality in overcoming OA.

Conclusion

Platelet-rich plasma has the potential as a biological agent against osteoarthritis in an in vivo study.

References

1. Boakye LA, Ross KA, Pinski JM. Platelet-rich plasma increases transforming growth factor-beta-1 expression at the graft-host interface following autologous osteochondral transplantation in a rabbit model. World J Orthop. 2015; 6(11):961.
2. Kim MJ, Lee JA, Shin MR, Park HJ, Roh SS. Improvement effect of *Corni Fructus* 30% ethanol extract by MIA-induced osteoarthritis animal model. Korean J Herbology. 2020; 35(1):35-44.
3. Sanchez M, Anitua E, Delgado D. A new strategy to tackle severe knee osteoarthritis: combination of intra-articular and intraosseous injections of platelet-rich plasma. Expert Opinion Biol Ther. 2016;16(5):627-43.
4. Moller KA, Klein S, Seeliger F, Finn A, Stenfors C, et al. Monosodium iodoacetate-induced monoarthritis develops differently in knee versus ankle joint in rats. Neurobiol Pain. 2019; 6:100036.
5. Asjid R, Faisal T, Qamar K, Malik S, Umbreen F, et al. Effect of platelet-rich plasma on mankin scoring in chemically-induced animal model of osteoarthritis. J Coll Physicians Surgeons Pakistan. 2019; 29(11):1067-71.
6. Jimbo S, Terashima Y, Teramoto A, et al. Antinociceptive effects of hyaluronic acid on monoiodoacetate-induced ankle osteoarthritis in rats. Journal of Pain Research. 2019: 12.
7. Preuss HG, Jarrell ST, Scheckenbach R, Lieberman S, Anderson RA. Comparative effects of chromium, vanadium and gymnema sylvestre on sugar-induced blood pressure elevations in SHR. Journal of the American College of Nutrition. 1998; 17(2): 116–23.
8. Beutler E, Duron O, Kelly BM. Improved method for the determination of blood glutathione. The Journal of Laboratory and Clinical Medicine. 1963; 61: 882–888.
9. Mannervik B, Guthenberg C. Glutathione transferase (human placenta), in Methods in Enzymology, Academic Press, London, UK. 1981.
10. Suvarna KS, Layton C, Bancroft JD. Bancroft's theory and practice of histological techniques e-book, Elsevier Health Sciences, London, UK. 2017.
11. Smit Y, Marais HJ, Thompson PN, Mahne, AT, Goddard A. Clinical findings, synovial fluid cytology and growth factor concentrations after intra-articular use of a platelet-

rich product in horses with osteoarthritis. Journal of the South African Veterinary Association. 2019; 90(1): 1–9.

12. Bruyère O, Cooper C, Pelletier JP, et al. An algorithm recommendation for the management of knee osteoarthritis in Europe and internationally: a report from a task force of the European Society for Clinical and Economic Aspects of Osteoporosis and Osteoarthritis (ESCEO). Seminars in Arthritis and Rheumatism. 2014: 44.
13. South S, Crabtree K, Vijayagopal P, Averitt D, Juma S, Dose-dependent effects of whole blueberries on cartilage health and pain in a monosodium iodoacetate (MIA) induced rat model of osteoarthritis, Current Developments in Nutrition. 2020; 4(2): 477.
14. Jasemian Y, Svendsen P, Deleuran B. Dagnaes-Hansen F. Refinement of the collagen induced arthritis model in rats by infrared thermography. British Journal of Medicine and Medical Research. 2011; 1(4):469–77.