

# THE USE OF PLATELET RICH PLASMA WITH BONE MARROW ASPIRATE IN PUDDU TIBIAL OSTEOTOMY

Caio Oliveira D'Elia<sup>1</sup>, Márcia Uchoa de Rezende<sup>2</sup>, Alexandre Carneiro Bitar<sup>3</sup>, Nelson Tatsui<sup>4</sup>, José Ricardo Pécora<sup>5</sup>, Gilberto Luis Camanho<sup>6</sup>

## ABSTRACT

**Objective:** The present study was performed in order to evaluate the use of platelet rich plasma associated to bone marrow aspirate, substituting autologous iliac bone graft in medial opening wedge osteotomy (OWHTO). **Methods:** Twenty-five patients were submitted to tibial opening wedge osteotomy, being divided into two groups. Iliac group: 14 patients submitted to OWHTO, using autologous iliac bone graft to fill the gap. PRP group: 11 patients using platelet rich plasma associated to bone marrow aspirate to fill the gap. We evaluated bleeding (hemoglobin and hematocrit levels) and pain (visual analogic scale-VAS),

then we compared the groups regarding these variables. **Results:** Differences between the groups were not found regarding hemoglobin levels ( $p=0.820$ ) and hematocrit levels ( $p=0.323$ ). The groups were not different regarding pain measured with VAS ( $p=0.538$ ). **Conclusion:** The use of platelet rich plasma associated to bone marrow aspirate in medial opening wedge osteotomy did not offer advantages over autologous iliac bone graft regarding bleeding and pain.

**Keywords** – *Platelet rich plasma; Osteotomy; Tibia; Bone substitutes*

## INTRODUCTION

Platelet-rich plasma (PRP) refers to a preparation obtained from autologous blood. Inside platelets, there are granules filled with growth factors, among the most important are platelet-derived growth factor (PDGF), transforming growth factor-beta (TGF- $\beta$ ) and vascular endothelial growth factor (VEGF)<sup>(1,2)</sup>.

Growth factors are polypeptides that are usually synthesized by specific tissues and act as local regulators of cellular function. These growth factors bind to receptors on the target cell membrane, activating an intracellular process that produces proteins to be used within the cell or exported<sup>(3)</sup>.

The normal concentration of platelets in peripheral blood is from 150,000/ $\mu$ l to 350,000/ $\mu$ l on average. For the PRP to be most efficiency, platelet concentration should be around 1,000,000/ $\mu$ l<sup>(1,2,4)</sup>. There are several methods for obtaining PRP<sup>(5)</sup>, each with its peculiar ability to concentrate the platelets, as well as in the release of growth factors by platelets.

The clinical use of PRP has grown considerably and has been applied in various situations: knee arthroplasty (TKA), tendon repair, treatment of cartilage lesions, and as a bone substitute<sup>(6-16)</sup>. PRP has been shown to reduce bleeding, pain, and the occurrence of arthrofibrosis when applied in the perioperative period in TKA<sup>(6,7)</sup>.

1 – Post-graduate Student, Department of Orthopedics and Traumatology, School of Medicine, USP.

2 – Assistant Physician, Knee Group, Institute of Orthopedics and Traumatology, HC/FMUSP.

3 – Orthopedist, Institute of Orthopedics and Traumatology, HC/FMUSP.

4 – Assistant Physician, Department of Hematology, HC/FMUSP.

5 – Head, Knee Group, Institute of Orthopedics and Traumatology, HC/FMUSP.

6 – Associate Professor, School of Medicine, Universidade de São Paulo.

Correspondence: Rua Dr. Ovídio Pires de Campos, 333 – São Paulo, SP, E-mail:caiodelia.vita@gmail.com

We declare no conflict of interest in this article.

The present study aims to evaluate the results with regard to pain and bleeding obtained with the application of PRP in the proximal tibial osteotomies by addition of a medial wedge (TOAMW).

## METHODS

This study was approved by the Research Ethics Committee of the Universidade de São Paulo. All patients signed an informed consent form.

The criteria for inclusion in the study were patients with varus knee deformity evaluated using a panoramic weight-bearing radiograph of the lower limbs, patients between 25 and 60 years of age, absence of systemic inflammatory diseases (rheumatoid arthritis, lupus, etc.), body mass index (BMI) less than 30kg/m<sup>2</sup>(17), need for correction with the use of wedges between 10 mm and 15 mm. Diagnosis of unicompartmental osteoarthritis, chronic ligament deficiencies, or deformities of the lower limbs. The indications for valgus osteotomy complied with those recommended in the literature(18-20).

The exclusion criteria were loss to follow-up and patient request to be excluded from the study. We studied 25 patients randomly divided into two groups by drawing lots with replacement performed on the day surgery had been scheduled. All surgical procedures in this study were performed by the same surgeon.

Patients were separated into two groups. The iliac group (IG) was comprised of 14 patients who underwent osteotomy with the use of an autologous iliac graft at the osteotomy site. The PRP group (PRPG) was comprised of 11 patients who underwent osteotomy with the use of a bone substitute composed of platelet-rich plasma (PRP) and bone marrow aspirate. We called this bone substitute “biological bone graft”.

The most frequent diagnosis in the two groups was chronic injury of the anterior cruciate ligament (ACL) (Table 1).

**Table 1** – Distribution of patients in the iliac and PRP groups according to diagnosis.

		Group		TOTAL
		Iliac	PRP	
<b>Diagnosis</b> <b>p = 0.168</b>	<b>Chronic injury of the ACL1</b>	6	9	15
		42.9%	81.2%	60.0%
	<b>Medial OA2</b>	5	1	6
		35.7%	9.1%	24.0%
	<b>Deformities3</b>	3	1	4
		21.4%	9.1%	16.0%
<b>Total</b>		14	11	25
		100%	100%	100%

P value regarding the Fisher's exact test.

1Double varus or triple varus.

2Also includes osteonecrosis.

3Genu varus or fracture sequela.

The groups were homogeneous with regard to the size of the wedges used (Table 2).The average age of the patients in the iliac and PRP groups was 45.9 and 37.8 years, respectively (p = 0.014).The technique employed to perform the osteotomy was, as noted earlier, the addition of a medial wedge, similar to the planning and surgical technique described by Puddu et al.(19,20).

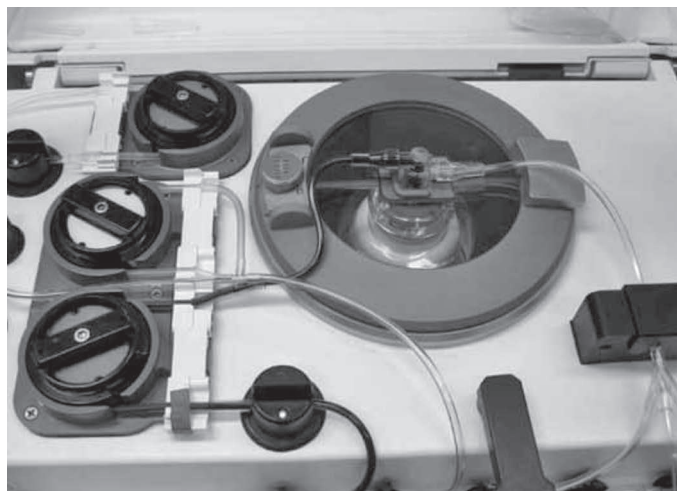
**Table 2** – Distribution of patients in the iliac and PRP groups according to size of the wedge used.

		Group		TOTAL
		Iliac	PRP	
<b>Wedge size</b> <b>p = 0.885</b>	10.0 mm	5	3	8
		35.7%	27.3%	32.0%
	12.5 mm	5	5	10
		35.7%	45.5%	40.0%
15.0 mm	4	3	7	
	28.6%	27.3%	28.0%	
<b>TOTAL</b>		14	11	25
		100%	100%	100%

P value regarding the Fisher's exact test.

After osteotomy and plate fixation, the space created in the metaphyseal region of the tibia was filled with one of the grafts being evaluated (iliac or biological).

To collect the platelets, we used a Haemonetics MCS+ 9000 automated cell separator and a 995-E plateletpheresis-specific kit (Haemonetics Corp.). In this system, the patient's blood was drained through a venipuncture in the antecubital fossa into a separation device under continuous centrifugation (Figure 1).



**Figure 1** – Haemonetics MCS+ 9000 automated cell separator.

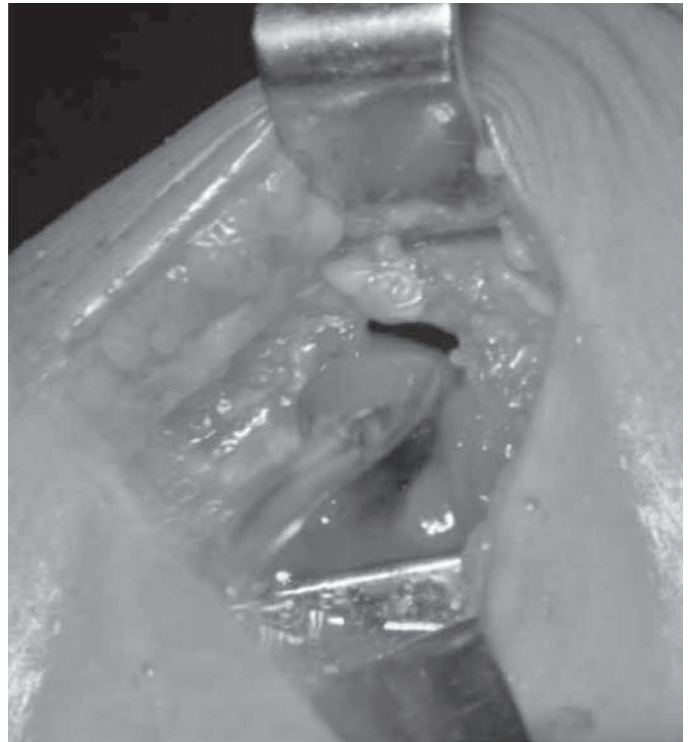
After blood fractionation, a refractive optical analyzer singled out the platelet layer and directed its collection into a specific disposable bag. The remaining blood was completely reinfused into the patient, marking the end of a cycle. Sodium citrate was used as an anticoagulant in a proportion of one for each 9 ml of processed whole blood. Two cycles were usually performed, yielding approximately 70 ml of platelet concentrate.

The bone marrow aspirate was obtained from the iliac crest via percutaneous puncture using the standard technique (Figure 2). Six punctures were made to obtain about 12 ml of bone marrow, not to exceed a volume of 2 ml per puncture. After each puncture, the needle was repositioned<sup>(21)</sup>. This material was anticoagulated with sodium citrate in the ratio of 1:5 citrate:bone marrow.

The biological bone graft was formed by adding the bone marrow aspirate to the platelet concentrate. Once the gel was formed, the surgeon placed the material in the surgical site (Figure 3).

For the evaluation of bleeding as a result of surgery, the RBC indices of hemoglobin (Hb) and hematocrit (Ht) were obtained<sup>(7)</sup>. These indices were obtained on the day of surgery and 24 hours postoperatively. In this manner, the variation of these indices before and after surgery was obtained. The groups were compared with respect to these variables (change in hemoglobin and hematocrit) by Student's t-test.

Pain assessment was performed using the visual analog scale for pain<sup>(22,23)</sup>, performed 24 hours after surgery. The groups were compared with respect to this variable by Student's t-test.

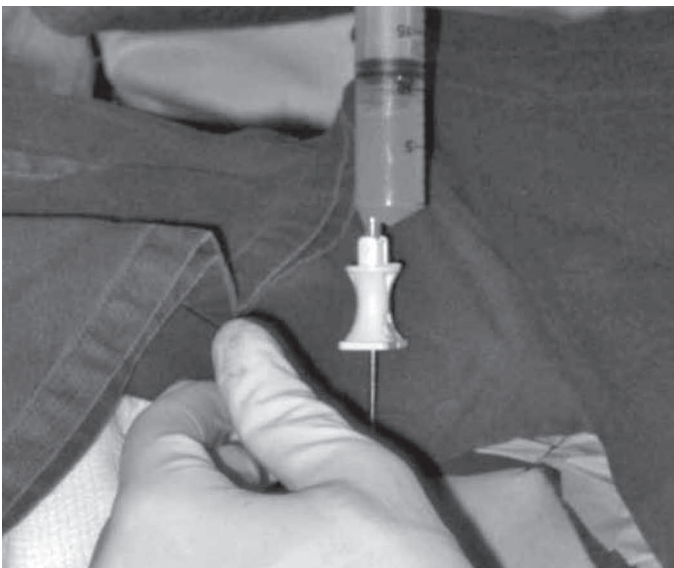


**Figure 3** – Placing the biological graft at the osteotomy site.

## RESULTS

There were no differences between the iliac and PRP groups in relation to the variation of levels of hemoglobin and hematocrit pre- and postoperatively (Tables 3 and 4).

Pain assessment performed using the visual analog scale for pain (VAS) was not different between groups (Table 5).



**Figure 2** – Obtaining the bone marrow aspirate.



**Table 3** – Changes in hemoglobin (Hb) levels.

		Iliac (n = 14)	PRP (n = 11)	Total
<b>Difference in Hb</b>	Mean	2.3	2.2	2.3
	SD	1.0	0.7	0.9
<b>pre- and postoperatively</b> <b>p = 0.820</b>	Median	2.4	2.1	2.2
	Minimum	-0.3	0.9	-0.3
	Maximum	3.7	3.2	3.7

P value regarding the Student's t-test.

**Tabela 4** – Changes in hematocrit (Ht) levels.

		Iliac (n = 14)	PRP (n = 11)	Total
<b>Difference in Ht</b>	Mean	6.4	5.3	5.9
	SD	2.3	3.1	2.7
<b>pre- and postoperatively</b> <b>p = 0.323</b>	Median	5.9	5.7	5.7
	Minimum	2.3	0.3	0.3
	Maximum	10.8	9.2	10.8

P value regarding the Student's t-test.

**Table 5** – Visual analog scale for pain<sup>(22)</sup>.

		Iliac (n = 14)	PRP (n = 11)	Total
<b>Pain scale</b>	Mean	5.1	4.4	4.8
	SD	2.9	2.7	2.8
<b>p = 0.538</b>	Median	6.0	3.0	5.0
	Minimum	1.0	1.0	1.0
	Maximum	9.0	9.0	9.0

P value regarding the Student's t-test.

## DISCUSSION

The valgus osteotomy of the tibia using the technique of adding a medial wedge is a common procedure in the treatment of several orthopedic pathologies, and today it has been thoroughly standardized<sup>(18,24-27)</sup>.

## REFERENCES

- Marx RE. Platelet-rich plasma (PRP): what is PRP and what is not PRP? *Implant Dent.* 2001;10(4):225-8.
- Marx RE. Platelet-rich plasma: evidence to support its use. *J Oral Maxillofac Surg.* 2004;62(4):489-96.
- Lind M. Growth factors: possible new clinical tools. A review. *Acta Orthop Scand.* 1996;67(4):407-17.
- Eppley BL, Woodell JE, Higgins J. Platelet quantification and growth factor analysis from platelet-rich plasma: implications for wound healing. *Plast Re-*

Platelet-rich plasma (PRP) has important osteoinductive properties, as has been demonstrated by several experimental studies<sup>(11,28-31)</sup>. PRP has wide clinical application in the field of oral and maxillofacial surgery and is used as an osteopromotive agent in various situations<sup>(31-33)</sup>. Its clinical use in orthopedics has been increasing, despite the absence of randomized prospective studies to assess the results of its application<sup>(30,34-36)</sup>.

In a study that aims to evaluate the effectiveness of PRP alone or in combination with other materials, it is important to remember that different methods are used to obtain PRP<sup>(1,5,30)</sup>, and the ability to obtain high concentrations of platelets is variable among the available methods. Most systems used in clinical practice are based on centrifugation; the centrifuges used for this purpose have been developed for diagnosis and not to obtain PRP, which is often generated with less than ideal platelets level. The method of obtaining PRP used in our study<sup>(37)</sup> is able to offer a platelet concentration above 1,000,000/ $\mu$ l.

The need for an autologous graft in TOAMW is considered a disadvantage by many surgeons, because of the morbidity associated with obtaining the autologous graft<sup>(38,39)</sup>. Pain is a frequent complaint in patients who undergo obtainment of autologous grafting. In our study there was no difference in pain between the groups after 24 hours ( $p = 0.538$ ).

PRP was used in TKA<sup>(6,7)</sup>, reducing bleeding and the occurrence of arthrofibrosis. In our model of the clinical application, the proximal tibial osteotomy by addition of a medial wedge, there was no difference in bleeding between the groups with the use of PRP associated with bone marrow aspirate ( $p = 0.820$  and  $p = 0.323$ ).

## CONCLUSION

The use of PRP associated with bone marrow in TOAMW showed no advantages over the use of autologous iliac bone graft with regard to pain and bleeding.

*constr Surg.* 2004;114(6):1502-8.

- Everts PA, Brown Mahoney C, Hoffmann JJ, Schonberger JP, Box HA, van Zundert A, et al. Platelet-rich plasma preparation using three devices: implications for platelet activation and platelet growth factor release. *Growth Factors.* 2006;24(3):165-71.
- Everts PA, Devilee RJ, Brown Mahoney C, Eeftinck-Schattenkerk M, Box HA, Knape JT, et al. Platelet gel and fibrin sealant reduce allogeneic blood transfusions in total knee arthroplasty. *Acta Anaesthesiol Scand.* 2006;50(5):593-9.

7. Everts PA, Devilee RJ, Oosterbos CJ, Mahoney CB, Schattenkerk ME, Knappe JT, et al. Autologous platelet gel and fibrin sealant enhance the efficacy of total knee arthroplasty: improved range of motion, decreased length of stay and a reduced incidence of arthrofibrosis. *Knee Surg Sports Traumatol Arthrosc.* 2007;15(7):888-94.
8. Sanchez M, Azofra J, Anitua E, Andia I, Padilla S, Santisteban J, et al. Plasma rich in growth factors to treat an articular cartilage avulsion: a case report. *Med Sci Sports Exerc.* 2003;35(10):1648-52.
9. Sanchez M, Anitua E, Azofra J, Andia I, Padilla S, Mujika I. Comparison of surgically repaired Achilles tendon tears using platelet-rich fibrin matrices. *Am J Sports Med.* 2007;35(2):245-51.
10. Lowery GL, Kulkarni S, Pennisi AE. Use of autologous growth factors in lumbar spinal fusion. *Bone.* 1999;25(2 Suppl):47S-50S.
11. Gandhi A, Doumas C, O'Connor JP, Parsons JR, Lin SS. The effects of local platelet rich plasma delivery on diabetic fracture healing. *Bone.* 2006;38(4):540-6.
12. Filho Cerruti H, Kerkis I, Kerkis A, Tatsui NH, da Costa Neves A, Bueno DF, et al. Allogeneous bone grafts improved by bone marrow stem cells and platelet growth factors: clinical case reports. *Artif Organs.* 2007;31(4):268-73.
13. Choi BH, Im CJ, Huh JY, Suh JJ, Lee SH. Effect of platelet-rich plasma on bone regeneration in autogenous bone graft. *Int J Oral Maxillofac Surg.* 2004;33(1):56-9.
14. Bielecki T, Gazdzik TS, Szczepanski T. Benefit of percutaneous injection of autologous platelet-leukocyte-rich gel in patients with delayed union and nonunion. *Eur Surg Res.* 2008;40(3):289-96.
15. Bielecki T, Gazdzik TS, Szczepanski T. Re: "The effects of local platelet rich plasma delivery on diabetic fracture healing". What do we use: Platelet-rich plasma or platelet-rich gel? *Bone.* 2006;39(6):1388.
16. Dallari D, Savarino L, Stagni C, Cenni E, Cenacchi A, Fornasari PM, et al. Enhanced tibial osteotomy healing with use of bone grafts supplemented with platelet gel or platelet gel and bone marrow stromal cells. *J Bone Joint Surg Am.* 2007;89(11):2413-20.
17. Romero-Corral A, Somers VK, Sierra-Johnson J, Thomas RJ, Collazo-Clavell ML, Korinek J, et al. Accuracy of body mass index in diagnosing obesity in the adult general population. *Int J Obes (Lond).* 2008;32(6):959-66.
18. Asik M, Sen C, Kilic B, Goksan SB, Ciftci F, Taser OF. High tibial osteotomy with Puddu plate for the treatment of varus gonarthrosis. *Knee Surg Sports Traumatol Arthrosc.* 2006;14(10):948-54.
19. Franco V, Cerullo G, Cipolla M, Gianni E, Puddu G. Open wedge high tibial osteotomy. *Tech Knee Surg.* 2002;1(1):43-53.
20. Puddu G, Cipolla M, Cerullo G, Franco V, Gianni E. Osteotomies: the surgical treatment of the valgus knee. *Sports Med Arthrosc.* 2007;15(1):15-22.
21. Muschler GF, Boehm C, Easley K. Aspiration to obtain osteoblast progenitor cells from human bone marrow: the influence of aspiration volume. *J Bone Joint Surg Am.* 1997;79(11):1699-709.
22. Willestaedt H, Levander G, Hult L. Studies in osteogenesis. *Acta Orthop Scand.* 1950;19(4):419-32.
23. DeLoach LJ, Higgins MS, Caplan AB, Stiff JL. The visual analog scale in the immediate postoperative period: intrasubject variability and correlation with a numeric scale. *Anesth Analg.* 1998;86(1):102-6.
24. Amendola A, Panarella L. High tibial osteotomy for the treatment of unicompartmental arthritis of the knee. *Orthop Clin North Am.* 2005;36(4):497-504.
25. Amendola A, Fowler PJ, Litchfield R, Kirkley S, Clatworthy M. Opening wedge high tibial osteotomy using a novel technique: early results and complications. *J Knee Surg.* 2004;17(3):164-9.
26. Brinkman JM, Lobenhoffer P, Agneskirchner JD, Staubli AE, Wymenga AB, van Heerwaarden RJ. Osteotomies around the knee: patient selection, stability of fixation and bone healing in high tibial osteotomies. *J Bone Joint Surg Br.* 2008;90(12):1548-57.
27. Koshino T, Murase T, Saito T. Medial opening-wedge high tibial osteotomy with use of porous hydroxyapatite to treat medial compartment osteoarthritis of the knee. *J Bone Joint Surg Am.* 2003;85-A(1):78-85.
28. Nash TJ, Howlett CR, Martin C, Steele J, Johnson KA, Hicklin DJ. Effect of platelet-derived growth factor on tibial osteotomies in rabbits. *Bone.* 1994;15(2):203-8.
29. Anitua E, Andia I, Ardanza B, Nurden P, Nurden AT. Autologous platelets as a source of proteins for healing and tissue regeneration. *Thromb Haemost.* 2004;91(1):4-15.
30. Floryan KM, Berghoff WJ. Intraoperative use of autologous platelet-rich and platelet-poor plasma for orthopedic surgery patients. *AORN J.* 2004;80(4):668-74.
31. Grageda E. Platelet-rich plasma and bone graft materials: a review and a standardized research protocol. *Implant Dent.* 2004;13(4):301-9.
32. Marx RE, Carlson ER, Eichstaedt RM, Schimmele SR, Strauss JE, Georgeff KR. Platelet-rich plasma: Growth factor enhancement for bone grafts. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.* 1998;85(6):638-46.
33. Sanchez AR, Sheridan PJ, Kupp LI. Is platelet-rich plasma the perfect enhancement factor? A current review. *Int J Oral Maxillofac Implants.* 2003;18(1):93-103.
34. Roldan JC, Jepsen S, Miller J, Freitag S, Rueger DC, Acil Y, et al. Bone formation in the presence of platelet-rich plasma vs. bone morphogenetic protein-7. *Bone.* 2004;34(1):80-90.
35. Solheim E. Growth factors in bone. *Int Orthop.* 1998;22(6):410-6.
36. Mariconda M, Cozzolino F, Cozzolino A, D'Agostino E, Bove A, Milano C. Platelet gel supplementation in long bone nonunions treated by external fixation. *J Orthop Trauma.* 2008;22(5):342-5.
37. O'Neill EM, Zalewski WM, Eaton LJ, Popovsky MA, Pivacek LE, Ragno G, et al. Autologous platelet-rich plasma isolated using the Haemonetics Cell Saver 5 and Haemonetics MCS+ for the preparation of platelet gel. *Vox Sang.* 2001;81(3):172-5.
38. Warden SJ, Morris HG, Crossley KM, Brukner PD, Bennell KL. Delayed- and non-union following opening wedge high tibial osteotomy: surgeons' results from 182 completed cases. *Knee Surg Sports Traumatol Arthrosc.* 2005;13(1):34-7.
39. Spahn G. Complications in high tibial (medial opening wedge) osteotomy. *Arch Orthop Trauma Surg.* 2004;124(10):649-53.