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Multifactorial Comparative Analysis of Platelet-Rich Plasma and Serum Prepared Using a Commercially Available Centrifugation Kit

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Abstract

Background

Platelet-rich plasma (PRP) is an autologous product prepared by centrifuging whole blood. PRP is reported to have high tissue repair potential and anti-inflammatory properties. Recently, PRP has become a potential treatment option for osteoarthritis, contributing to pain relief and locomotive improvement. However, the underlying therapeutic mechanisms and key biochemical factors in PRP remain unclear. This study aimed to estimate the major factors for tissue repair involved in PRP treatment by comparing between serum and PRP prepared from the same patients using the Luminex assay.

Methodology

Blood samples were collected from nine healthy volunteers, and serum and PRP were prepared. PRP was prepared using a PEAK® PRP SYSTEM kit of DePuy Synthes Mitek Sports Medicine (Raynham, Massachusetts, USA), which is a commercially available PRP preparation kit. The white blood cell count, hemoglobin level, and platelet count were automatically measured for both whole blood and PRP in the hospital's clinical laboratory using the XE-5000™ Automated Hematology System (Sysmex, Kobe, Japan). Comparative analysis of biological factors was then performed using the Luminex assay on serum and PRP.

Results

PRP was found to have significantly higher white blood cell and platelet counts and lower hemoglobin levels than whole blood. Furthermore, PRP contained significantly higher levels of various factors, including interleukin (IL)-1ra, IL-10, IL-13, C-C motif chemokine ligand (CCL)-2, CCL3, CCL4, CCL8, CCL13, CCL21, C-X-C motif chemokine ligand (CXCL)-10, matrix metalloproteinase (MMP)-3, MMP-9, cluster of differentiation (CD) 40 ligand, vascular endothelial growth factor (VEGF), VEGF-C, platelet-derived growth factor (PDGF)-AB, PDGF-BB, and bone morphogenic protein (BMP)-2. Additionally, IL-1ra and IL-4 showed significant correlations with white blood cell counts in PRP, whereas VEGF had a significant correlation with platelet counts.

Conclusions

PRP contains various factors in higher quantities than serum. Specifically, the notable increase in the anti-inflammatory cytokine IL-1ra is suggested to play a key role as a major therapeutic mechanism of PRP.

Categories: Rheumatology, Orthopedics, Sports Medicine

Keywords: vascular endothelial growth factor (vegf), leukocyte-rich prp, il-1ra, luminex assay, osteoarthritis (oa), platelet-rich plasma (prp)

Introduction

Autologous cellular therapies that use platelet-rich plasma (PRP) have the potential to play an important role in various regenerative treatments [1]. Tissue repair strategies for treating musculoskeletal disorders and chronic recalcitrant wounds or skin ulcers are needed worldwide. Platelet-derived growth factors contained in PRP are expected to promote angiogenesis and mesenchymal stem cell proliferation and have been increasingly used in sports medicine, mainly to promote tissue repair for muscle injuries and rotator cuff tears of the shoulder [2,3]. Recently, some studies have shown the efficacy of PRP therapy in treating tendinopathies, such as patellar tendinitis and Achilles tendinitis improving pain and function, and shortening the time from injury to return to sports, raising interest in its anti-inflammatory effects [3]. Considering both the anti-inflammatory and tissue repair properties of PRP therapy, its use in treating osteoarthritis is gaining interest [4].

How to cite this article

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Osteoarthritis is a chronic degenerative disease in which patients suffer from pain and locomotive dysfunction due to joint deformity and inflammatory response [5]. Because the pathogenesis of the disease has been elucidated, it has become clear that chronic mild inflammation in the synovium and abnormal bone metabolism in the subchondral bone, including bone cysts and osteophyte formation, are involved in its pathogenesis [6]. Nonsteroidal anti-inflammatory drugs and corticosteroids are recommended as treatment strategies for this disease, along with patient education and exercise therapy as core treatment strategies [7]. Intra-articular hyaluronic acid injection is also used in outpatient practice; however, PRP has shown a better clinical outcome in a recent study comparing hyaluronic acid [8,9]. To elucidate the superior therapeutic mechanism of PRP, analyzing the biochemical factors contained in PRP may be needed, particularly considering the factors involved in the pathophysiology of osteoarthritis, which is not fully understood, although several studies have been conducted [10]. Therefore, this study aimed to determine the factors contained in PRP prepared using a commercially available centrifugation kit by comparing them with those contained in serum.

Materials And Methods

In this study, we analyzed PRP obtained from generally healthy adults. People with medical ailments, surgical history, and internal medication use, aged under 20 or over 60 years were excluded from this study. This study involved the examination of blood counts and a comparative multifactorial analysis of serum and PRP obtained from nine generally healthy adults (including six males and three females) with ages ranging from 28 to 51 years. Before blood sample collection, the purpose and protocol of the study were explained to each participant, and informed consent for the study was obtained after confirmation of their lack of medical ailments, surgical history, and internal medication use. This study was conducted according to the ethical guidelines for research involving human subjects at Showa University. The investigation was implemented after obtaining approval from Showa University (22-149-B). After obtaining informed consent and providing explanations to the research participants, blood samples were collected. The blood sample collection procedure was as follows: 2 mL of blood was collected for whole blood cell count, 8 mL for serum preparation, and 27 mL for PRP preparation. To prepare PRP, 3 mL of acid citrate dextrose solution A (ACD-A) was mixed into the collection syringe to prevent coagulation.

The blood samples were subjected to centrifugation following the protocol of the PEAK® PRP SYSTEM kit of DePuy Synthes Mitek Sports Medicine (Raynham, Massachusetts, USA), resulting in the retrieval of 3 mL of PRP. Of the retrieved 3 mL of PRP, 1 mL was allocated for multiplex assays, 1 mL for blood cell counts, and 1 mL for storage. The 8 mL of blood collected for serum preparation was stored at room temperature for 30 minutes to allow clot formation. Subsequently, the blood sample was centrifuged at $1,000 \times g$ for 15 minutes; then, the supernatant was collected. The PRP and serum samples for comparative multifactorial assay and storage were frozen at -80°C until the time of analysis.

Whole blood samples for blood cell counts were placed in ethylenediaminetetraacetic acid-containing collection tubes and immediately analyzed in the facility's testing laboratory using the XE-5000™ Automated Hematology System (Sysmex, Kobe, Japan). Blood cell counts, including white blood cell (WBC) count, hemoglobin (Hb) level, and platelet (Plt) count, were measured for both whole blood and PRP, and comparative analyses were performed.

The prepared serum and PRP were outsourced to Genetic Lab Co., Ltd. (Hokkaido, Japan) for the Luminex assay. The analysis was performed using the BioPlex-200 system from Bio-Rad Laboratories (Richmond, CA), and measurements were performed for the following parameters: interleukin (IL)-1 beta, IL-1ra, IL-4, IL-6, IL-10, IL-13, and IL-17; C-C motif chemokine ligand (CCL) 2, CCL3, CCL4, CCL8, CCL13, CCL15, and CCL21; C-X3-C motif chemokine ligand (CX3CL)-1; C-X-C motif chemokine ligand (CXCL)-8 and CXCL10; matrix metalloproteinase (MMP)-1, MMP-3, MMP-9, and MMP-13; cluster of differentiation (CD)-40 ligand and CD44; vascular endothelial growth factor (VEGF) and VEGF-C; platelet-derived growth factor (PDGF)-AB and PDGF-BB; and bone morphogenic protein (BMP)-2. Conducting a Luminex assay typically involves several key steps, and a general overview of the process is presented below.

First, the Bio-Plex 200 system was set up and calibrated according to the manufacturer's instructions. After preparing all necessary reagents, including assay beads coated with specific antibodies, standards, and samples, the lyophilized bead mix was reconstituted with the supplied buffer or diluent, and the bead suspension was thoroughly vortexed for at least 30 seconds. Samples or standards were then added to the microplate wells. The samples were estimated to fall within the linear range of the assay, which were prepared to be diluted three times. The microplate containing the samples, standards, and bead suspension was then incubated at room temperature for two hours to allow the antigen-antibody reaction to occur. After incubation, the microplate wells were washed to remove any unbound material or debris. The detection antibody specific to the analyte of interest in each well was then added and incubated at room temperature for one hour. The microplate was washed again to remove unbound detection antibodies, and an appropriate streptavidin-phycoerythrin detection reagent was applied to each well for signal amplification and detection. For data acquisition, the prepared microplate was loaded into the Bio-Plex 200 system, and the assay was run to read the fluorescence signals from the beads in each well. The system collected data on the fluorescence intensity of each bead. Data were then analyzed using the Bio-Plex Manager software.

For statistical analysis, JMP Pro ver. 16 was used. The blood counts for whole blood and PRP and the levels of each factor in serum and PRP measured using the Luminex assay were expressed as means and standard deviations. Because this was a comparative analysis using biological samples collected from the same patients, a paired t-test was performed for each, and the significance level was set at 5%. Correlations with blood counts were analyzed for each analyte measured with the Luminex assay.

Results

The WBC count was $5,116 \pm 1,139/\mu\text{L}$ in whole blood and $23,006 \pm 6,819/\mu\text{L}$ in PRP. The Hb level was $14.3 \pm 1.0 \text{ g/dL}$ in whole blood and $3.7 \pm 0.8 \text{ g/dL}$ in PRP. The Plt count was $23.8 \pm 4.4 \times 10^4/\mu\text{L}$ in whole blood and $143.4 \pm 39.1 \times 10^4/\mu\text{L}$ in PRP. PRP had significantly higher WBC count ($p < 0.001$), lower Hb level ($p < 0.001$), and higher Plt count ($p < 0.001$) (Figure 1).

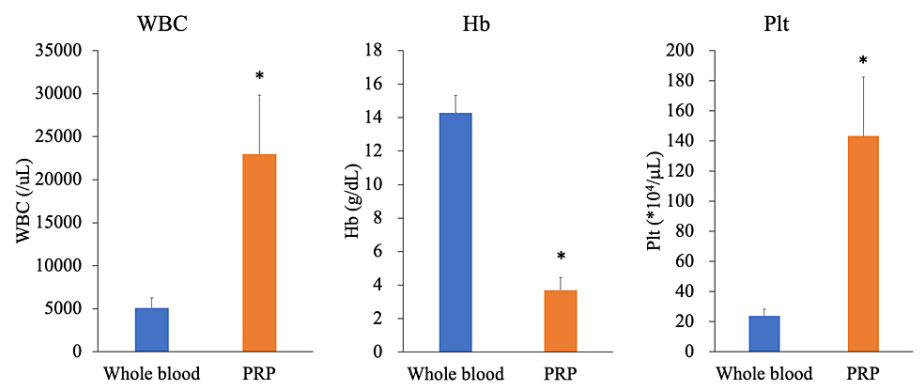


FIGURE 1: Blood cell count for whole blood and PRP.

*: $P < 0.05$ vs. whole blood assessed by the paired t-test.

WBC = white blood cell; Hb = hemoglobin; Plt = platelet; PRP = platelet-rich plasma

Comparison of serum and PRP showed that various factors were significantly more abundant in the PRP group than those in the serum group (Figure 2). Specifically, the levels of IL-1ra ($p = 0.004$), IL-10 ($p = 0.024$), IL-13 ($p = 0.026$), CCL2 ($p = 0.001$), CCL3 ($p = 0.005$), CCL4 ($p < 0.001$), CCL8 ($p = 0.012$), CCL13 ($p < 0.001$), CCL21 ($p = 0.045$), CXCL10 ($p = 0.006$), MMP-3 ($p = 0.041$), MMP-9 ($p = 0.010$), CD40 ligand ($p = 0.006$), VEGF ($p = 0.006$), VEGF-C ($p = 0.008$), PDGF-AB ($p = 0.001$), PDGF-BB ($p < 0.001$), and BMP-2 ($p < 0.001$) were significantly higher in PRP than in serum.

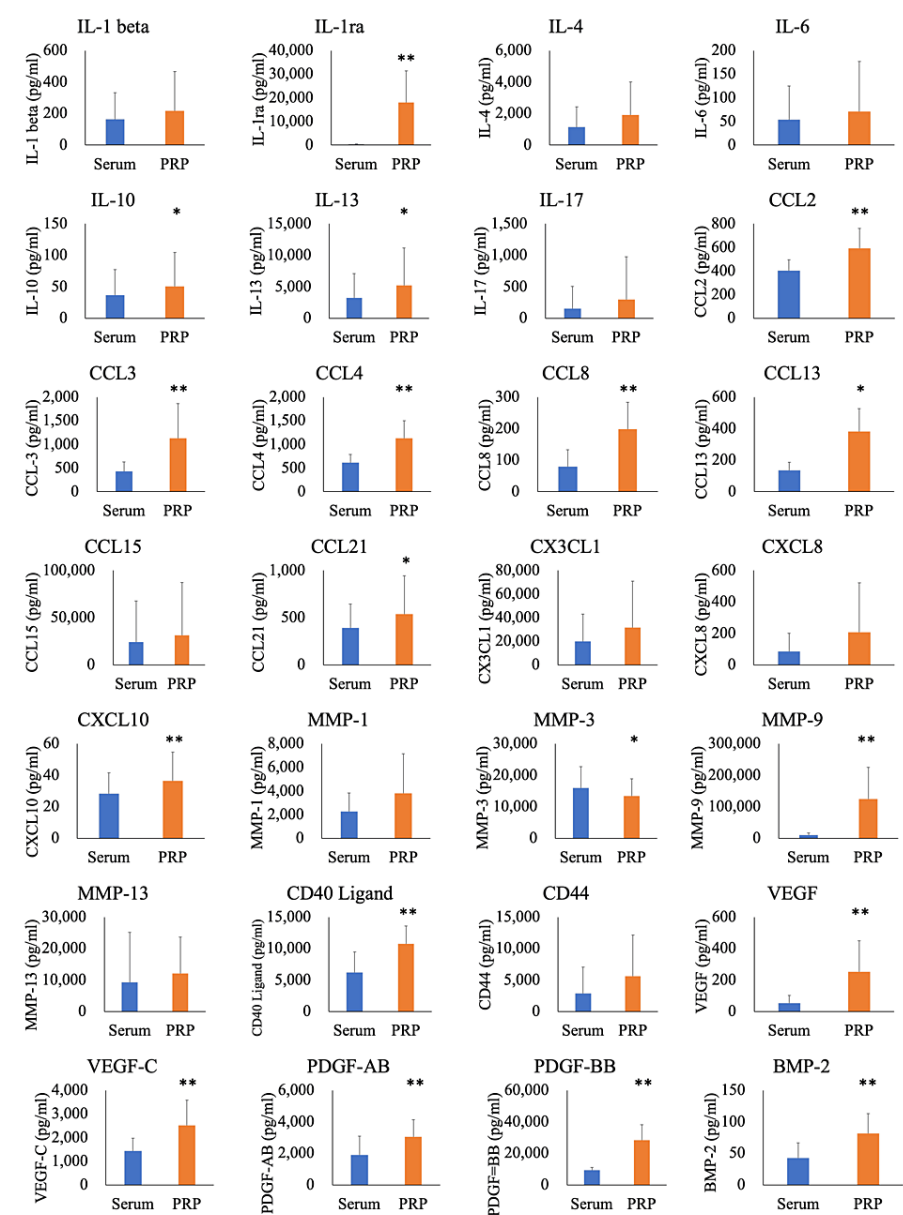


FIGURE 2: Comparison between serum and PRP using the Luminex assay.

*, P < 0.05, **, P < 0.01 vs. serum assessed by the paired t-test.

IL = interleukin; CCL = C-C motif chemokine ligand; CXCL = C-X-C motif chemokine ligand; CX3CL = C-X3-C motif chemokine ligand; MMP = matrix metalloproteinase; CD = cluster of differentiation; VEGF = vascular endothelial growth factor; PDGF = platelet-derived growth factor; BMP = bone morphogenic protein; PRP = platelet-rich plasma

Subsequently, a correlation analysis was performed between each analyte and the WBC count, Hb level, and Plt count in PRP (Table 1). Significant correlations were found between WBC counts and IL-1ra (correlation coefficient (CC) = 0.842, 95% confidence interval (CI) = 0.404-0.966, p = 0.004), IL-4 (CC = 0.869, 95% CI = 0.483-0.972, p = 0.002), and CCL4 (CC = 0.771, 95% CI = 0.218-0.949, p = 0.015). No factors were found to be correlated with Hb levels. VEGF (CC = 0.684, 95% CI = 0.036-0.927, p = 0.042) was significantly correlated with Plt count.

	WBC					Hb					Plt				
	Correlation coefficient	95% confidence interval			P-value	Correlation coefficient	95% confidence interval			P-value	Correlation coefficient	95% confidence interval			P-value
IL-1 beta	0.006	-0.660	~	0.667	0.988	0.095	-0.607	~	0.714	0.808	-0.022	-0.676	~	0.652	0.955
IL-1ra	0.842	0.404	~	0.966	0.004**	-0.059	-0.696	~	0.630	0.880	0.629	-0.059	~	0.912	0.069
IL-4	0.869	0.483	~	0.972	0.002**	-0.543	-0.887	~	0.188	0.130	0.160	-0.564	~	0.745	0.682
IL-6	-0.159	-0.744	~	0.565	0.682	-0.354	-0.824	~	0.405	0.350	-0.528	-0.882	~	0.208	0.143
IL-10	0.417	-0.341	~	0.847	0.264	-0.649	-0.917	~	0.025	0.058	-0.264	-0.789	~	0.485	0.492
IL-13	-0.011	-0.670	~	0.658	0.976	0.224	-0.517	~	0.773	0.563	0.217	-0.522	~	0.770	0.574
IL-17	0.027	-0.648	~	0.679	0.945	-0.491	-0.871	~	0.256	0.179	-0.125	-0.729	~	0.587	0.747
CCL2	-0.378	-0.833	~	0.381	0.315	-0.020	-0.675	~	0.653	0.959	-0.478	-0.866	~	0.273	0.193
CCL3	-0.404	-0.842	~	0.355	0.280	-0.012	-0.671	~	0.657	0.974	-0.192	-0.759	~	0.541	0.620
CCL4	0.771	0.218	~	0.949	0.015*	-0.304	-0.805	~	0.451	0.426	0.389	-0.370	~	0.837	0.300
CCL8	0.024	-0.692	~	0.717	0.954	0.625	-0.141	~	0.923	0.097	0.457	-0.364	~	0.879	0.255
CCL13	0.613	-0.086	~	0.908	0.079	-0.178	-0.75	~	0.551	0.646	0.193	-0.540	~	0.760	0.620
CCL15	-0.205	-0.765	~	0.531	0.595	0.286	-0.466	~	0.799	0.455	-0.013	-0.671	~	0.656	0.972
CCL21	0.480	-0.270	~	0.868	0.191	-0.500	-0.873	~	0.246	0.170	-0.043	-0.687	~	0.639	0.912
CX3CL1	-0.291	-0.800	~	0.462	0.447	0.352	-0.406	~	0.824	0.352	0.051	-0.634	~	0.692	0.897
CX3CL8	0.618	-0.079	~	0.909	0.076	-0.617	-0.908	~	0.080	0.077	-0.126	-0.729	~	0.587	0.745
CXCL10	-0.433	-0.852	~	0.324	0.244	0.205	-0.531	~	0.765	0.597	-0.421	-0.848	~	0.337	0.258
MMP-1	-0.280	-0.796	~	0.471	0.465	0.184	-0.546	~	0.756	0.635	0.321	-0.436	~	0.812	0.400
MMP-3	0.352	-0.407	~	0.824	0.352	-0.390	-0.837	~	0.370	0.299	-0.027	-0.679	~	0.648	0.943
MMP-9	0.617	-0.079	~	0.909	0.077	0.061	-0.628	~	0.697	0.876	0.525	-0.213	~	0.882	0.147
MMP-13	0.075	-0.620	~	0.704	0.847	-0.457	-0.860	~	0.297	0.216	-0.047	-0.690	~	0.637	0.903
CD40 ligand	0.270	-0.480	~	0.792	0.483	-0.051	-0.691	~	0.634	0.895	0.525	-0.213	~	0.882	0.147
CD44	0.377	-0.383	~	0.833	0.317	0.360	-0.399	~	0.827	0.341	0.522	-0.218	~	0.881	0.150
VEGF	0.197	-0.537	~	0.761	0.612	0.435	-0.322	~	0.853	0.242	0.684	0.036	~	0.927	0.042*
VEGF-C	-0.186	-0.756	~	0.545	0.632	0.284	-0.468	~	0.798	0.459	0.192	-0.541	~	0.759	0.621
PDGF-AB	0.218	-0.521	~	0.771	0.573	0.072	-0.621	~	0.703	0.854	0.100	-0.604	~	0.716	0.799
PDGF-BB	0.161	-0.563	~	0.746	0.679	0.297	-0.457	~	0.803	0.438	0.607	-0.095	~	0.906	0.083
BMP-2	0.194	-0.539	~	0.760	0.616	-0.404	-0.842	~	0.355	0.280	-0.436	-0.853	~	0.320	0.240

TABLE 1: Correlation analysis between biochemical factor levels of PRP and blood cell counts.

IL = interleukin; CCL = C-C motif chemokine ligand; CXCL = C-X-C motif chemokine ligand; CX3CL = C-X3-C motif chemokine ligand; MMP = matrix metalloproteinase; CD = cluster of differentiation; VEGF = vascular endothelial growth factor; PDGF = platelet-derived growth factor; BMP = bone morphogenic protein; PRP = platelet-rich plasma

Discussion

PRP is an autologous product prepared by centrifuging whole blood, and it is reported to have high tissue repair potential and anti-inflammatory properties [1]. Recently, PRP treatment has been considered a therapeutic strategy for osteoarthritis, contributing to pain relief and locomotive improvement [3,4].

PRP preparations are categorized mainly into two types, namely, leukocyte-rich PRP (LR-PRP) preparations,

having more WBCs than baseline, and leukocyte-poor PRP (LP-PRP) preparations, having less than baseline [11]. Recently, whether LR-PRP provides a superior therapeutic benefit over LP-PRP has been debated. However, several randomized controlled trials have found no significant differences in clinical outcomes between the two PRP types [12,13]. Furthermore, a study comparing placebo (saline), LR-PRP, and LP-PRP in treating patellar tendinopathy also failed to show significant differences in clinical outcomes [14], casting skepticism on the efficacy of PRP therapy itself. In contrast, it has been reported that LR-PRP resulted in significant histological improvements in a rabbit Achilles tendonitis model compared with LP-PRP [15].

In this study, LR-PRP was prepared using a commercially available centrifugation kit. To assess the blood count in PRP, whole blood collected from the same subjects was used as a control. PRP exhibited significantly higher WBC and Plt count than whole blood. It has been reported that for PRP treatment to be effective in treating osteoarthritis, the Plt count in PRP should be at least 10 billion/8 mL (1.25 billion/mL) [16]. In this study, the Plt count was approximately 1.43 billion/mL, suggesting the potential for favorable clinical outcomes. In contrast, Hb levels were significantly lower. Additionally, because Hb has been reported to be responsible for exacerbating synovitis, which is crucial to the development of osteoarthritis [17], PRP with lower Hb levels is desirable, and the agents prepared in this study revealed a lower level.

It has also become evident that the concentrations of IL-1ra and VEGF were notably elevated in PRP compared with those in serum. Furthermore, these two factors showed significant positive correlations with WBC and Plt count, respectively. IL-1ra is an anti-inflammatory cytokine that antagonizes IL-1 alpha and IL-1 beta signaling and exists in two subtypes: a 17-kDa form secreted by monocytes, neutrophils, and macrophages, and an 18-kDa form that remains in the cytoplasm of epithelial cells, endothelial cells, and fibroblasts [18,19]. In this study, IL-1ra showed a significant positive correlation with WBC count, suggesting the superiority of LR-PRP. In contrast, according to previous reports, the average concentration of IL-1ra in LP-PRP was lower than that in this study [20], indicating that using LR-PRP may be more advantageous due to its enhanced anti-inflammatory effects mediated by IL-1ra. At present, IL-1ra is not currently in clinical use, and only a pilot study has been reported to assess its safety and efficacy in patients with anterior cruciate ligament injuries [21]. VEGF plays an important role in tissue repair by promoting endothelial cell proliferation and tissue vascularization and stimulating the accumulation of stem and immune cells necessary for tissue repair in inflamed and injured tissues [22]. In this study, VEGF showed a significant positive correlation with the Plt count, consistent with previous findings [23,24]. VEGF has been reported to promote tissue repair in meniscus injuries [25] but has also been associated with degenerative changes in articular cartilage [26,27], raising concerns about its contribution to osteoarthritis progression. Another important aspect to consider is the effect of PRP on synovial cells. It has been reported that PRP promotes the secretion of MMPs, which are enzymes involved in the degradation of articular cartilage [28].

PRP comprises a lot of components that may have various effects on anabolic and catabolic changes in tissues. It has generally shown positive clinical results, including pain relief and improved locomotive function in patients with osteoarthritis [4,8,9]. However, long-term studies are required to clarify its potential to inhibit disease progression. Based on the results of this study, which highlighted IL-1ra as an important anti-inflammatory factor, further investigation of the therapeutic effects of LR-PRP is needed.

This study has several limitations. First, the participants were healthy adults; therefore, further analysis of PRP in patients with osteoarthritis is required. However, the validity of this study is to investigate the influence of preparing PRP from blood samples affecting the level of biochemical mediators contained in PRP, so it is necessary to remove as many possible biases as we can, such as past medical history, current ailment, surgical history, and medications. Therefore, we believe it is applicable that only healthy adults were included in this study. Second, the number of subjects was relatively small. Although no outliers were observed in any parameter, a larger sample size would have strengthened the validity of the findings. However, a previous study has shown that PRP has much IL-1ra [20], which is relevant to our study, so it may be appropriate to compare PRP and serum originating from the same subject using a paired t-test. Finally, this study only focused on LR-PRP prepared using only one preparation kit, and a similar analysis should be performed for LP-PRP.

Conclusions

In this study, a multifactorial analysis using the Luminex assay was performed on serum and PRP prepared from blood samples of the same subject and PRP contained a larger amount of various kinds of biochemical mediators than serum. Furthermore, IL-1ra and VEGF were significantly more concentrated and correlated with leukocyte and platelet counts, respectively. In light of previous reports on these two biological factors, LR-PRP prepared using the PEAK PRP SYSTEM may have favorable clinical results. In particular, the significant increase in IL-1ra, about 40 times higher than serum, suggests that it plays an important role in PRP treatment, and the anti-inflammatory effect may shed light on the therapeutic mechanism of PRP. However, the results of this study are not directly applicable to clinical practice because the study was performed on a relatively small number of subjects, especially healthy adults without any ailments in medical history. Thus, to determine if PRP is a potential therapeutic strategy for osteoarthritis or other degenerative diseases, it is necessary to examine whether PRP made from patients with the disease has similar biochemical factor profiles to those of healthy adults. Therefore, further studies are needed to elucidate the efficacy of PRP treatment for osteoarthritis patients, such as multifactorial investigation for

PRP in osteoarthritis patients.

Additional Information

Author Contributions

All authors have reviewed the final version to be published and agreed to be accountable for all aspects of the work.

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Drafting of the manuscript: Takayuki Okumo

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Supervision: Masataka Sunagawa, Koji Kanzaki

Disclosures

Human subjects: Consent was obtained or waived by all participants in this study. Institutional Review Board of Showa University issued approval 22-149-B. This study was conducted in accordance with the ethical guidelines for research involving human subjects at Showa University, and the investigation was implemented after obtaining the following approval number: 22-149-B. **Animal subjects:** All authors have confirmed that this study did not involve animal subjects or tissue. **Conflicts of interest:** In compliance with the ICMJE uniform disclosure form, all authors declare the following: **Payment/services info:** PEAK® PRP SYSTEM, a PRP preparation kit, was provided by DePuy Synthes Mitek Sports Medicine. **Financial relationships:** All authors have declared that they have no financial relationships at present or within the previous three years with any organizations that might have an interest in the submitted work. **Other relationships:** All authors have declared that there are no other relationships or activities that could appear to have influenced the submitted work.

References

1. Everts P, Onishi K, Jayaram P, Lana JF, Mautner K: Platelet-rich plasma: New performance understandings and therapeutic considerations in 2020. *Int J Mol Sci.* 2020, 21:7794. [10.3390/ijms21207794](https://doi.org/10.3390/ijms21207794)
2. Mlynarek RA, Kuhn AW, Bedi A: Platelet-rich plasma (PRP) in Orthopedic Sports Medicine . *Am J Orthop (Belle Mead NJ).* 2016, 45:290-326.
3. Collins T, Alexander D, Barkatali B: Platelet-rich plasma: a narrative review . *EFORT Open Rev.* 2021, 6:225-35. [10.1302/2058-5241.6.200017](https://doi.org/10.1302/2058-5241.6.200017)
4. Szwedowski D, Szczepanek J, Paczesny Ł, Zabrzynski J, Gagat M, Mobasher A, Jeka S: The effect of platelet-rich plasma on the intra-articular microenvironment in knee osteoarthritis. *Int J Mol Sci.* 2021, 22:5492. [10.3390/ijms22115492](https://doi.org/10.3390/ijms22115492)
5. Michael JW, Schlüter-Brust KU, Eysel P: The epidemiology, etiology, diagnosis, and treatment of osteoarthritis of the knee. *Dtsch Arztebl Int.* 2010, 107:152-62. [10.3238/arztebl.2010.0152](https://doi.org/10.3238/arztebl.2010.0152)
6. Loeser RF, Goldring SR, Scanzello CR, Goldring MB: Osteoarthritis: a disease of the joint as an organ . *Arthritis Rheum.* 2012, 64:1697-707. [10.1002/art.34453](https://doi.org/10.1002/art.34453)
7. Bannuru RR, Osani MC, Vaysbrot EE, et al.: OARSJ guidelines for the non-surgical management of knee, hip, and polyarticular osteoarthritis. *Osteoarthritis Cartilage.* 2019, 27:1578-89. [10.1016/j.joca.2019.06.011](https://doi.org/10.1016/j.joca.2019.06.011)
8. Tang JZ, Nie MJ, Zhao JZ, Zhang GC, Zhang Q, Wang B: Platelet-rich plasma versus hyaluronic acid in the treatment of knee osteoarthritis: a meta-analysis. *J Orthop Surg Res.* 2020, 15:403. [10.1186/s13018-020-01919-9](https://doi.org/10.1186/s13018-020-01919-9)
9. Chen L, Jin S, Yao Y, He S, He J: Comparison of clinical efficiency between intra-articular injection of platelet-rich plasma and hyaluronic acid for osteoarthritis: a meta-analysis of randomized controlled trials. *Ther Adv Musculoskelet Dis.* 2023, 15:1759720X231157043. [10.1177/1759720X231157043](https://doi.org/10.1177/1759720X231157043)
10. Mussano F, Genova T, Munaron L, Petrillo S, Erovigni F, Carossa S: Cytokine, chemokine, and growth factor profile of platelet-rich plasma. *Platelets.* 2016, 27:467-71. [10.3109/09537104.2016.1143922](https://doi.org/10.3109/09537104.2016.1143922)
11. Le AD, Enweze L, DeBaun MR, Dragoo JL: Current clinical recommendations for use of platelet-rich plasma . *Curr Rev Musculoskelet Med.* 2018, 11:624-34. [10.1007/s12178-018-9527-7](https://doi.org/10.1007/s12178-018-9527-7)
12. Di Martino A, Boffa A, Andriolo L, et al.: Leukocyte-rich versus leukocyte-poor platelet-rich plasma for the treatment of knee osteoarthritis: a double-blind randomized trial. *Am J Sports Med.* 2022, 50:609-17. [10.1177/03635465211064303](https://doi.org/10.1177/03635465211064303)
13. Muthu S, Patel S, Selvaraj P, Jeyaraman M: Comparative analysis of leukocyte poor vs leukocyte rich platelet-rich plasma in the management of lateral epicondylitis: systematic review & meta-analysis of randomised controlled trials. *J Clin Orthop Trauma.* 2021, 19:96-107. [10.1016/j.jcot.2021.05.020](https://doi.org/10.1016/j.jcot.2021.05.020)
14. Scott A, LaPrade RF, Harmon KG, et al.: Platelet-rich plasma for patellar tendinopathy: a randomized controlled trial of leukocyte-rich PRP or leukocyte-poor PRP versus saline. *Am J Sports Med.* 2019, 47:1654-61. [10.1177/0363546519837954](https://doi.org/10.1177/0363546519837954)

15. Jiang G, Wu Y, Meng J, et al.: Comparison of leukocyte-rich platelet-rich plasma and leukocyte-poor platelet-rich plasma on Achilles tendinopathy at an early stage in a rabbit model. *Am J Sports Med.* 2020, 48:1189-99. [10.1177/0363546520906142](https://doi.org/10.1177/0363546520906142)
16. Bansal H, Leon J, Pont JL, Wilson DA, Bansal A, Agarwal D, Preoteasa I: Author correction: platelet-rich plasma (PRP) in osteoarthritis (OA) knee: correct dose critical for long term clinical efficacy. *Sci Rep.* 2021, 11:18612. [10.1038/s41598-021-98365-2](https://doi.org/10.1038/s41598-021-98365-2)
17. Tajima T, Sekimoto T, Yamaguchi N, Taniguchi N, Kurogi S, Maruyama M, Chosa E: Hemoglobin stimulates the expression of ADAMTS-5 and ADAMTS-9 by synovial cells: a possible cause of articular cartilage damage after intra-articular hemorrhage. *BMC Musculoskelet Disord.* 2017, 18:449. [10.1186/s12891-017-1815-7](https://doi.org/10.1186/s12891-017-1815-7)
18. Gabay C, Lamacchia C, Palmer G: IL-1 pathways in inflammation and human diseases. *Nat Rev Rheumatol.* 2010, 6:232-41. [10.1038/nrrheum.2010.4](https://doi.org/10.1038/nrrheum.2010.4)
19. Arend WP, Malyak M, Guthridge CJ, Gabay C: Interleukin-1 receptor antagonist: role in biology. *Annu Rev Immunol.* 1998, 16:27-55. [10.1146/annurev.immunol.16.1.27](https://doi.org/10.1146/annurev.immunol.16.1.27)
20. Jayaram P, Mitchell PJ, Shybut TB, Moseley BJ, Lee B: Leukocyte-rich platelet-rich plasma is predominantly anti-inflammatory compared with leukocyte-poor platelet-rich plasma in patients with mild-moderate knee osteoarthritis: a prospective, descriptive laboratory study. *Am J Sports Med.* 2023, 51:2133-40. [10.1177/03635465231170394](https://doi.org/10.1177/03635465231170394)
21. Kraus VB, Birmingham J, Stabler TV, et al.: Effects of intraarticular IL1-Ra for acute anterior cruciate ligament knee injury: a randomized controlled pilot trial (NCT00332254). *Osteoarthritis Cartilage.* 2012, 20:271-8. [10.1016/j.joca.2011.12.009](https://doi.org/10.1016/j.joca.2011.12.009)
22. Hoeben A, Landuyt B, Highley MS, Wildiers H, Van Oosterom AT, De Bruijn EA: Vascular endothelial growth factor and angiogenesis. *Pharmacol Rev.* 2004, 56:549-80. [10.1124/pr.56.4.3](https://doi.org/10.1124/pr.56.4.3)
23. Wasterlain AS, Hillary JB, Jason LD: Contents and formulations of platelet-rich plasma. *Oper Tech Orthop.* 2012, 1:33-42. [10.1053/j.oto.2011.11.001](https://doi.org/10.1053/j.oto.2011.11.001)
24. Ha CW, Park YB, Jang JW, Kim M, Kim JA, Park YG: Variability of the composition of growth factors and cytokines in platelet-rich plasma from the knee with osteoarthritis. *Arthroscopy.* 2019, 35:2878-84.e1. [10.1016/j.arthro.2019.04.010](https://doi.org/10.1016/j.arthro.2019.04.010)
25. Lu Z, Furumatsu T, Fujii M, Maehara A, Ozaki T: The distribution of vascular endothelial growth factor in human meniscus and a meniscal injury model. *J Orthop Sci.* 2017, 22:715-21. [10.1016/j.jos.2017.02.006](https://doi.org/10.1016/j.jos.2017.02.006)
26. Murata M, Yudoh K, Masuko K: The potential role of vascular endothelial growth factor (VEGF) in cartilage: how the angiogenic factor could be involved in the pathogenesis of osteoarthritis?. *Osteoarthritis Cartilage.* 2008, 16:279-86. [10.1016/j.joca.2007.09.003](https://doi.org/10.1016/j.joca.2007.09.003)
27. Hamilton JL, Nagao M, Levine BR, Chen D, Olsen BR, Im HJ: Targeting VEGF and its receptors for the treatment of osteoarthritis and associated pain. *J Bone Miner Res.* 2016, 31:911-24. [10.1002/jbmr.2828](https://doi.org/10.1002/jbmr.2828)
28. Browning SR, Weiser AM, Woolf N, et al.: Platelet-rich plasma increases matrix metalloproteinases in cultures of human synovial fibroblasts. *J Bone Joint Surg Am.* 2012, 94:e1721-7. [10.2106/JBJS.K.01501](https://doi.org/10.2106/JBJS.K.01501)