Leukocyte-Rich Spin Regimens in ACP Max™ System

Arthrex Research

OBJECTIVE

This study aimed to determine spin regimens for 30, 60, and 90 mL in the ACP Max system that produce a leukocyte-rich platelet-rich plasma (PRP) and to characterize the cellular content of these regimens.[1,2]

MATERIALS AND METHODS

Blood Collection

Blood was collected from donors (N = 6) using 13.3% acid citrate dextrose solution A (ACD-A) as the anticoagulant. A small volume of anticoagulated blood from each donor was aliquoted for baseline complete blood count (CBC) analyses.

PRP Preparation

PRP was prepared for each donor as described below in Table 1.

Table 1. Spin regimens to prepare the various groups.

Step	30 mL Leukocyte-Rich PRP	60 mL Leukocyte-Rich PRP	90 mL Leukocyte-Rich PRP
1	Centrifuge at 3200 rpm for 3 minutes	Centrifuge at 3200 rpm for 6 minutes	Centrifuge at 3200 rpm for 9 minutes
2	Remove platelet-poor plasma to 2 tick marks above buffy coat		
3	Collect next 15 mL of fluid into double syringe		
4	Gently invert double syringe 15-20 times		
5	Centrifuge at 1500 rpm for 5 minutes		
6	Collect PRP until the red blood cell (RBC) layer is reached and continue collecting the next 1 mL of fluid	Collect PRP without any RBCs, along with an additional 1-1.2 mL of RBCs	Collect PRP until the RBC layer is reached and continue collecting the next 1.2 mL of fluid

The PRP volumes were recorded. A small aliquot of each PRP was collected for each device, and a CBC with differential was performed.

Data Analysis

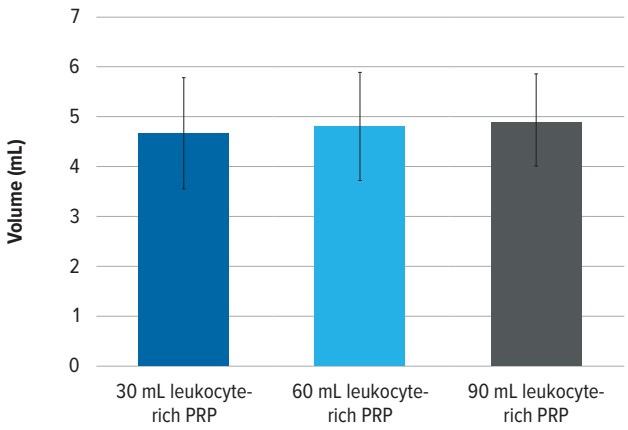
The following analyses were performed on all CBC results, focusing on the platelet (PLT), neutrophil (NE), white blood cell (WBC), and monocyte (MONO) groups.

- The average volume of each group was determined without any additional processing.
- The fold change in the concentration of each cell type relative to baseline was calculated by dividing the values obtained from the PRP by the corresponding values from the respective whole blood.
- The dose was calculated by multiplying the cell concentrations by the recovered fluid volume.
- Following the calculations for each device, the data were averaged across the 6 donors for each group.

RESULTS

The volume of each group was analyzed without any additional processing (Figure 1). The average volume of each leukocyte-rich group was between 4.7 and 4.9 mL.

Figure 1. Average volume of leukocyte-rich PRP, with standard deviation, for 30, 60, and 90 mL spin regimens (N = 6, except 90 mL PRP, which used N = 5).



The fold change in cell concentration relative to baseline was calculated for platelets, white blood cells, monocytes, and neutrophils (Figure 2). The average fold change and standard deviation are listed in Table 2.

Figure 2. The average fold change over baseline, with standard deviation, of PLT, WBC, MONO, and NE (N = 6, except 90 mL PRP, which used N = 5).

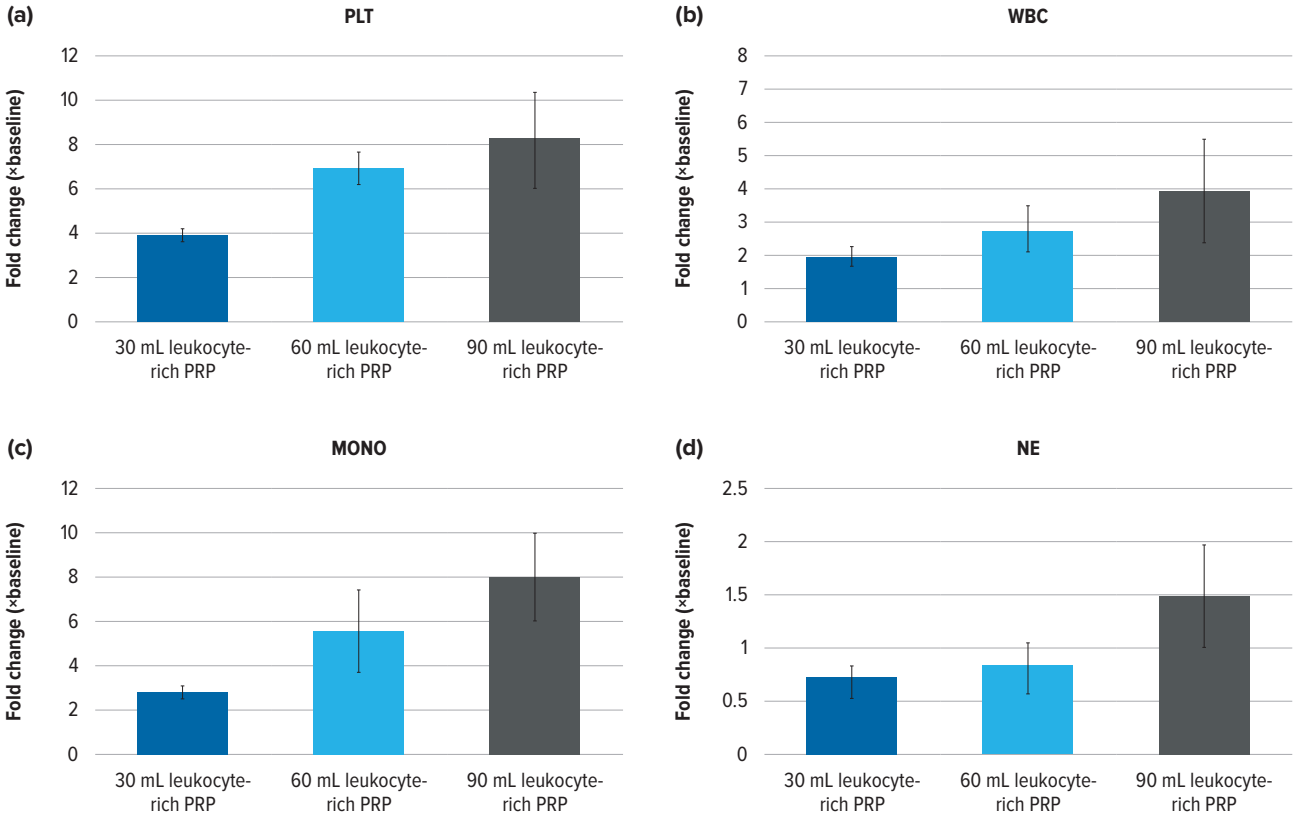


Table 2. Average fold change, with standard deviation, of the analyzed cells.

	30 mL	60 mL	90 mL
PLT	3.89 ± 0.39	6.88 ± 0.74	8.21 ± 2.09
WBC	1.95 ± 0.31	2.77 ± 0.66	3.93 ± 1.57
MONO	2.92 ± 0.23	5.57 ± 1.81	8.02 ± 1.92
NE	0.68 ± 0.15	0.83 ± 0.24	1.48 ± 0.48

The delivered dose of each cell type was calculated based on the CBC results and the recovered volumes (Figure 3). The average delivered dose and standard deviation for the respective volumes are listed in Table 3.

Figure 3. The average delivered dose, with standard deviation, of PLT, WBC, MONO, and NE (N = 6, except 90 mL PRP, which used N = 5).

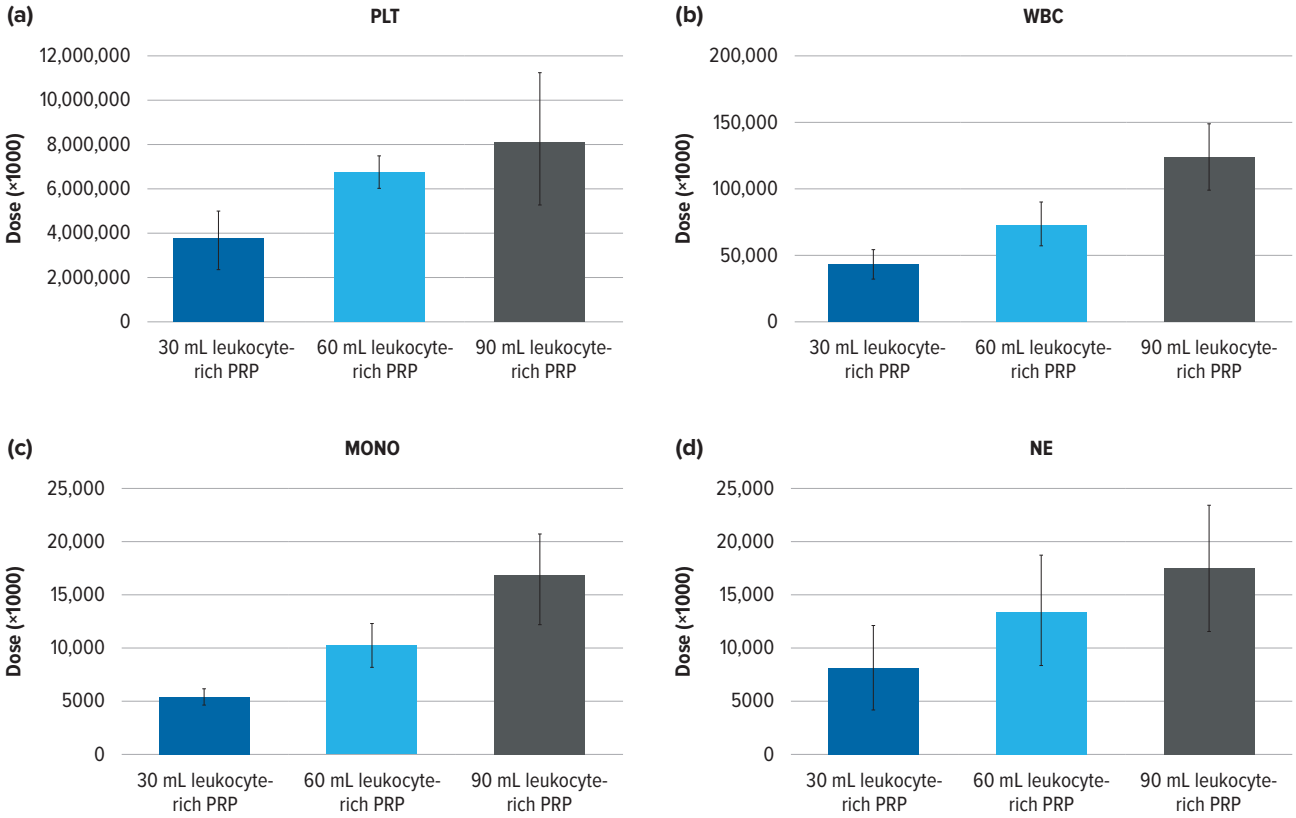


Table 3. Average delivered dose, with standard deviation, of the analyzed cells.

	30 mL	60 mL	90 mL
PLT (×1000)	3,714,058 ± 1,204,639	6,644,400 ± 838,334	8,122,075 ± 2,802,160
WBC (×1000)	43,602 ± 9287	71,360 ± 16,850	122,767 ± 25,367
MONO (×1000)	5253 ± 1068	10,252 ± 1978	16,623 ± 4442
NE (×1000)	7931 ± 3826	13,384 ± 4752	17,422 ± 5792

DISCUSSION

Based on the data presented here, the ACP Max™ system spin regimen can be successfully altered to create leukocyte-rich PRP instead of the normal leukocyte-poor PRP. PRPs were created that concentrated white blood cells 1.95×, 2.77×, and 3.93× over baseline for 30, 60, and 90 mL regimens, respectively. More specifically, monocytes were concentrated 2.92×, 5.57×, and 8.02× over baseline, respectively, while neutrophils were concentrated 0.68×, 0.83×, and 1.48× over baseline, respectively. Additionally, platelets were concentrated 3.89×, 6.88×, and 8.21× over baseline for 30, 60, and 90 mL, respectively. The delivered dose of cells followed similar trends to the fold change, with all white blood cell counts significantly increasing with larger spin volumes. It is important to note that the doses may differ between patients, depending on their baseline cell concentrations.

According to the PAW classification of PRP, these new PRP formulations would be classified as P4-Aα, as the platelet concentrations exceed 1.25M/µL, while the white blood cell and neutrophil counts are above baseline. In contrast, the normal ACP Max PRP is classified as P4-Bβ.[3] PRP is a common conservative treatment employed by orthopedic surgeons, with different compositions of PRP being used to treat different ailments. Notably, leukocyte-rich PRP may be better suited to a tendon environment, as leukocyte-rich PRPs have been shown to stimulate tenocyte proliferation better.[4,5] Conversely, leukocyte-poor PRP may be better suited to joint environments, as leukocyte- and red blood cell-rich concentrates have been shown to cause greater synoviocyte cell death than leukocyte-poor concentrates.[5,6] The ACP Max system normally produces leukocyte-poor PRP. However, this study demonstrates that it can be used to create leukocyte-rich PRP with only slight changes to the spin regimen, allowing surgeons to tailor their PRP preparation to the treatment.

References

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