

Ortobiológicos

conceito e atualização no emprego
de plasma rico em plaquetas e
lisados plaquetários no tratamento
de afecções articulares

MV. Me. Sarah Seidel

Sumário

Conceito e classificação
Processos de obtenção
Constituição produto final
Ação em tecido articular
Evidência científica
Perspectivas
Considerações finais

Conceitos

Ortobiológicos

Conjunto de produtos, autólogos ou não, formados por um concentrado de células ou fragmentos celulares

Hemocomponentes

Componentes lábeis do sangue submetidos a um processo **FÍSICO** que dá origem a um produto formado por parte do sangue total

Hemoderivados

Produtos obtidos a partir do plasma por meio de processos **FÍSICO-QUÍMICOS** ou **BIOTECNOLÓGICOS**

Conceitos

Plasma Rico em Plaquetas

Hemocomponente que concentra plaquetas acima dos níveis basais do sangue total, com ou sem enriquecimento leucocitário

DMOAD

Droga Modificadora da Doença Osteoartrósica

Métodos multimodais

Transplante

Estímulo celular

Arcabouço biodegradável

Hemocomponentes



Hemocomponentes

PRP

- Citrato de sódio
- Grande variabilidade
 - *Protocolo*
 - *Produto final*
- Tempo

PPA

- Heparina
- Incubação (24h)
- Filtração

PRF

- Sem anticoagulante
- Sólido

LP

- Ruptura membrana
- Liberação fatores
- Acelular

Liofilizado

- Congelamento
- Secagem
- Crioprotetores

Sistemas de classificação

| AUTOR | CLASSIFICAÇÃO | METODOLOGIA |
|-------------------------|--------------------|---|
| MISHRA et al. (2012) | Medicina esportiva | Concentração plaquetária Ativador exógeno Concentração leucocitária |
| EHRENFEST et al. (2014) | PRP PRF | Conteúdo celular Arquitetura de fibrina |
| DELONG et al. (2012) | PAW | Concentração plaquetária Ativador exógeno Concentração leucocitária |
| MAUTNER et al. (2015) | PLRA | Volume e frequência |
| MAGALON et al. (2016) | DEPA | Dose Eficiência Pureza Ativação |

Métodos de obtenção: PRP



Centrifugação

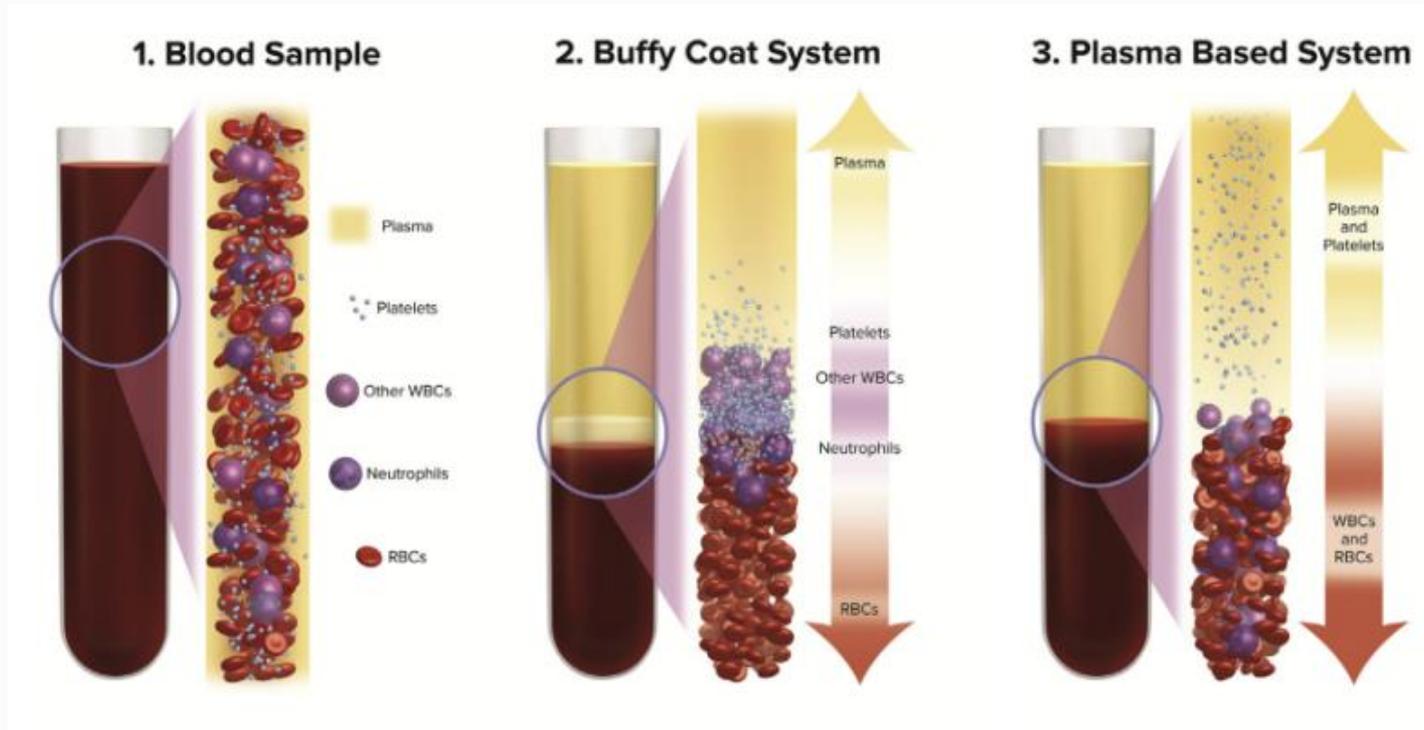


Plasmaferese



Filtração

Métodos de obtenção: PRP



Métodos de obtenção: PRP

Ativação?

Fracionamento

Homogeneização

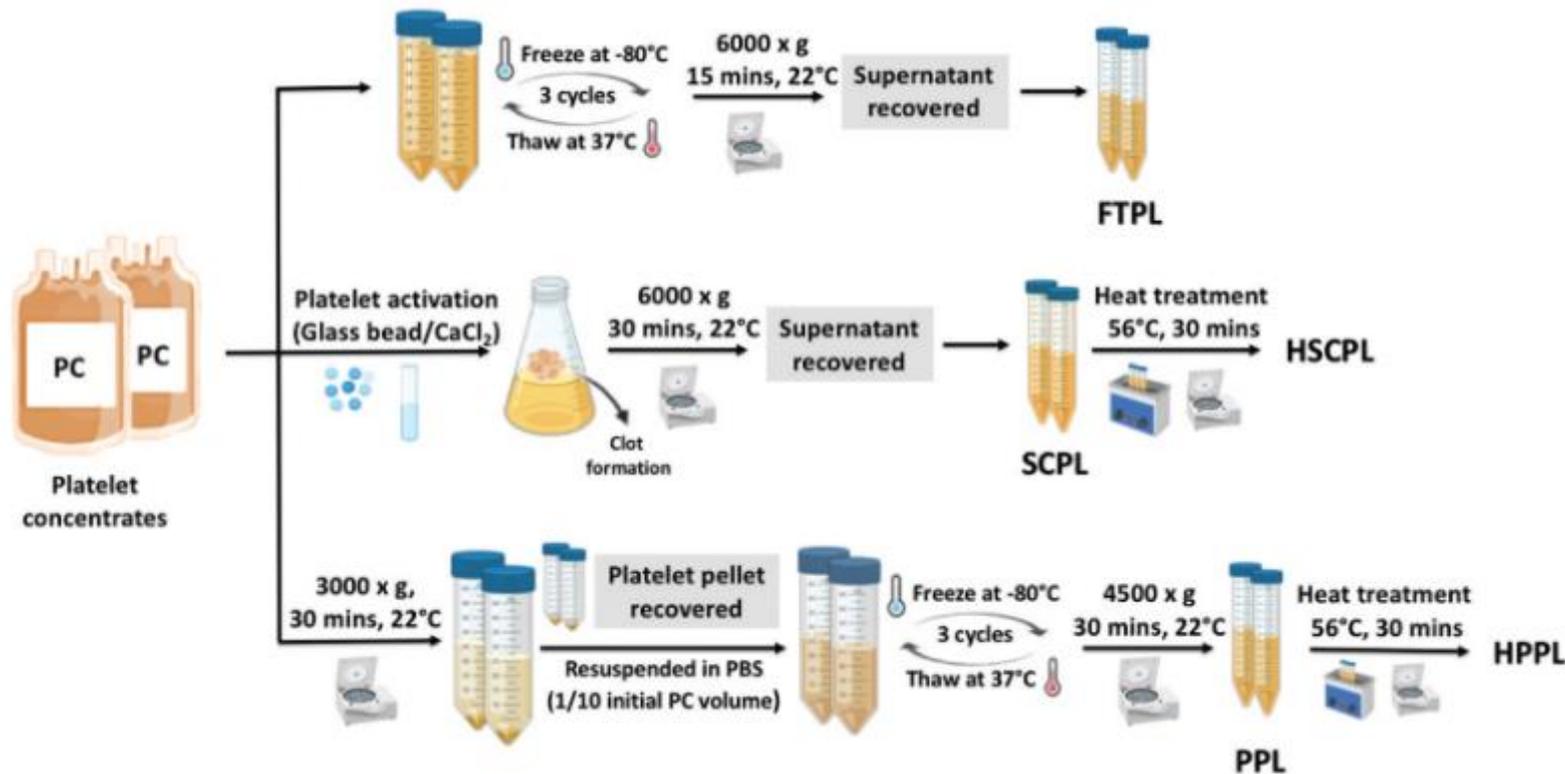
Proporção ST:AC

Anticoagulante: citrato

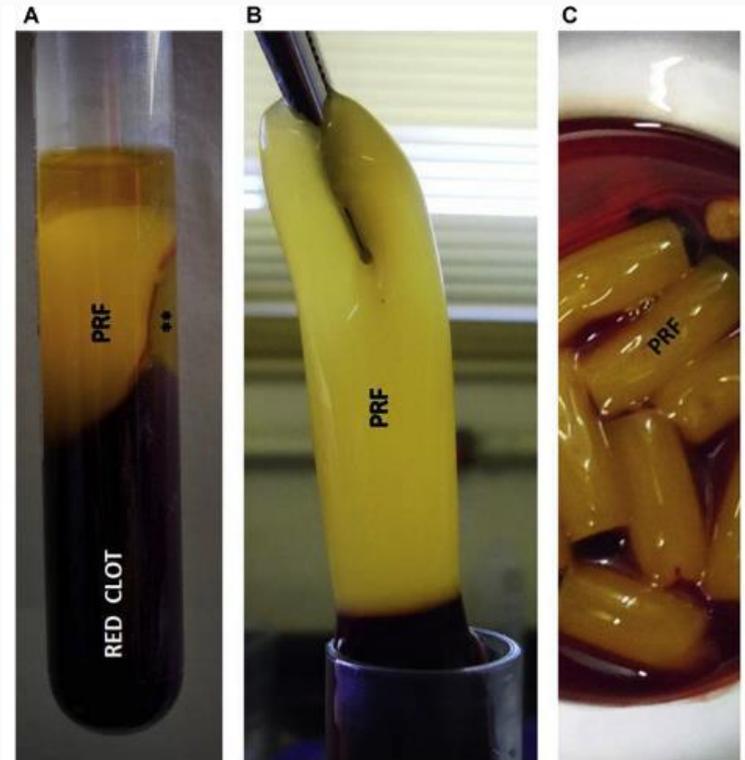
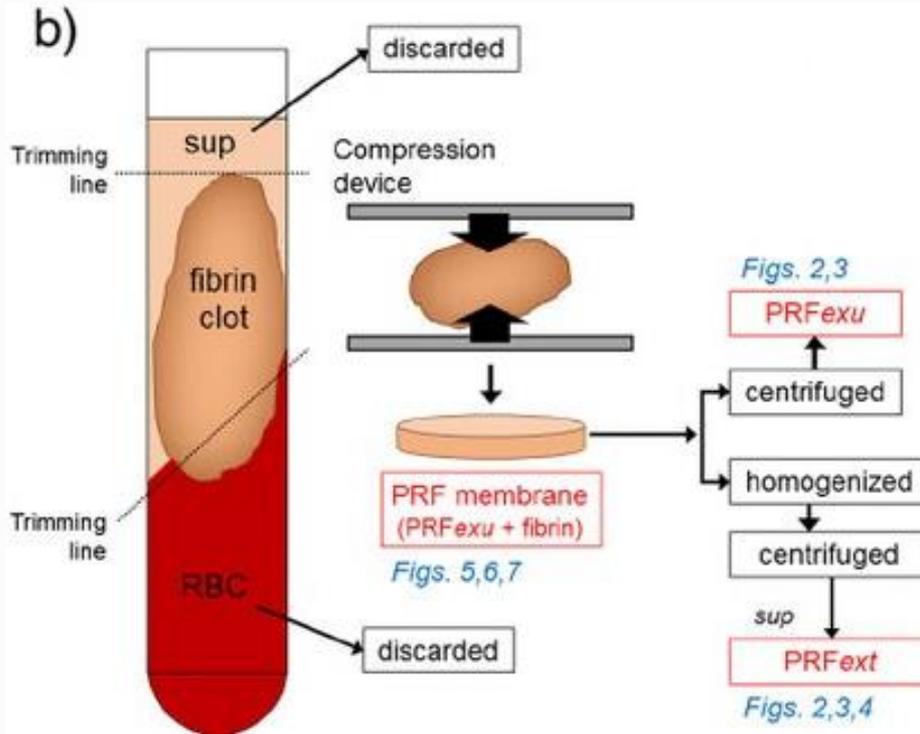
Calibre agulha

Antissepsia

Métodos de obtenção: Lisado Plaquetário



Métodos de obtenção: Plaqueta Rica em Fibrina



Jiménez-Aristazábal et al. Changes on the Structural Architecture and Growth Factor Release, and Degradation in Equine Platelet-Rich Fibrin Clots Cultured Over Time. 2019.

Qual protocolo escolher?



Article

Does Double Centrifugation Lead to Premature Platelet Aggregation and Decreased TGF- β 1 Concentrations in Equine Platelet-Rich Plasma?

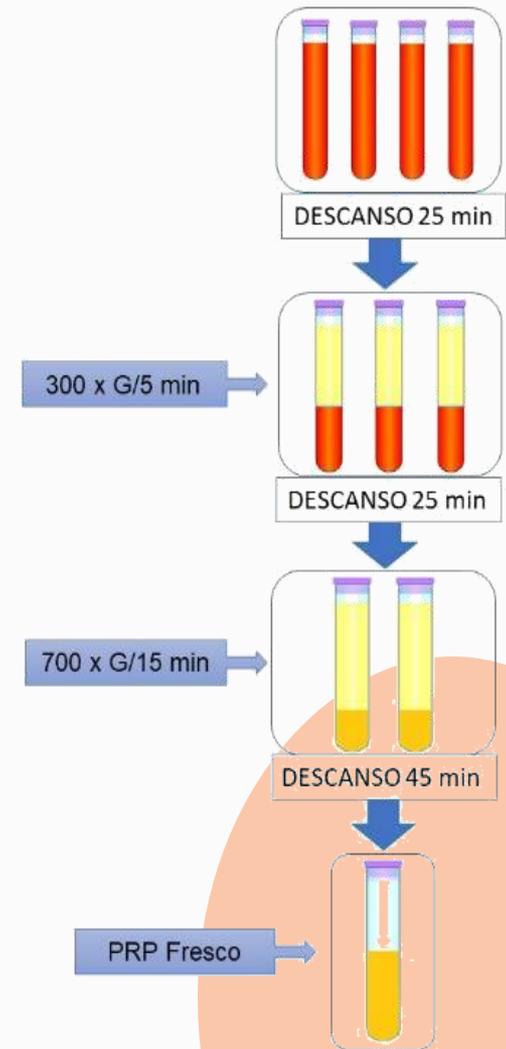
Sarah R. T. Seidel ^{1,*}, Cynthia P. Vendruscolo ¹, Juliana J. Moreira ¹, Joice Fülber ¹,
Tatiana F. Ottaiano ², Maria L. V. Oliva ², Yara M. Michelacci ² and Raquel Y. A. Baccarin ^{1,*}

¹ Departamento de Clínica Médica, Faculdade de Medicina Veterinária e Zootecnia, Universidade de São Paulo, São Paulo 05508-270, Brazil

² Departamento de Bioquímica, Escola Paulista de Medicina, UNIFESP, São Paulo 04023-062, Brazil

* Correspondence: sarahseidel@usp.br (S.R.T.S.); baccarin@usp.br (R.Y.A.B.); Tel.: +55-11-3091-1323 (R.Y.A.B.)

Received: 31 May 2019; Accepted: 31 July 2019; Published: 21 August 2019



Constituição produto final

VEGF
FC Endotelial Vascular
Aumenta angiogênese e permeabilidade vascular, mitogênico células endoteliais

HGF
FC de Hepatócito
Morfogênese, proliferação celular, anti-inflamatório, proteção, reparo e regeneração

EGF
FC Epidermal
Crescimento endotelial, angiogênese, regula colagenase, reparação pele

TGF- β
FC Transformante
Migração celular, neovascularização e angiogênese, síntese COL, inibe degradação MEC, ação mitogênica sobre FCs



KGF
FC Queratinócito
Crescimento e neoformação de queratinócitos

FGF
FC Fibroblástico
Reparo tecidual, crescimento e diferenciação de condrócitos e osteoblastos

IGF
FC Tipo Insulina
Formação MO, síntese FA e COL-1, proliferação e diferenciação de MSC

PDGF
FC Derivado de PLT
Reparo tecidual, crescimento e diferenciação de condrócitos e osteoblastos

CTGF
FC de Tecido Conjuntivo
Angiogênese, reparo cartilagem, fibrose e adesão PLT

Constituição produto final

VEGF
FC Endotelial Vascular
Aumenta angiogênese e permeabilidade vascular, mitogênico células endoteliais

HGF
FC de Hepatócito
Morfogênese, proliferação celular, anti-inflamatório, proteção, reparo e regeneração

EGF
FC Epidermal
Crescimento endotelial, angiogênese, regula colagenase, reparação pele

TGF- β
FC Transformante
Migração celular, neovascularização e angiogênese, síntese COL, inibe degradação MEC, ação mitogênica sobre FCs



KGF
FC Queratinócito
Crescimento e neoformação de queratinócitos

FGF
FC Fibroblástico
Reparo tecidual, crescimento e diferenciação de condrócitos e osteoblastos

IGF
FC Tipo Insulina
Formação MO, síntese FA e COL-1, proliferação e diferenciação de MSC

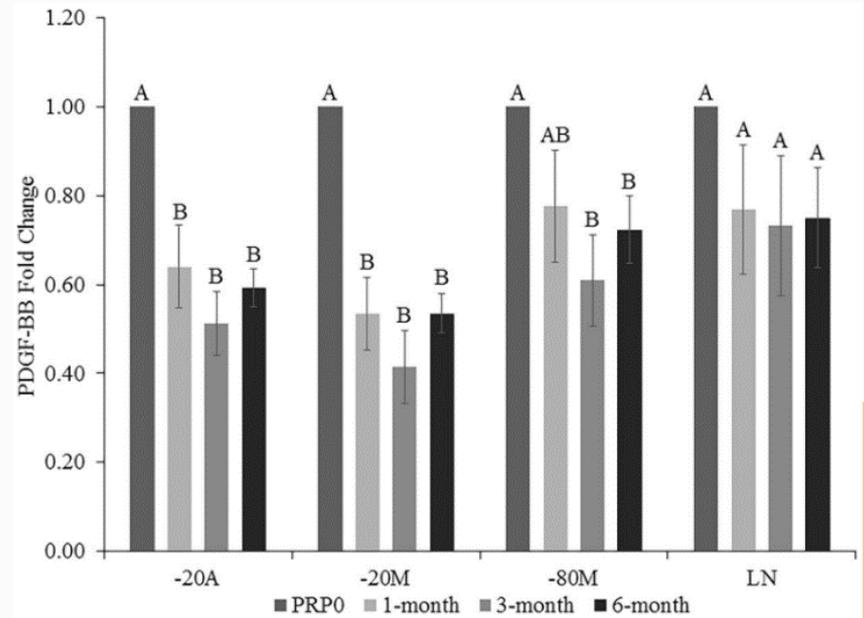
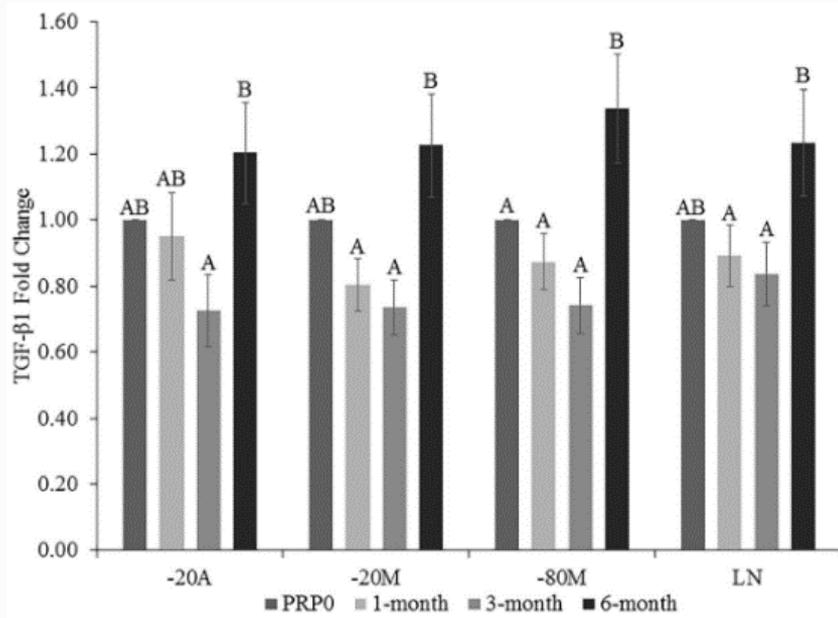
PDGF
FC Derivado de PLT
Reparo tecidual, crescimento e diferenciação de condrócitos e osteoblastos

CTGF
FC de Tecido Conjuntivo
Angiogênese, reparo cartilagem, fibrose e adesão PLT

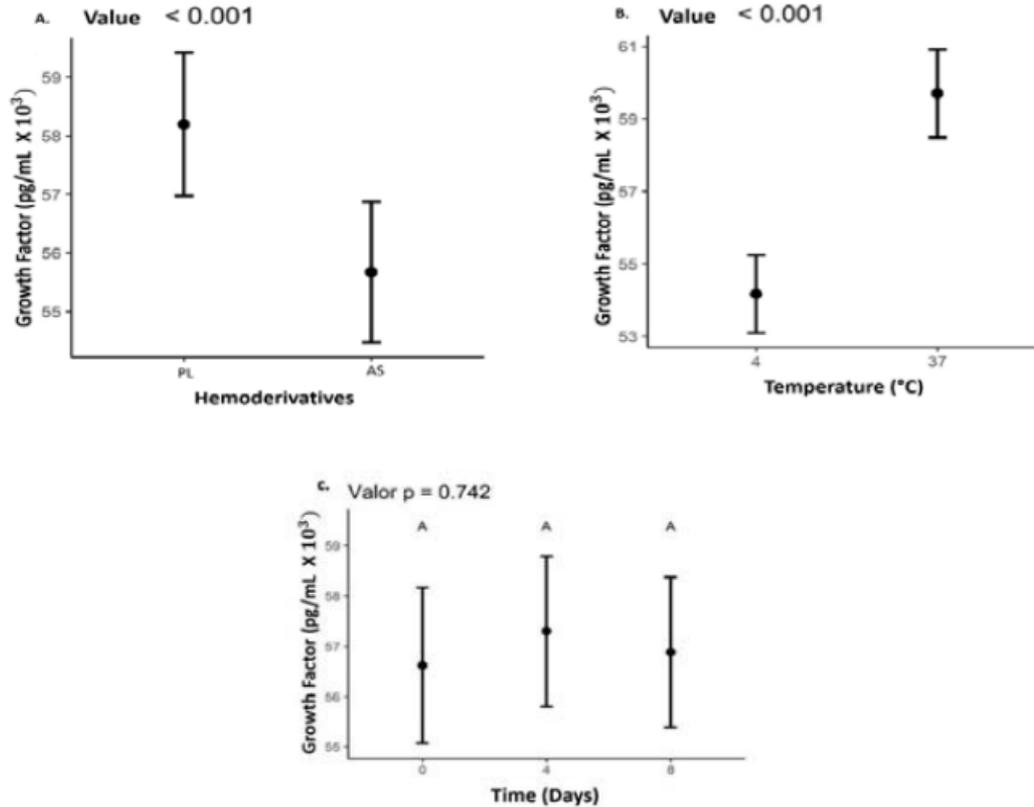
Constituição produto final

| | | | |
|------------------|---|---------------------|---|
| Plasma | | Leukocytes | |
| Proteins | Albumin, globulins, fibrinogen, complement, and clotting factors | Neutrophils | |
| Electrolytes | Chloride, sodium, potassium, and calcium | Primary granules | Myeloperoxidase, acid hydrolases, defensins, and serine proteases |
| Hormones | IGF-1, estrogens, progesterone, androgens, ACTH, and HGH | Secondary granules | Collagenase, lactoferrin, cathelicidin, bactericidal phagocytins, and lysozyme |
| Biomarkers | COMP, CD11b, protein C, microRNA, osteocalcin, and osteonectin | Tertiary granules | Gelatinase and proteases |
| Platelets | | Monocytes | Platelet-activating factor, TGF- β , VEGF, FGF, and EGF |
| Alpha granules | Adhesive proteins, clotting factors, and GFs PDGF, TGF- β , VEGF, FGF, EGF, and HGF | Erythrocytes | ATP, S-nitrosothiols, nitric oxide, hydrogen sulfide, hemoglobin, and free radicals |
| Dense bodies | Calcium and neurotransmitters | | |
| Lysosomes | Lysosomal enzymes | | |

Estabilidade produto final: PRP

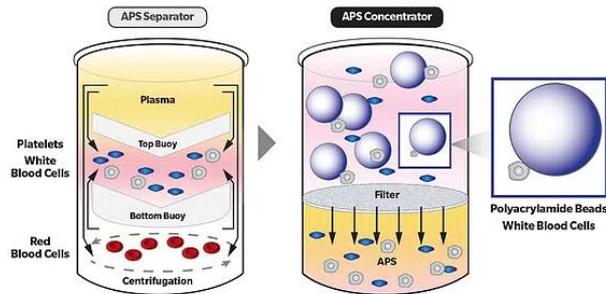


Estabilidade produto final: LP



Padronização

Pro-Stride Injection™



PRECISEPRP™
Equine 

PrecisePRP™ Equine
Leucoreduced allogeneic pooled freeze-dried platelet-rich plasma
VetStem, Inc.

PrecisePRP™ Equine
Leucoreduced allogeneic pooled freeze-dried platelet-rich plasma

For intra-articular injection in horses only

4.0×10^9 equine platelets / vial, freeze-dried

Single patient use vial for rehydration to 8 mL with sterile water

A freeze-dried species-specific source of concentrated platelets in plasma



Ação em tecido articular

Como o PRP pode influenciar o
ambiente articular?



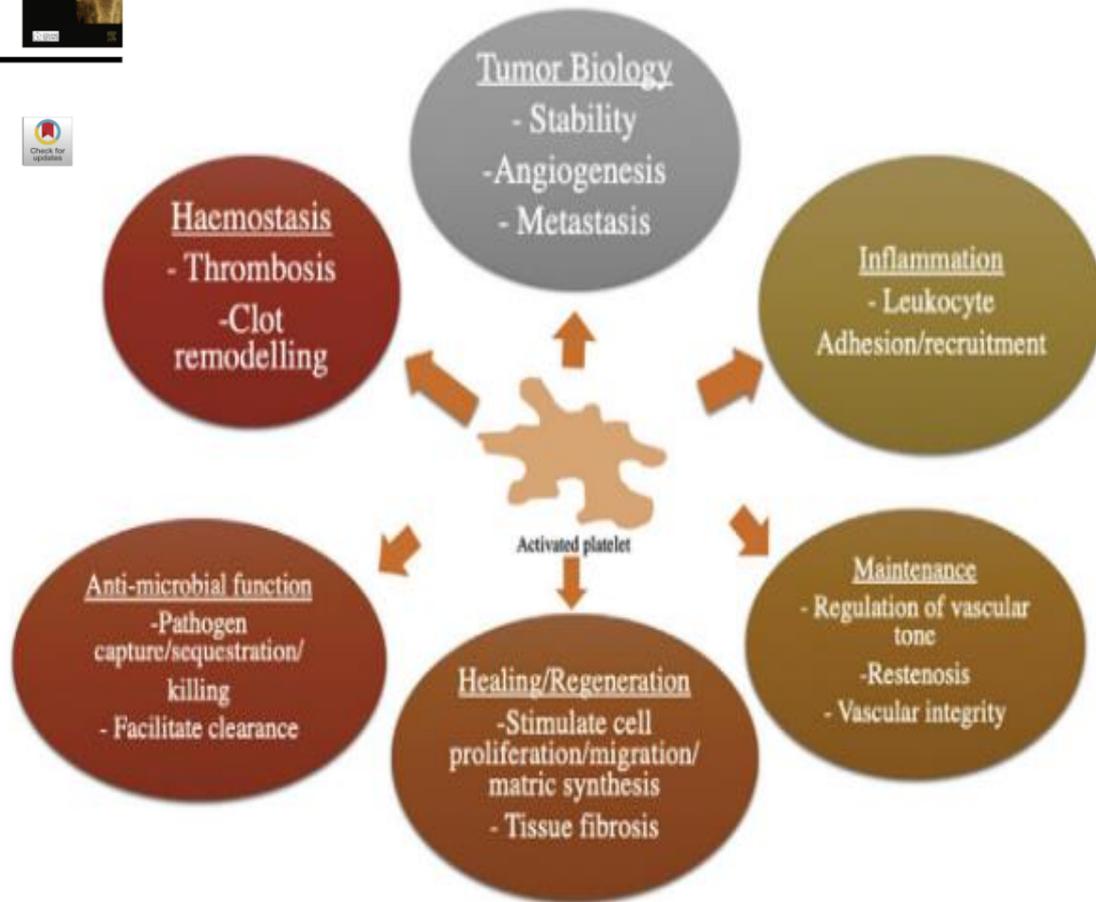
ELSEVIER



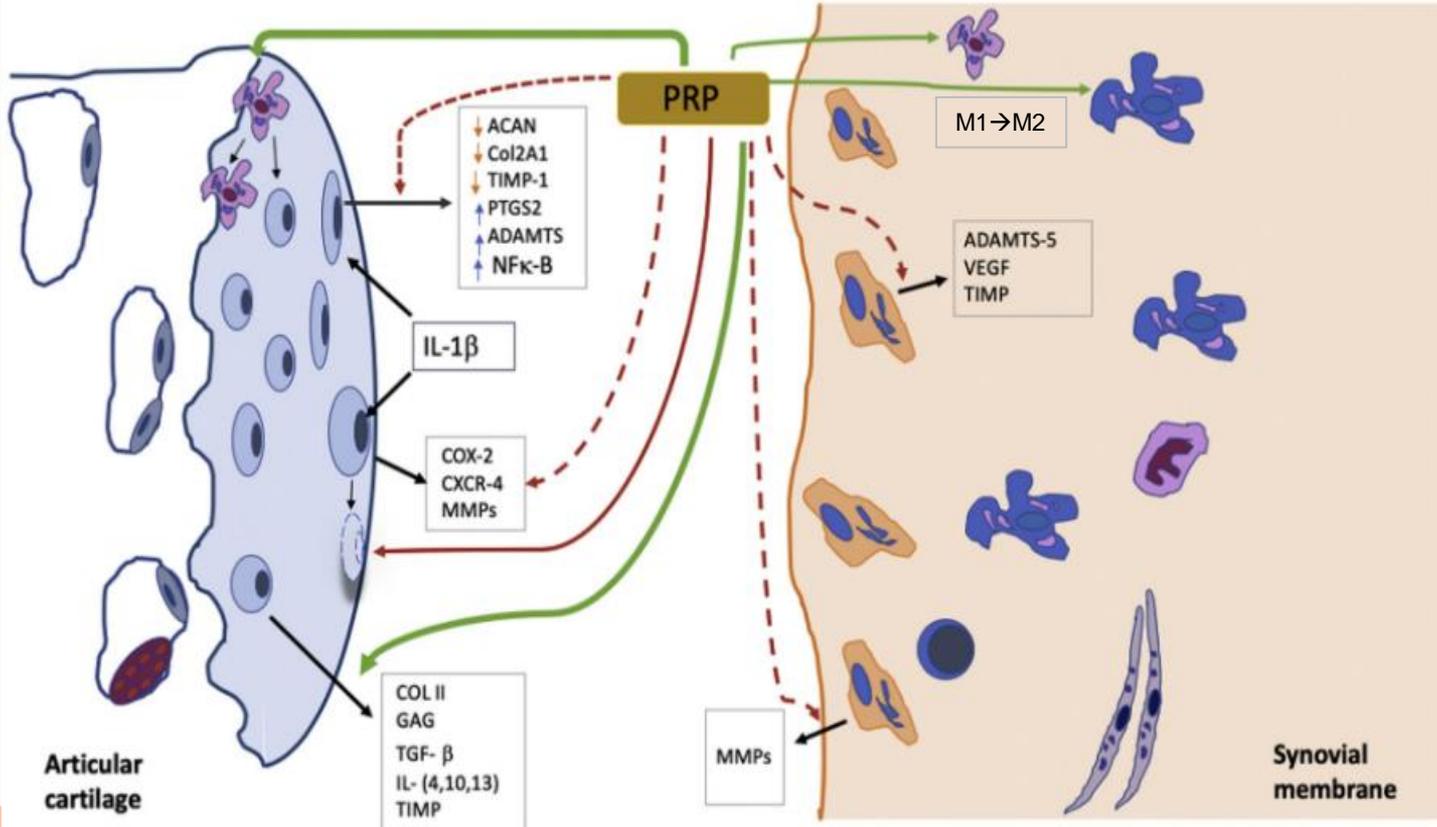
Review Article

Platelet-Rich Products and Their Application to Osteoarthritis

Livia Camargo Garbin ^{a,*}, Christine S. Olver ^b



Ação do PRP na OA



REVIEW

Biology of platelet-rich plasma and its clinical application in cartilage repair

Xuetao Xie^{1,2}, Changqing Zhang¹ and Rocky S Tuan^{2*}

Efeito anabólico

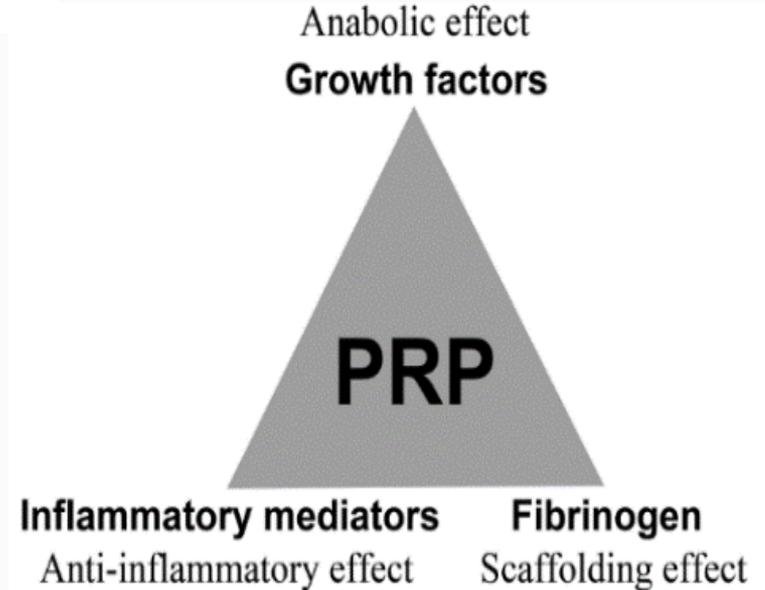
- Condrócitos, CTMs e sinoviócitos
- Acúmulo de MEC
- Produção de HA

Arcabouço bioativo

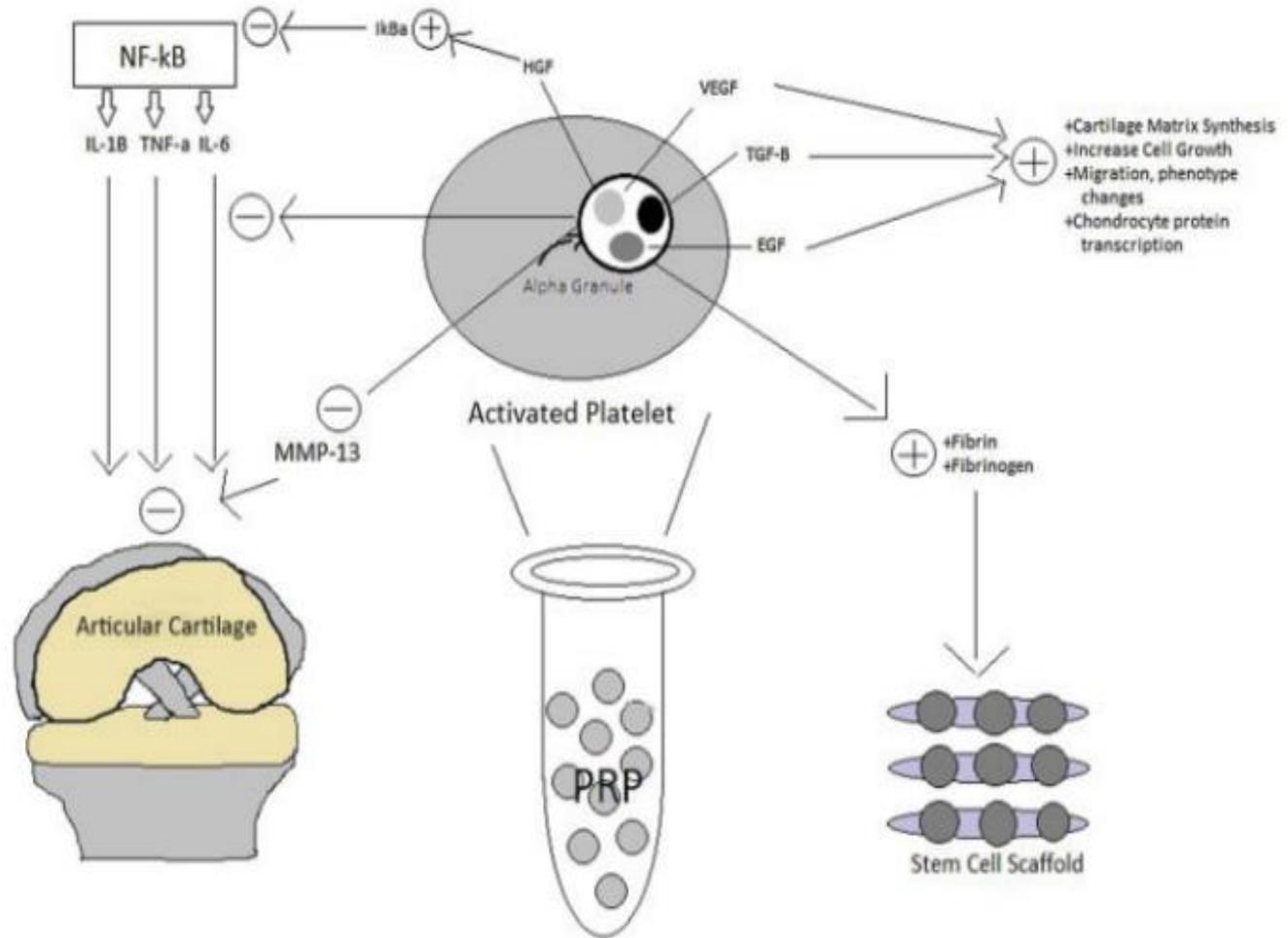
- Correção de defeitos
- Liberação gradativa

Mediadores inflamatórios

- Inibe inflamação
- Melhora sintomatologia OA



Inibição NF- κ B
Via catabólica
Transcrição de citocinas
IL-1 β , TNF α , IL-6
Diminui expressão MMP-13





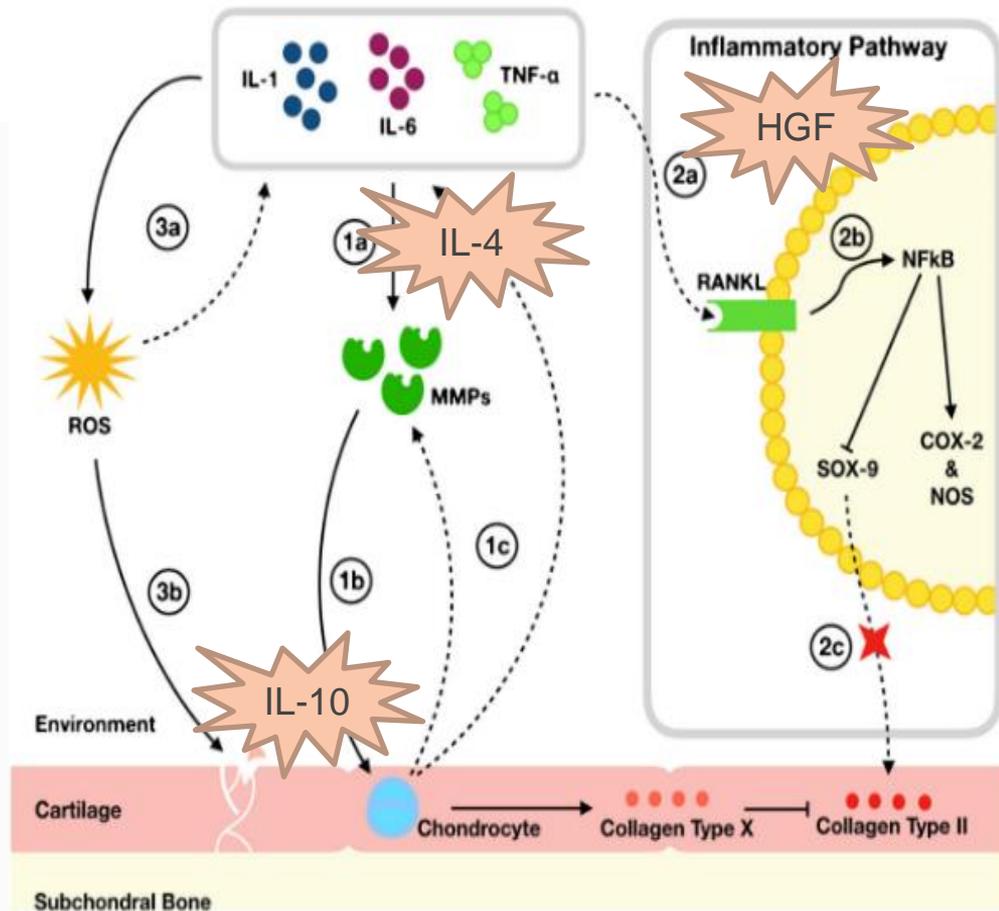
Platelet-Rich Plasma and Cartilage Repair

Mitchell I. Kennedy¹ • Kaitlyn Whitney¹ • Thos Evans² • Robert F. LaPrade^{1,2}

1
Síntese de MMPs,
Degradação COL
Produção de COL X,
Degradação cartilagem e condrócitos

2
RANKL ativa NFκB
Diminui SOX-9 e COL-II
Aumenta COX2 e NOS

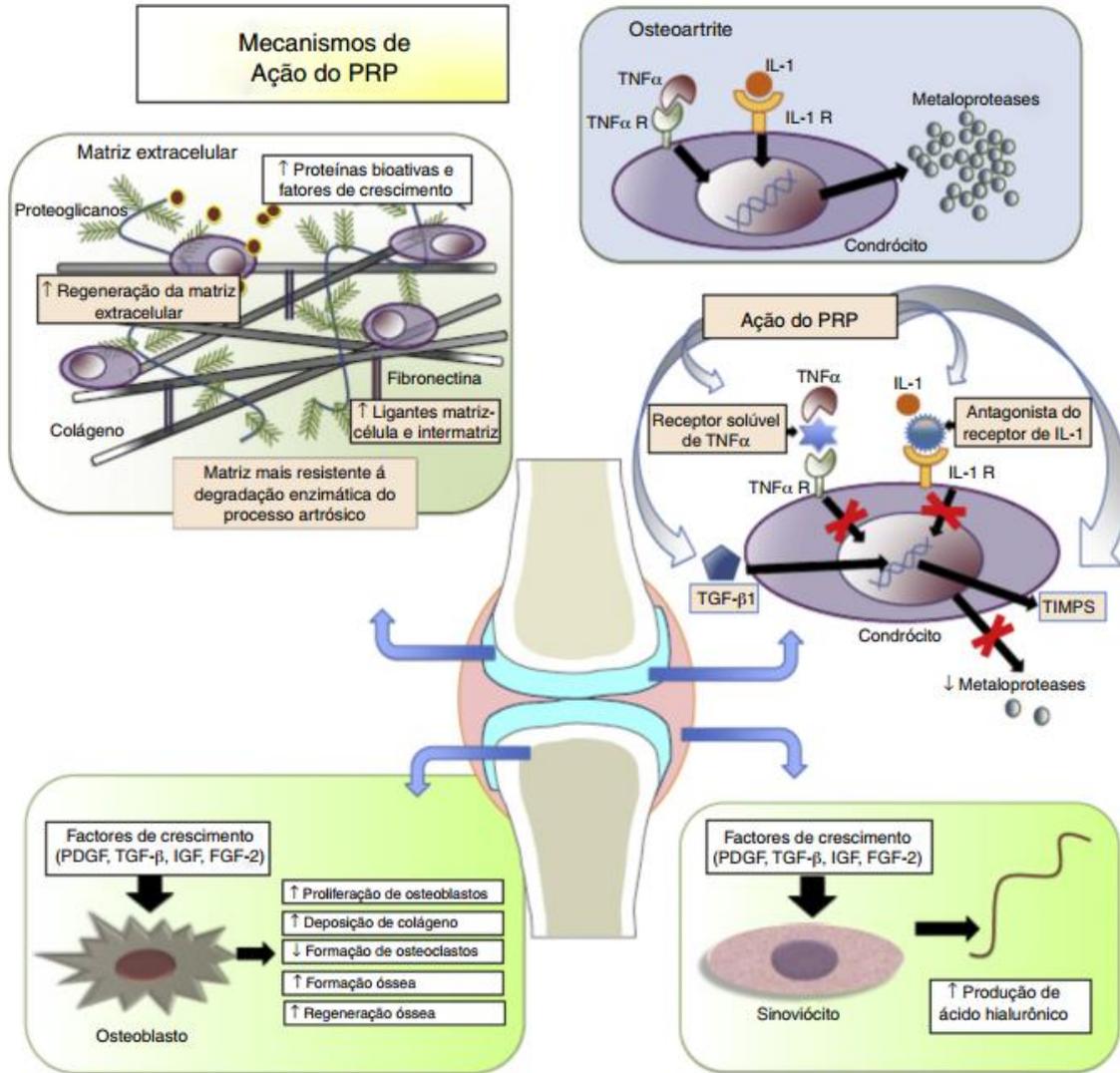
3
Estímulo ROS
Apoptose do condrócito
Fatores catabólicos e degenerativos



Artigo de revisão

Plasma rico em plaquetas no tratamento da osteoartrite

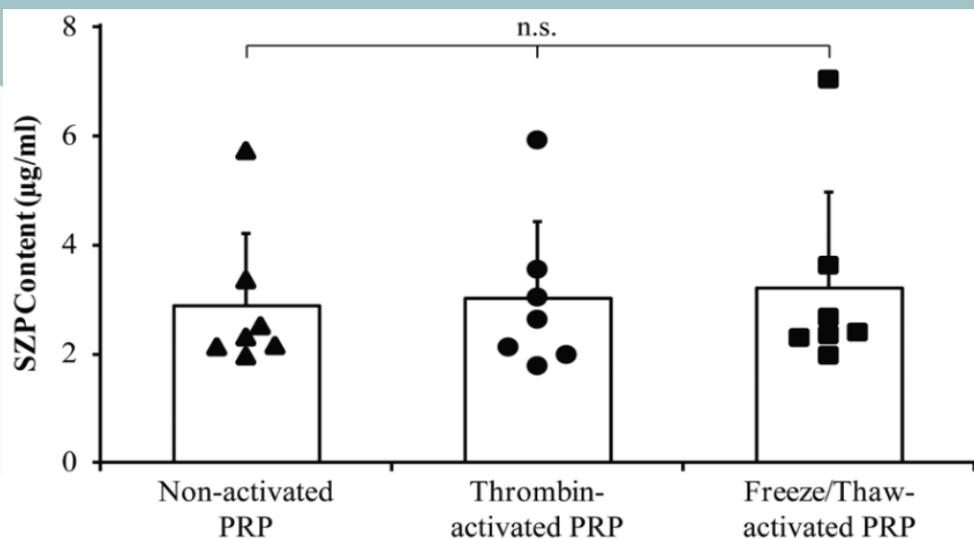
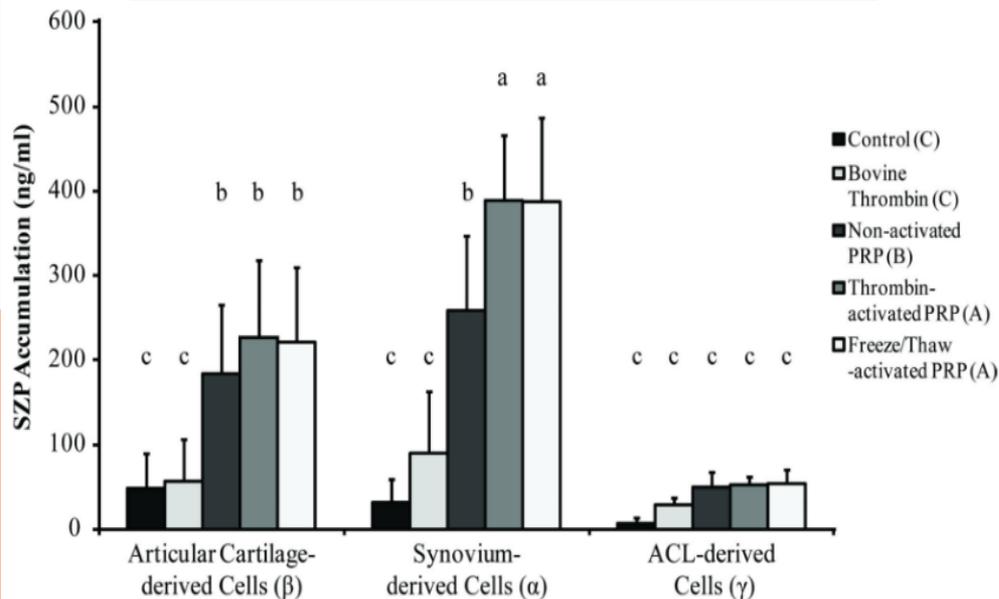
Eduardo Knop*, Luiz Eduardo de Paula e Ricardo Fuller



Stimulation of the Superficial Zone Protein and Lubrication in the Articular Cartilage by Human Platelet-Rich Plasma

Ryosuke Sakata, MD, Sean M. McNary, PhD, Kazumasa Miyatake, MD, Cassandra A. Lee, MD, James M. Van den Bogaerde, MD, Richard A. Marder, MD, and A. Hari Reddi, PhD

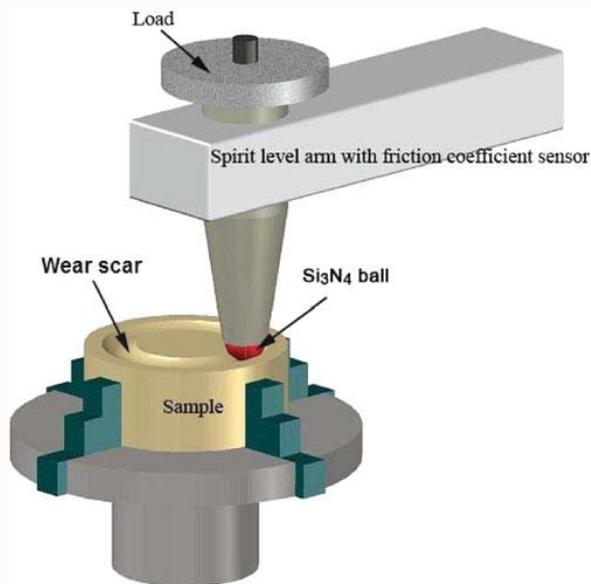
Concentração lubrificina meios de cultivo



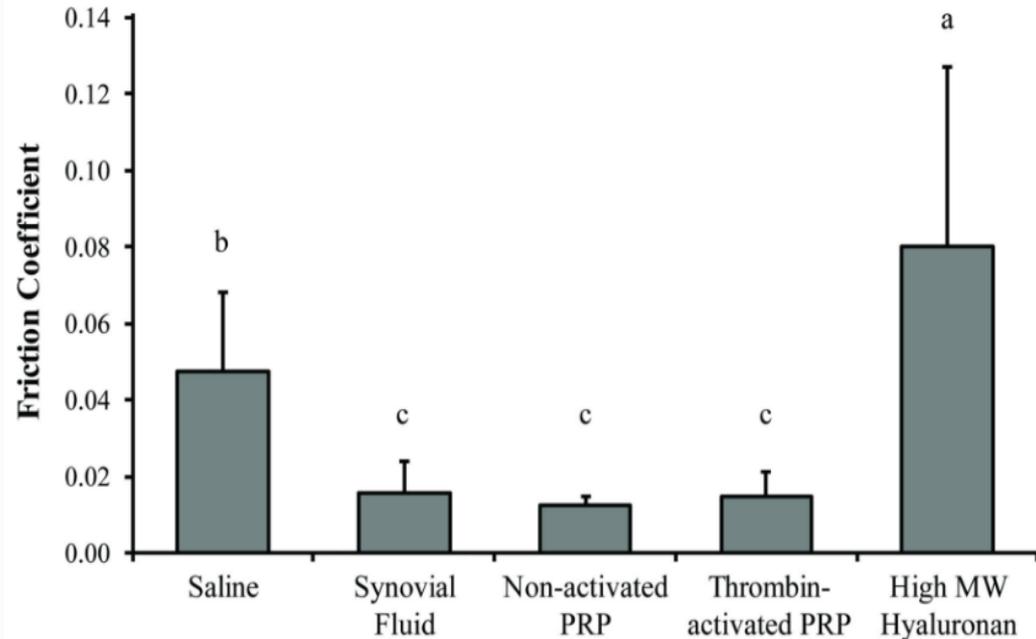
Concentração lubrificina PRP

Stimulation of the Superficial Zone Protein and Lubrication in the Articular Cartilage by Human Platelet-Rich Plasma

Ryosuke Sakata, MD, Sean M. McNary, PhD, Kazumasa Miyatake, MD, Cassandra A. Lee, MD, James M. Van den Bogaerde, MD, Richard A. Marder, MD, and A. Hari Reddi, PhD



Coeficiente de fricção cartilagem bovina



Onde os ortobiológicos irão agir?

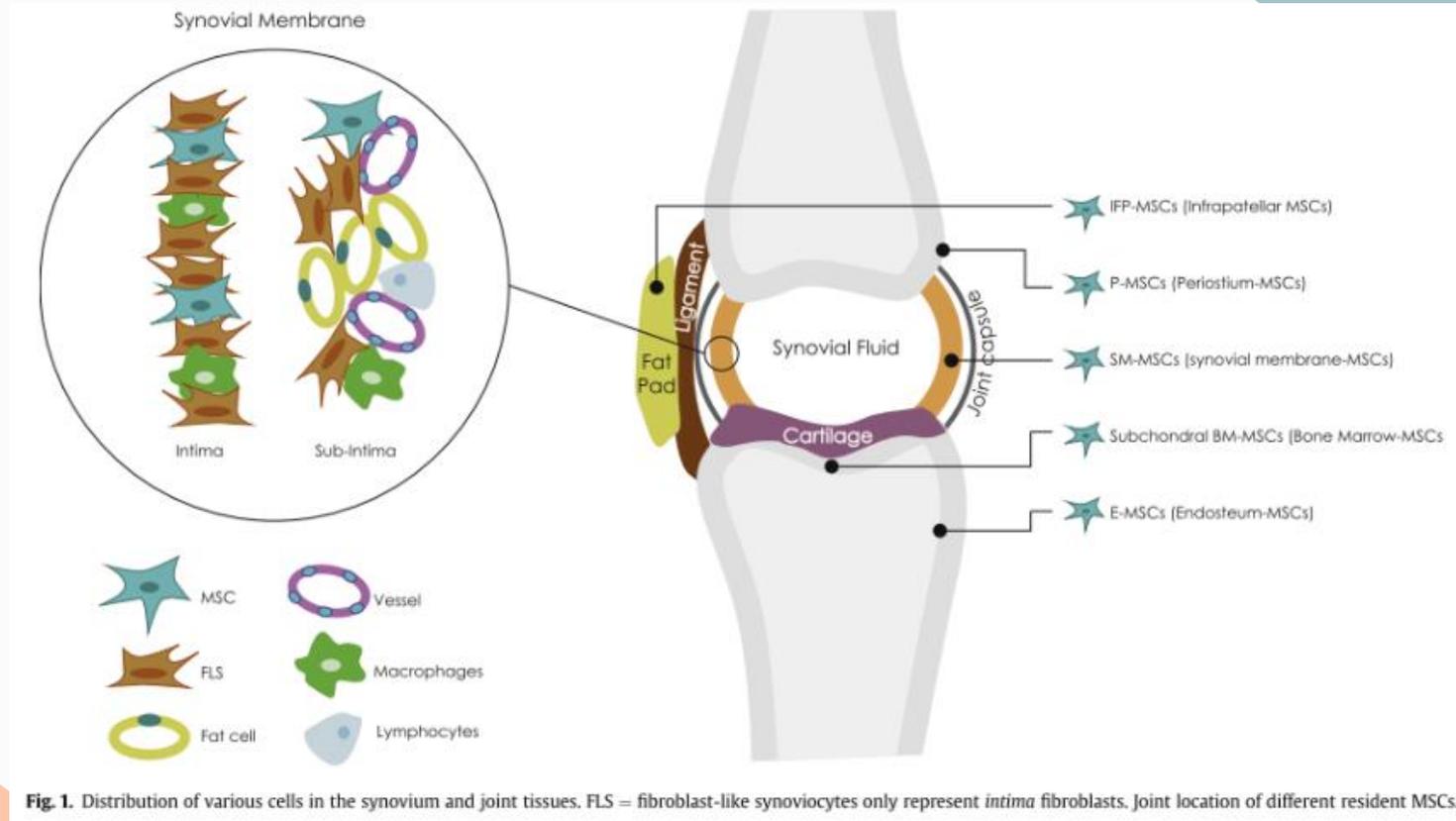
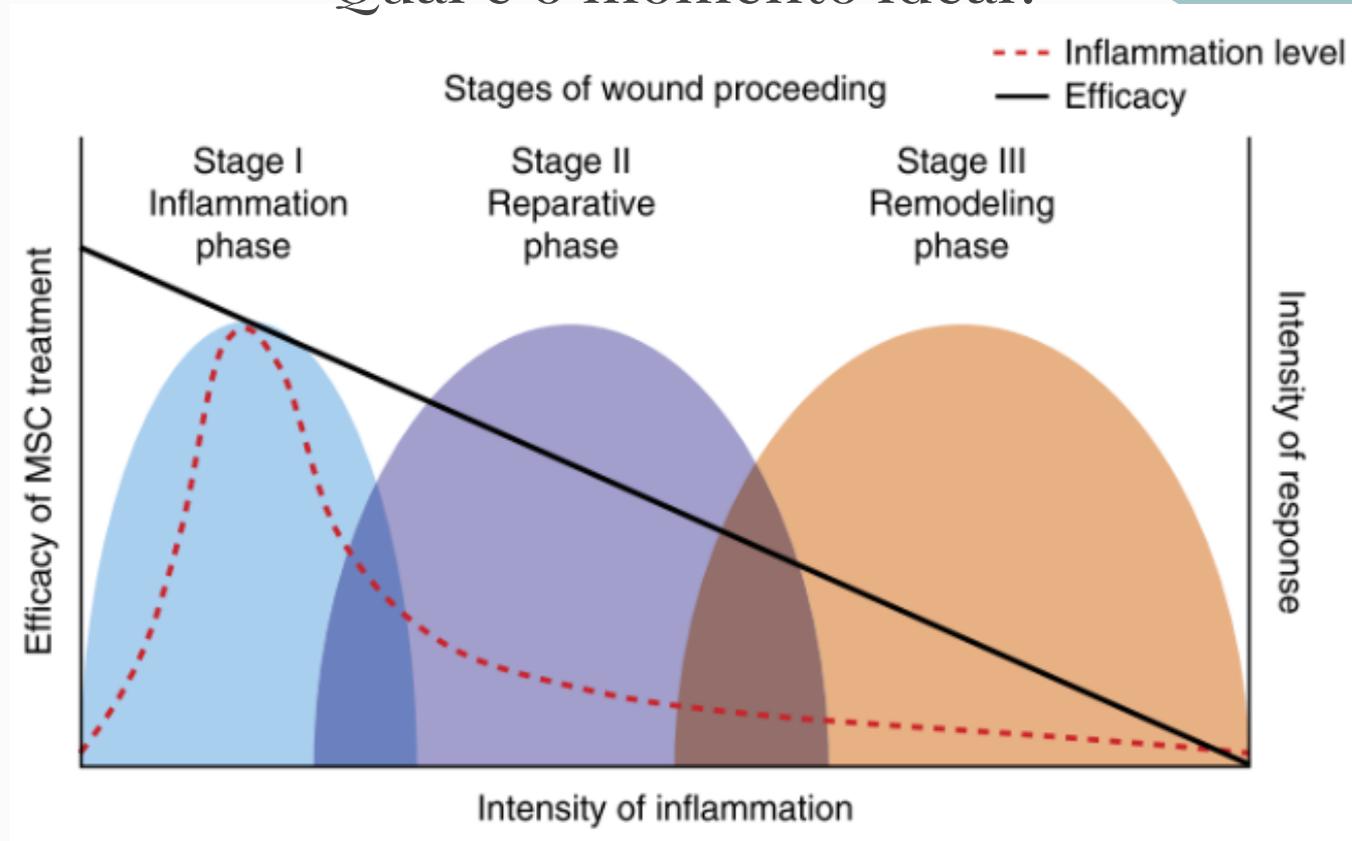


Fig. 1. Distribution of various cells in the synovium and joint tissues. FLS = fibroblast-like synoviocytes only represent *intima* fibroblasts. Joint location of different resident MSCs.

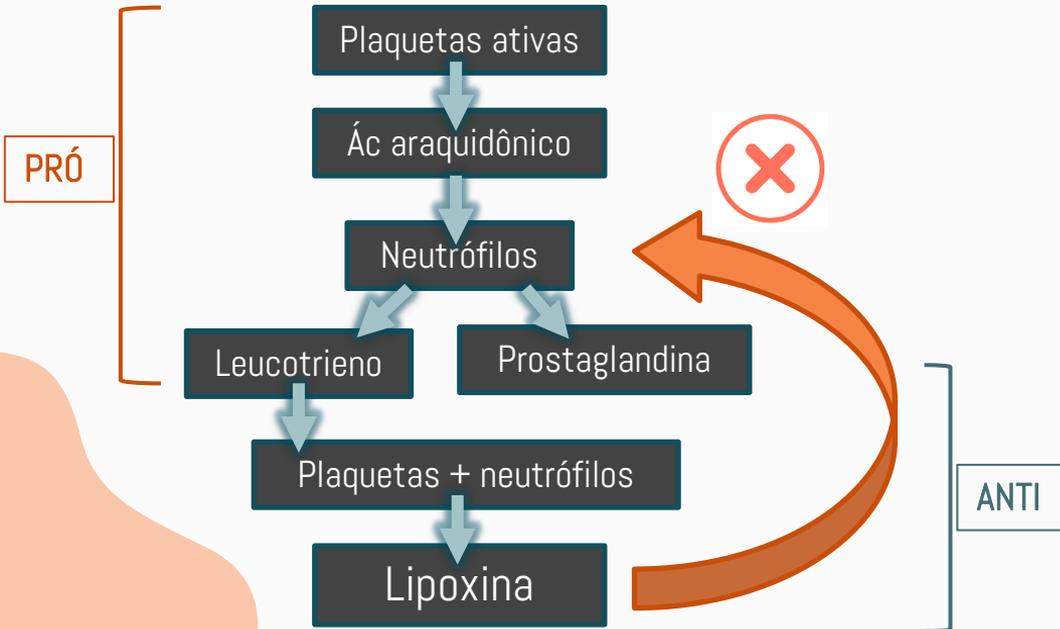
Qual é o momento ideal?





Leukocyte-rich PRP for knee osteoarthritis: Current concepts

José Fábio Lana ^a, Alex Macedo ^a, Icaro Lanzoni Gallo Ingrao ^a, Stephany Cares Huber ^b, Gabriel Silva Santos ^{a,*}, Maria Helena Andrade Santana ^b



Leucócitos

Fase inflamatória

Liberam moléculas pró e anti-inflamatórias
+ plaquetas ativas: maior potencial terapêutico

LR-PRP

Propriedade antimicrobiana

Maior produção de VEGF

Neutrófilos

Secreção de citocinas inflamatórias e MMPs

Lipoxina em OA

Manutenção do microambiente

Previne progressão

Inflamação prolongada e degeneração



Literatura vigente

O que podemos extrair dos artigos?



Pooled Platelet-Rich Plasma Lysate Therapy Increases Synoviocyte Proliferation and Hyaluronic Acid Production While Protecting Chondrocytes From Synoviocyte-Derived Inflammatory Mediators

Jessica M. Gilbertie^{1,2}, Julie M. Long¹, Alicia G. Schubert¹, Alix K. Berglund^{1,2}, Thomas P. Schaer³ and Lauren V. Schnabel^{1,2*}



Cultured Synoviocytes

Allowed to adhere and grow for 48hr

24hr stimulation with:

IL-1 β (10ng/mL)
LPS (100ng/mL)
Unstimulated control

Synoviocytes washed and treated for 48hr with:

25% PPP-L
25% PRP-L
No treatment control

Synoviocyte cell numbers assessed via density

Cultured Chondrocytes



Allowed to adhere and grow for 48hr

Added (50%) to chondrocyte media for 48hr

Synoviocyte conditioned media collected

ELISAs:
HA
IL-1 β
IL-6
TNF- α

qPCR:

COL1A1 PRG4
COL2A1 MMP-3
COL3A1 MMP-13
ACAN 18S

Obtenção do PRP

6 animais: machos e fêmeas

Repouso por 30 min e coleta do plasma total

250 G/5 min + 1.500 G/15 min

Sobrenadante (PPP) e pellet ressuspendido (PRP)

5 ciclos congelamento/descongelamento

Centrifugação 20.000 G/20 min

Amostras filtradas e congeladas

Caracterização do PRP

PPP

0.05×10^3 leucócitos/ μ L

9.38×10^3 plaquetas/ μ L

PRP

1.39 leucócitos/ μ L

1.226,38 plaquetas / μ L

Basal

5.60×10^3 leucócitos/ μ L

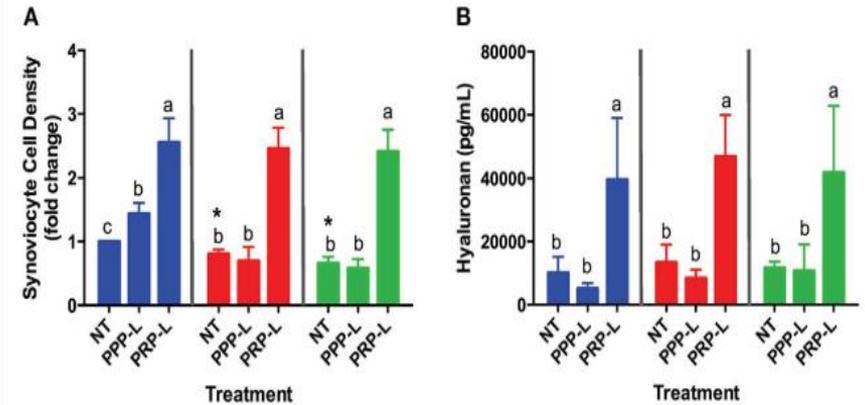
134.5×10^3 plaquetas/ μ L



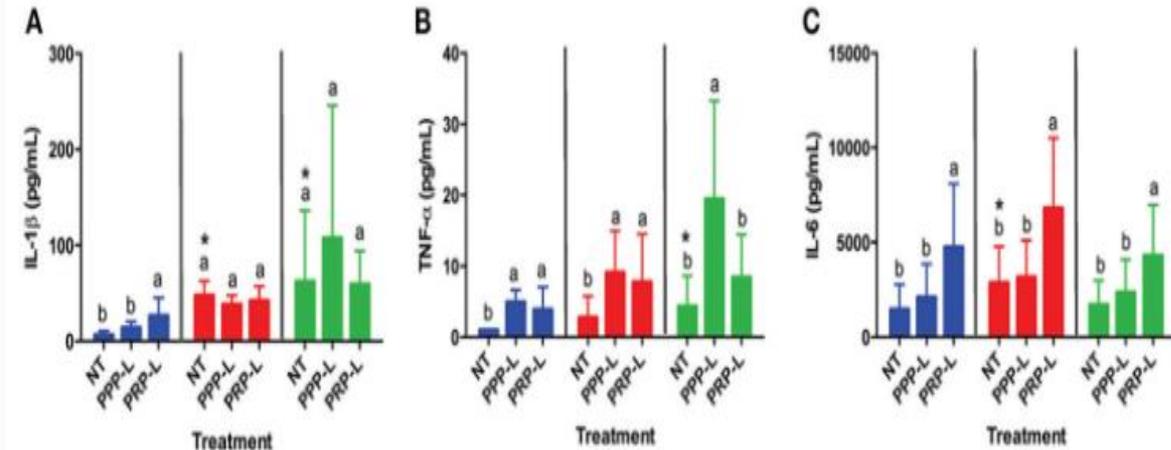
Pooled Platelet-Rich Plasma Lysate Therapy Increases Synoviocyte Proliferation and Hyaluronic Acid Production While Protecting Chondrocytes From Synoviocyte-Derived Inflammatory Mediators

Jessica M. Gilbertie^{1,2}, Julie M. Long¹, Alicia G. Schubert¹, Alix K. Berglund^{1,2}, Thomas P. Schaefer³ and Lauren V. Schnabel^{1,2*}

Crescimento de sinoviócitos e produção de HA



Mensuração de IL-1 β , TNF α e IL-6



Meios da cultura de sinoviócitos

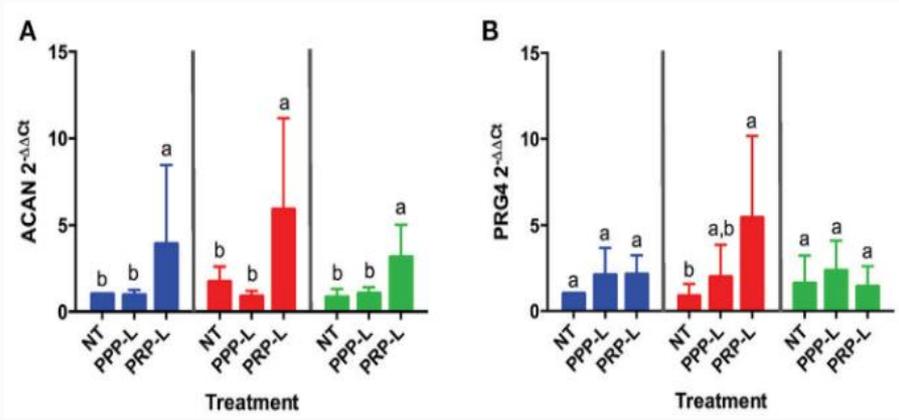




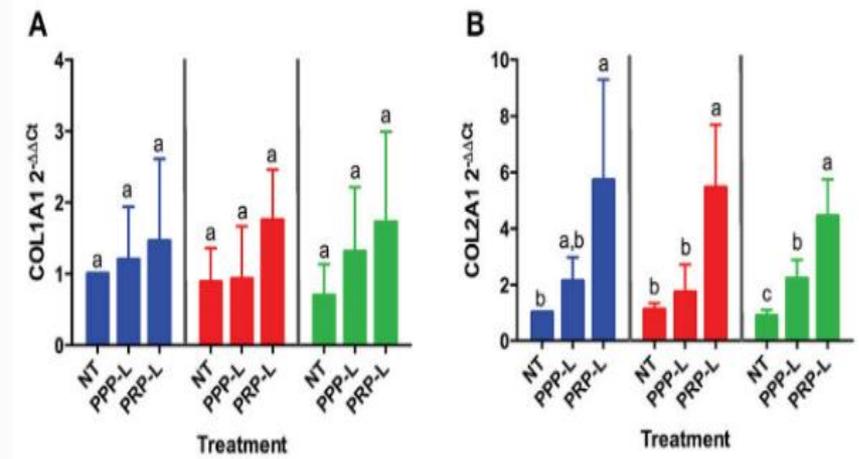
Pooled Platelet-Rich Plasma Lysate Therapy Increases Synoviocyte Proliferation and Hyaluronic Acid Production While Protecting Chondrocytes From Synoviocyte-Derived Inflammatory Mediators

Jessica M. Gilbertie^{1,2}, Julie M. Long¹, Alicia G. Schubert¹, Alix K. Berglund^{1,2}, Thomas P. Schaer³ and Lauren V. Schnabel^{1,2*}

Mensuração de agrecan e lubricina

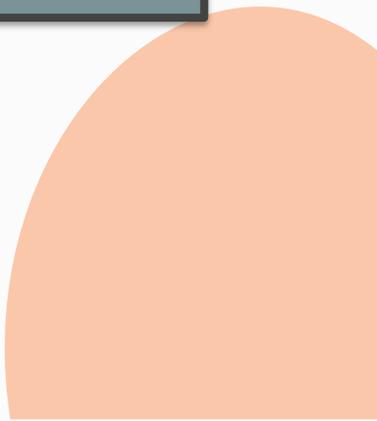


Mensuração de colágeno



Avaliação condrócitos

■ Unstimulated
■ IL-1β
■ LPS



Effects of blood-derived products and sodium hyaluronate on equine synovial fluid cells and on synovial fluid from osteochondrotic joints of horses after arthroscopy and administration of treatment

Thaís S. L. Machado DVM, PhD

Cristina O. Massoco DVM, PhD

Luis Cláudio L. C. Silva DVM, PhD

Joice Fülber DVM, MS

Juliana J. Moreira DVM, MS

Raquel Y. A. Baccarin DVM, PhD

AJVR • Vol 80 • No. 7 • July 2019

Received April 30, 2018.

Accepted November 19, 2018.

Comparou efeito de 3 hemocomponentes e HA

PRP x IRAP x APP x HA

In vitro

Células de LS de articulação sadia

Hemocomponentes homólogos

In vivo

Animais com OCD tibiotársica

Hemocomponentes autólogos

2 ml IA

Preparo hemocomponentes

PRP

150 G/5 min + 800 G/10 min

Descarte 75% e homogeneiza 1h

APP

Tubos com heparina

Incuba a 37°C/24h

300 G/10 min + 900 G/10 min

Filtra

IRAP

Incuba a 37°C/24h

3200 G/20 min

Coleta soro e filtra

Effects of blood-derived products and sodium hyaluronate on equine synovial fluid cells and on synovial fluid from osteochondrotic joints of horses after arthroscopy and administration of treatment

Thais S. L. Machado DVM, PhD

Cristina O. Massoco DVM, PhD

Luis Cláudio L. C. Silva DVM, PhD

Joice Fülber DVM, MS

Juliana J. Moreira DVM, MS

Raquel Y. A. Baccarin DVM, PhD

AJVR • Vol 80 • No. 7 • July 2019

IN VITRO

Received April 30, 2018.
Accepted November 19, 2018.

Table 1—Median (interquartile [25th to 75th percentile] range) concentrations of PGE₂, IL-1β, TNF-α, IL-10, and IL-1ra in supernatant of equine synovial cell cultures (n = 8 samples) after challenge exposure with LPS for 3 hours and incubation with various treatments for 24 hours.

| Treatment | PGE ₂ (pg/mL) | IL-1β (pg/mL) | TNF-α (pg/mL) | IL-10 (pg/mL) | IL-1ra (pg/mL) |
|--------------------|---|-----------------------|------------------|--------------------|--|
| Control | 4,332.5 ^a (3,683.0–5,805.4) | 0 (0–0) | 0 (0–0) | 0 (0–0.5) | 0 ^a (0–7.3) |
| Phenylbutazone | 153.1 ^b (90.2–917.3) | 0 (0–0) | 0 (0–0) | 0 (0–28.2) | 0 ^a (0–26.2) |
| PRP | 7,481.1 ^c (6,390.5–9,249.2) | 0 (0–0) | 0 (0–0) | 0 (0–2.3) | 1,141.2 ^b (584.7–1,397.4) |
| IRAP | 6,499.5 ^{a,c,d} (0–8,803.6) | 135.2 (34.4–168.5) | 0 (0–0) | 19.2 (0–48.1) | 9,603.1 ^c (6,364.7–10,163.4) |
| APP | 4,476.5 ^{a,d} (0–7,100.9) | 180.3 (31.4–239.1) | 0 (0–0) | 18.9 (7.7–52.9) | 20,996.1 ^d (19,333.0–25,816.0) |
| Sodium hyaluronate | 5,050.3 ^{a,c,d} (4,777.0–7,147.3) | 0 (0–0) | 0 (0–0) | 0 (0–12.4) | 0 ^a (0–18.8) |

Effects of blood-derived products and sodium hyaluronate on equine synovial fluid cells and on synovial fluid from osteochondrotic joints of horses after arthroscopy and administration of treatment

Thais S. L. Machado DVM, PhD
 Cristina O. Massoco DVM, PhD
 Luis Cláudio L. C. Silva DVM, PhD

AJVR • Vol 80 • No. 7 • July 2019

IN VIVO

Joice Fülber DVM, MS
 Juliana J. Moreira DVM, M
 Raquel Y. A. Baccarin D

Received April 30, 2018.
 Accepted November 19, 2018.

Table 3—Median (interquartile [25th to 75th percentile] range) concentrations of PGE₂, IL-1β, and IL-1ra and least squares mean (95% confidence interval) concentrations of TNF-α and IL-10 in synovial fluid of osteochondrotic tibiotarsal joints of horses (8 joints/treatment) before (time 0) and 48 hours after arthroscopy and injection of various treatments.

| Treatment | PGE ₂ (pg/mL) | | IL-1β (pg/mL) | | IL-1ra (pg/mL) | | TNF-α (pg/mL) | | IL-10 (pg/mL) | |
|--------------------|---------------------------------------|-------------------------|---------------------|---------------------|----------------|----------------------------|------------------------|------------------------|--------------------------|---------------------------|
| | 0 hours | 48 hours | 0 hours | 48 hours | 0 hours | 48 hours | 0 hours | 48 hours | 0 hours | 48 hours |
| Control | 33.9 ^a (23.9 to 52.1) | 19.8* (15.5 to 29.9) | 2.0 (1.5 to 2.2) | 2.1 (1.6 to 2.5) | 0 (0 to 0) | 1,721* (536 to 3,266) | 1.14 (0.34 to 1.94) | 1.52 (0.33 to 2.70) | 23.9 (-52.7 to 100.0) | 33.2 (-54.4 to 120.0) |
| PRP | 15.8 ^a (11.1 to 32.7) | 30.3* (19.8 to 33.1) | 3.1 (1.7 to 3.6) | 2.1 (1.8 to 3.6) | 0 (0 to 0) | 3,730* (802 to 5,965) | 1.59 (1.3 to 1.9) | 1.67 (0.84 to 2.5) | 47.7 (8.3 to 87.2) | 52.4 (-53.4 to 158.0) |
| IRAP | 70.9 ^b (39.8 to 77.5) | 29.1* (21.3 to 37.7) | 2.5 (1.9 to 2.5) | 2.9 (2.6 to 3.2) | 0 (0 to 0) | 3,229* (1,779 to 5,126) | 1.83 (1.13 to 2.53) | 1.53 (0.84 to 2.48) | 33.1 (-29.3 to 95.6) | 30.6 (-17.4 to 78.3) |
| APP | 13.8 ^a (12.5 to 19.0) | 18.4 (16.4 to 43.1) | 2.7 (2.2 to 4.1) | 2.6 (2.4 to 3.5) | 0 (0 to 0) | 1,827* (292 to 4,487) | 1.74 (0.44 to 3.04) | 1.89 (1.46 to 2.33) | 23.1 (-12.3 to 58.6) | 77.1 (-119.0 to 273.0) |
| Sodium hyaluronate | 32.4 ^{a,b} (21.2 to 37.7) | 27.5 (16.4 to 38.6) | 3.2 (2.5 to 4.1) | 3.6 (1.7 to 5.9) | 0 (0 to 0) | 9,520* (247 to 12,627) | 1.55 (0.78 to 2.31) | 2.5 (-0.23 to 5.22) | 17.4 (-23.5 to 58.4) | 29.2 (-39.4 to 97.9) |

Effects of blood-derived products and sodium hyaluronate on equine synovial fluid cells and on synovial fluid from osteochondrotic joints of horses after arthroscopy and administration of treatment

Thais S. L. Machado DVM, PhD

Cristina O. Massoco DVM, PhD

Luis Cláudio L. C. Silva DVM, PhD

Joice Fülber DVM, MS

Juliana J. Moreira DVM, MS

Raquel Y. A. Baccarin DVM, PhD

AJVR • Vol 80 • No. 7 • July 2019

Received April 30, 2018.
Accepted November 19, 2018.

IN VIVO

Table 4—Median (interquartile [25th to 75th percentile] range) concentrations of chondroitin sulfate and hyaluronic acid and percentage of high-molecular-weight (HMW) hyaluronic acid in synovial fluid of osteochondrotic tibiotarsal joints of horses (8 joints/treatment) before and 48 hours after arthroscopy and injection of various treatments.

| Treatment | Chondroitin sulfate ($\mu\text{g/mL}$) | | Hyaluronic acid ($\mu\text{g/mL}$) | | HMW hyaluronic acid (%) | |
|--------------------|---|-------------------------|---|---------------------------------------|----------------------------|----------------------|
| | 0 hours | 48 hours | 0 hours | 48 hours | 0 hours | 48 hours |
| Control | 75.4 (40.6–136.7) | 195.8* (88.6–356.4) | 676.0 (419.0–744.9) | 802.7 ^{a,b} (326.9–877.3) | 74.0 (62.2–87.0) | 45.5* (14.7–61.2) |
| PRP | 38.0 (23.9–58.2) | 68.9* (61.2–164.4) | 377.6 (267.6–630.7) | 390.4 ^a (241.8–583.9) | 80.0 (75.0–89.0) | 54.0* (35.0–82.0) |
| IRAP | 40.5 (34.1–66.8) | 109.4* (74.0–165.1) | 288.2 (193.0–426.2) | 206.5 ^c (174.5–247.9) | 77.0 (63.0–81.0) | 57.7 (39.5–63.0) |
| APP | 65.5 (50.7–103.2) | 181.0* (127.4–382.6) | 541.6 (309.2–609.4) | 406.2 ^{a,b} (239.9–605.4) | 79.0 (38.5–90.3) | 43* (29.3–60.3) |
| Sodium hyaluronate | 47.3 (19.6–53.1) | 159.9* (135.0–180.7) | 492.2 (476.4–577.5) | 584.1 ^b (526.7–664.5) | 83.5 (62.7–91.5) | 51.5* (24.7–71.5) |

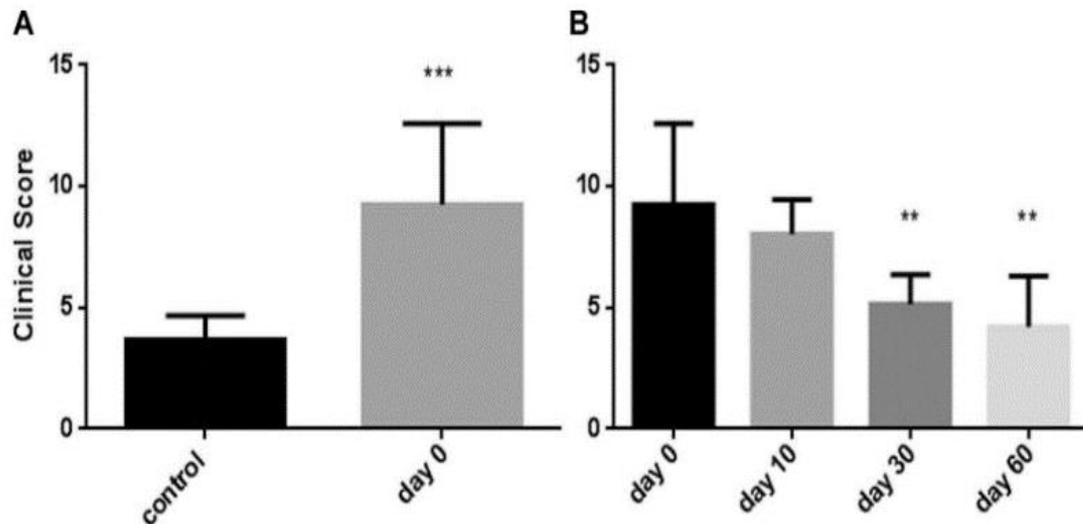


Original Research

Treatment With Platelet Lysate Inhibits Proteases of Synovial Fluid in Equines With Osteoarthritis

Gustavo Perrone^a, Yael Lastra^b, Camilo González^a, Nicolás Caggiano^b, Rubén Giménez^a, Román Pareja^b, Emilio De Simone^{b,*}

Escore clínico animais



A: controle x tratados, dia 0
B: tratados, dia 0 e pós tratamento

PL em OA

23 cavalos esporte (T) + 21 sadios (C)

PRP

1000 G/10 min + 1500 G/10 min

1 ml pellet

3 ciclos congelamento/descongelamento

Contagem plaquetária

400-700 × 10³ plaquetas/μL

1 ml IA → única

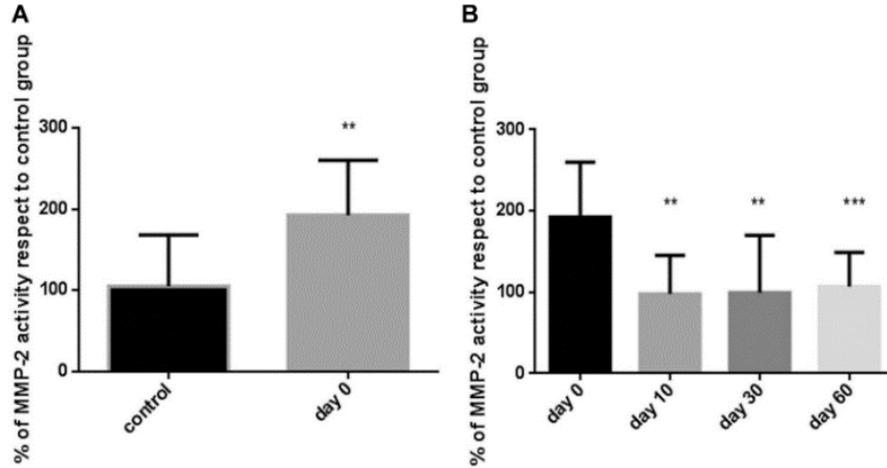


Original Research

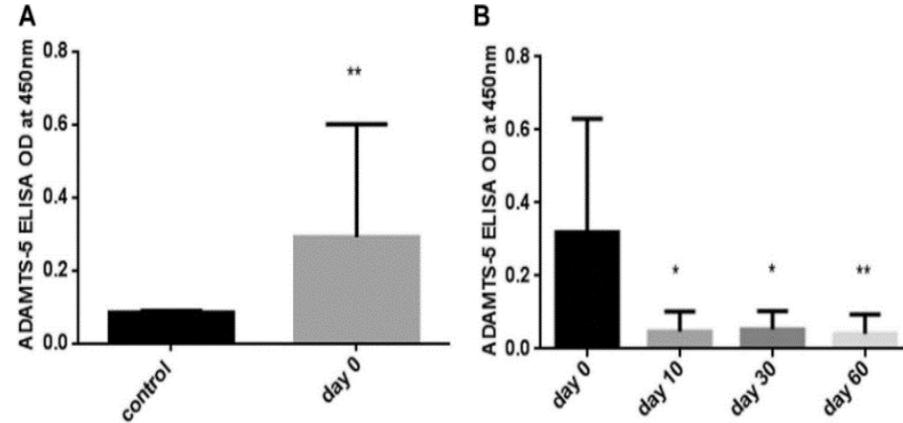
Treatment With Platelet Lysate Inhibits Proteases of Synovial Fluid in Equines With Osteoarthritis

Gustavo Perrone ^a, Yael Lastra ^b, Camilo González ^a, Nicolás Caggiano ^b, Rubén Giménez ^a, Román Pareja ^b, Emilio De Simone ^{b, *}

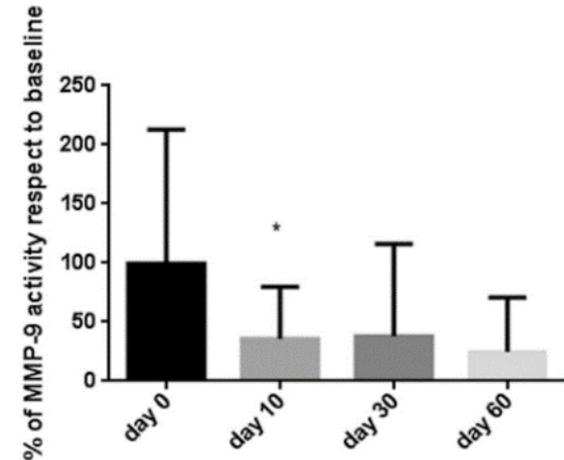
Atividade de MMP-2



Mensuração ADAMTS



Atividade de MMP-9



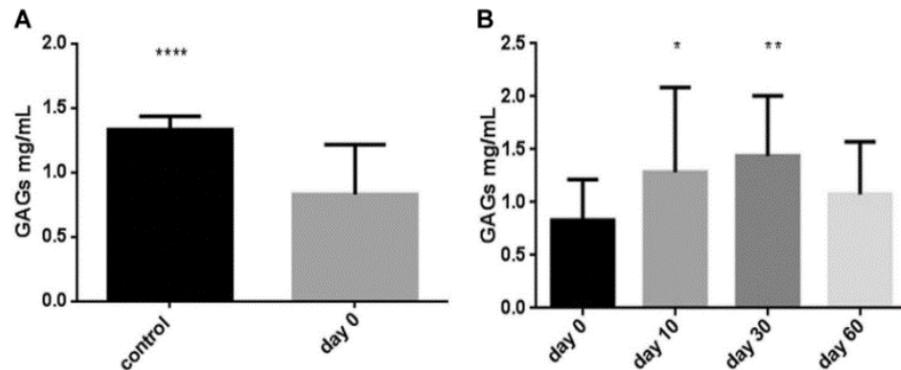
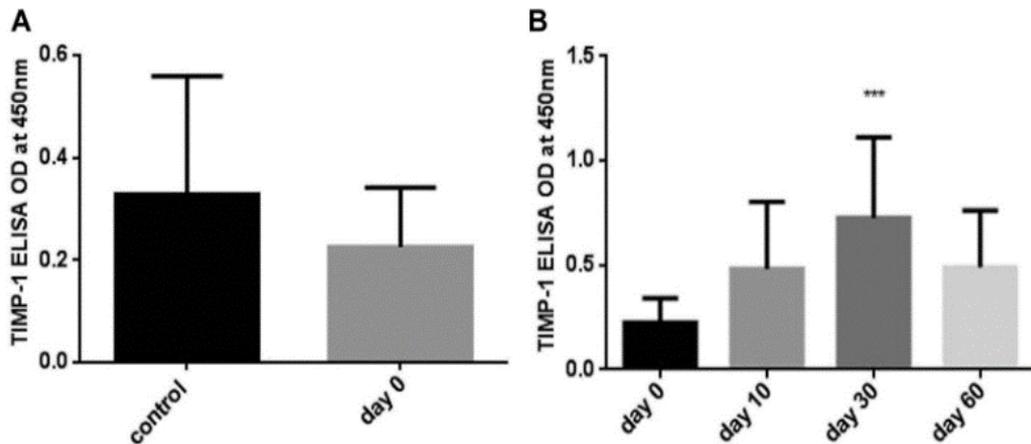


Original Research

Treatment With Platelet Lysate Inhibits Proteases of Synovial Fluid in Equines With Osteoarthritis

Gustavo Perrone^a, Yael Lastra^b, Camilo González^a, Nicolás Caggiano^b, Rubén Giménez^a, Román Pareja^b, Emilio De Simone^{b,*}

Mensuração TIMP-1



Mensuração GAGs

Considerações dos autores

PL alivia dor e sinais clínicos em OA

Sem reações adversas

↓ MMP-9

Inibição de recrutamento de leucócitos,

Ação de TGF-B1 e IGF

↑ TIMP

TGF-B1

↑ GAGs

Deterioração articular

Clinical findings, synovial fluid cytology and growth factor concentrations after intra-articular use of a platelet-rich product in horses with osteoarthritis



PRP IA: sadio e OA

5 sadios x 5 OA

PRP

Sistema de filtração

Concentração

Plaquetária 4.7x

Leucocitária 2.1x

Avaliação clínica

| Group | Control | OA |
|--------------------------------------|---------------|---------------|
| OA scale (0–3) | 0 ± 0 | 1.80 ± 0.83 |
| Synovial effusion score (0–3) | | |
| Day 0 | 0.00 ± 0.00** | 1.00 ± 1.22** |
| Day 1 | 1.00 ± 0.00* | 1.40 ± 0.89 |
| Day 2 | 1.00 ± 0.00* | 1.60 ± 0.54 |
| Day 5 | 0.60 ± 0.54 | 0.80 ± 0.83 |
| Day 21 | 0.60 ± 0.54 | 1.00 ± 0.70 |
| Day 56 | 0.00 ± 0.00 | 0.40 ± 0.89 |
| Flexion score (0–3) | | |
| Day 0 | 0.00 ± 0.00 | 0.20 ± 0.44 |
| Day 1 | 0.40 ± 0.54 | 0.40 ± 0.54 |
| Day 2 | 0.00 ± 0.00 | 0.20 ± 0.44 |
| Day 5 | 0.00 ± 0.00 | 0.20 ± 0.44 |
| Day 21 | 0.00 ± 0.00 | 0.00 ± 0.00 |
| Day 56 | 0.00 ± 0.00 | 0.00 ± 0.00 |
| Periarticular signs (yes/no) | | |
| Day 0 | 0/5 | 1/4 |
| Day 1 | 5/0 | 3/2 |
| Day 2 | 5/0 | 5/0 |
| Day 5 | 3/2 | 2/3 |
| Day 21 | 3/2 | 2/3 |
| Day 56 | 0/5 | 0/5 |

Clinical findings, synovial fluid cytology and growth factor concentrations after intra-articular use of a platelet-rich product in horses with osteoarthritis



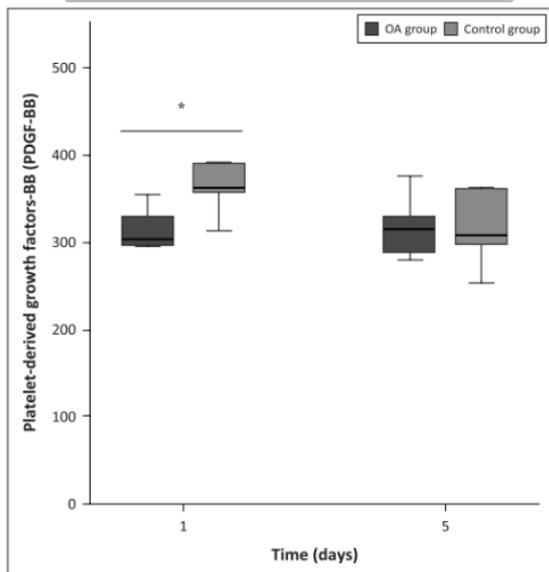
Análise do líquido sinovial

| Day | NCC (cells/ μ L) | Neutrophils (%) | TP (g/L) | SAA (mg/L) | PDGF-BB (pg/mL) | TGF- β 1 (pg/mL) |
|---------------|---|------------------------------------|--------------------------------|-----------------|--------------------------------------|---|
| Day 0 | | | | | | |
| Control | 111.60 \pm 39.89 | 1.60 \pm 2.45 | 15.48 \pm 6.31 | 0.00 \pm 0.00 | - | - |
| OA | 87.40 \pm 53.64 | 5.40 \pm 8.50 | 14.22 \pm 2.56 | 0.00 \pm 0.00 | - | - |
| Day 1 | | | | | | |
| Control | 22940.00 \pm 11103.28 ^{*,****} | 83.40 \pm 2.70 ^{*,****} | 46.64 \pm 11.11 [*] | 0.00 \pm 0.00 | 390.47 \pm 82.15 ^{**,***} | 4021.27 \pm 783.00 ^{**,***,****} |
| OA | 20464.00 \pm 12055.93 ^{*,****} | 71.20 \pm 5.40 ^{*,****} | 39.06 \pm 7.10 [*] | 0.00 \pm 0.00 | 302.27 \pm 47.44 ^{**} | 2792.48 \pm 615.46 ^{**,***,****} |
| Day 2 | | | | | | |
| Control | 13212.00 \pm 3413.46 [*] | 64.00 \pm 4.00 ^{*,**} | 30.18 \pm 11.55 [*] | 0.00 \pm 0.00 | - | - |
| OA | 7372.00 \pm 6122.10 | 38.80 \pm 14.32 ^{*,**} | 22.46 \pm 5.90 [*] | 0.00 \pm 0.00 | - | - |
| Day 5 | | | | | | |
| Control | 3654.00 \pm 2037.59 ^{***} | 14.80 \pm 11.86 ^{****} | 20.56 \pm 8.02 | 0.95 \pm 2.14 | 316.42 \pm 45.69 ^{***} | 851.73 \pm 351.82 ^{***,****} |
| OA | 1660.40 \pm 1250.72 ^{***} | 8.20 \pm 15.54 ^{****} | 16.54 \pm 6.09 | 0.00 \pm 0.00 | 318.06 \pm 37.82 | 678.35 \pm 289.58 ^{***,****} |
| Day 21 | | | | | | |
| Control | 207.60 \pm 49.90 | 2.60 \pm 1.67 | 17.14 \pm 6.61 | 0.00 \pm 0.00 | - | - |
| OA | 213.00 \pm 123.80 | 5.20 \pm 6.97 | 12.18 \pm 3.18 | 0.00 \pm 0.00 | - | - |
| Day 56 | | | | | | |
| Control | 224.80 \pm 71.94 | 10.80 \pm 11.16 | 14.62 \pm 4.51 | 0.00 \pm 0.00 | - | - |
| OA | 95.60 \pm 71.40 | 4.20 \pm 6.79 | 13.78 \pm 3.26 | 0.00 \pm 0.00 | - | - |

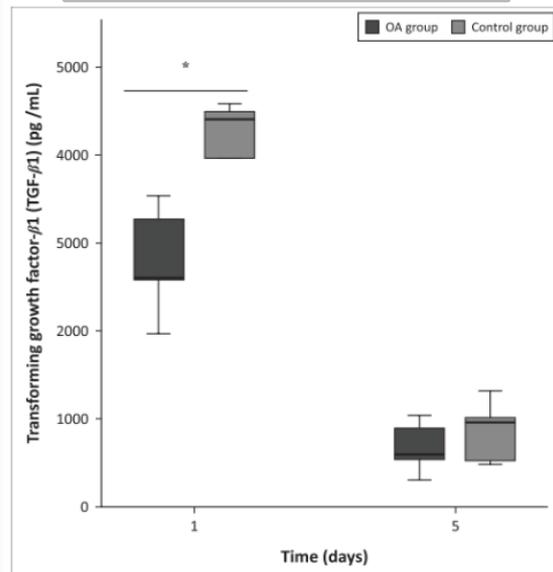
Clinical findings, synovial fluid cytology and growth factor concentrations after intra-articular use of a platelet-rich product in horses with osteoarthritis



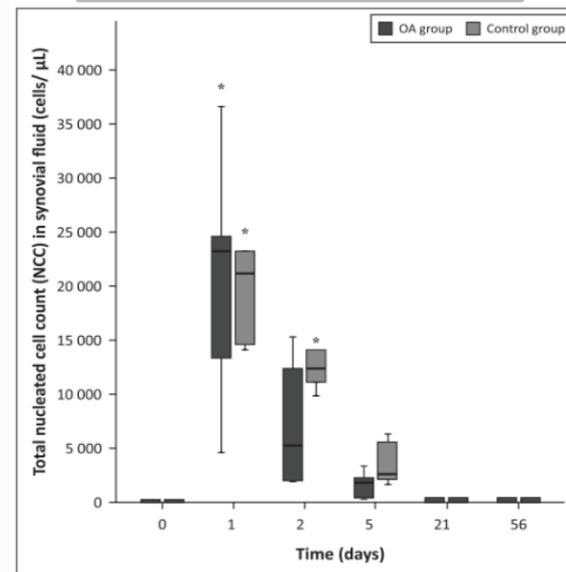
Mensuração PDGF-BB



Mensuração TGF-β1



Mensuração NCC



RESEARCH

Open Access



A safety evaluation of allogeneic freeze-dried platelet-rich plasma or conditioned serum compared to autologous frozen products equivalents in equine healthy joints

Livia Camargo Garbin^{1,2}, Erin K. Contino³, Christine S. Olver⁴ and David D. Frisbie^{3*}

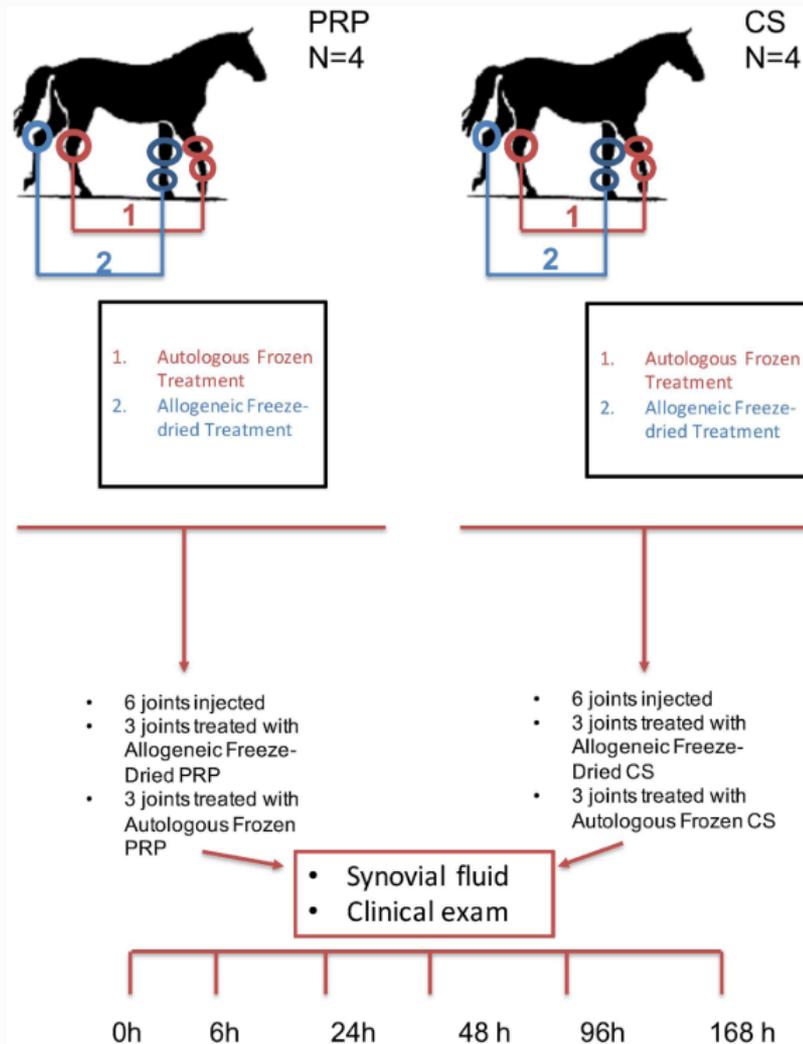
Segurança hemocomponentes liofilizados PRP/SCA

Autólogo: congelamento

Alógeno: secagem de fração dos autólogos

Aplicação: 4 doadores/fração + RL

Animal: próprio controle



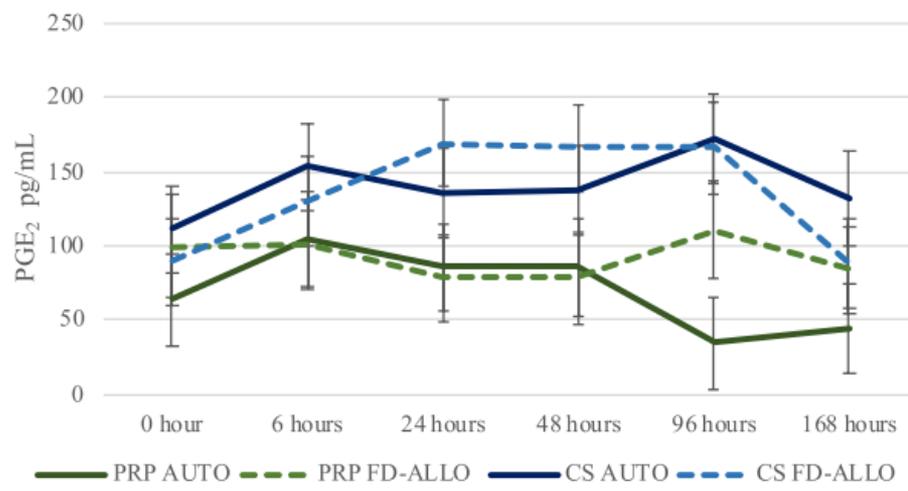
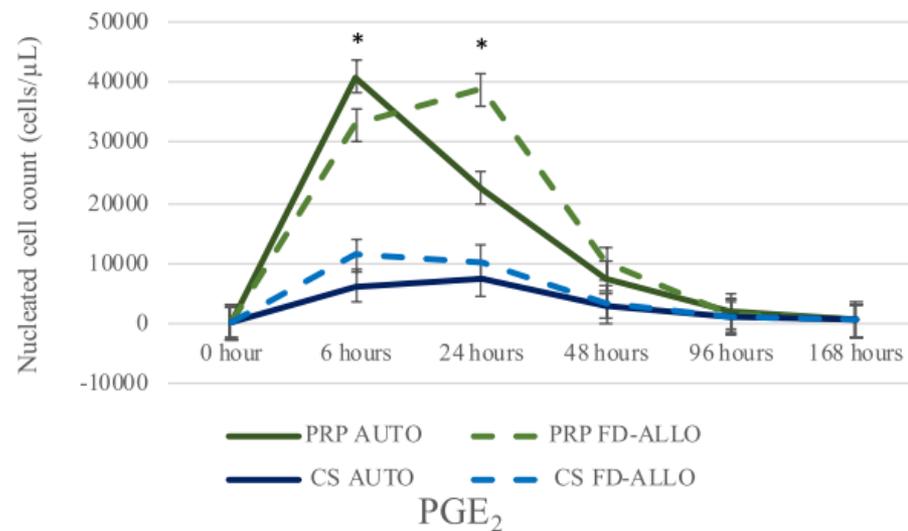
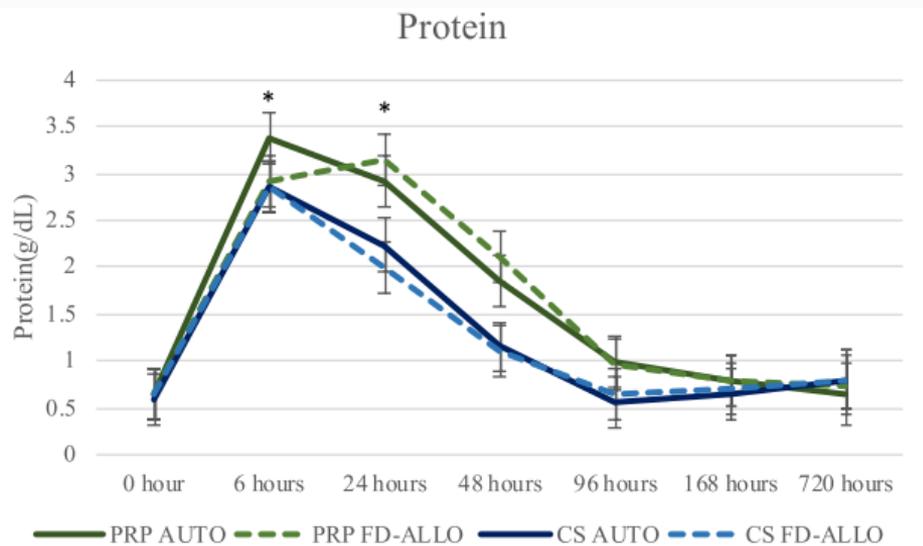
RESEARCH

Open Access



A safety evaluation of allogeneic freeze-dried platelet-rich plasma or conditioned serum compared to autologous frozen products equivalents in equine healthy joints

Livia Camargo Garbin^{1,2}, Erin K. Contino³, Christine S. Olver⁴ and David D. Frisbie^{3*}



A Platelet-Rich Plasma-Derived Biologic Clears *Staphylococcus aureus* Biofilms While Mitigating Cartilage Degeneration and Joint Inflammation in a Clinically Relevant Large Animal Infectious Arthritis Model

Jessica M. Gilbertie^{1,2}, Thomas P. Schaefer^{3†}, Julie B. Engiles^{3,4}, Gabriela S. Seiler⁵, Bennett L. Deddens⁵, Alicia G. Schubert¹, Megan E. Jacob^{2,6}, Darko Stefanovski³, Gordon Ruthel⁴, Noreen J. Hickok⁷, Devorah M. Stowe⁶, Alexa Frink¹ and Lauren V. Schnabel^{1,2†}

Indução:

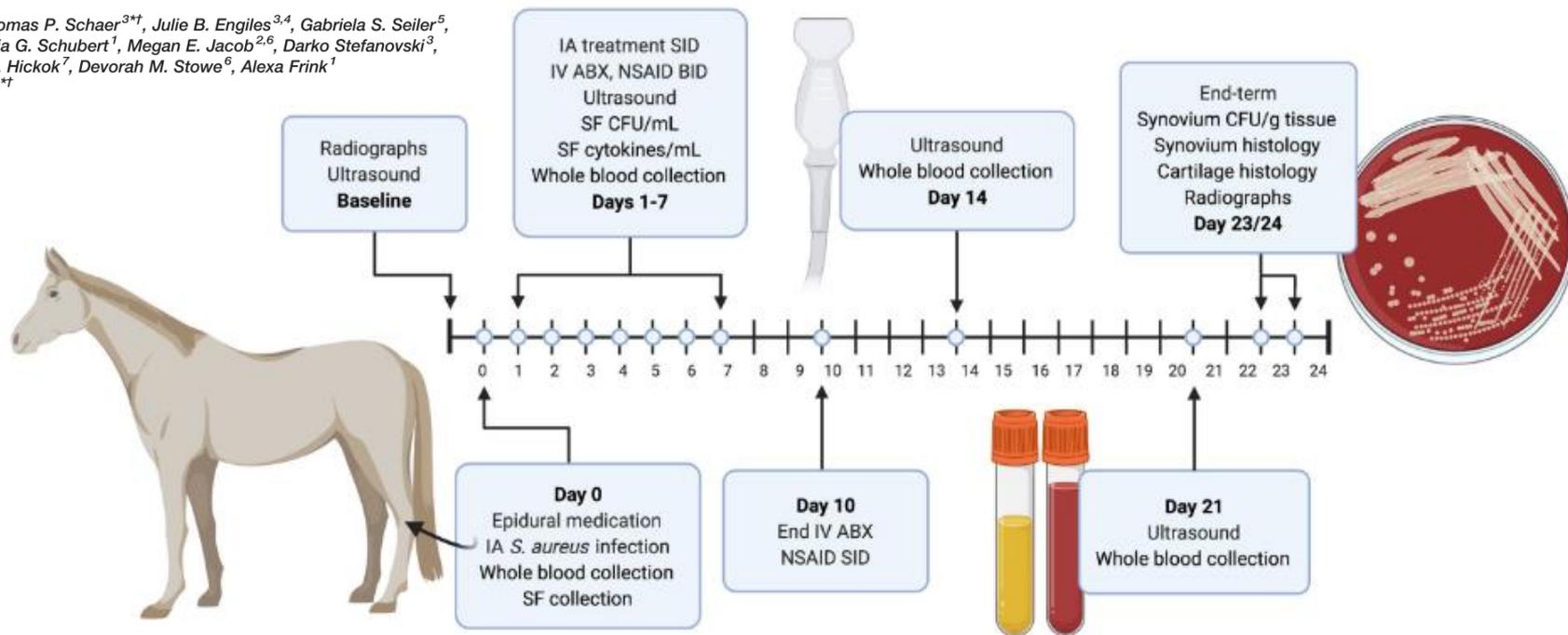
Staphylococcus aureus 1x10⁶ CFU

Tratamento:

24h pós-indução

500mg amicacina + 5mL salina

500mg amicacina + 5mL BIO-PLY

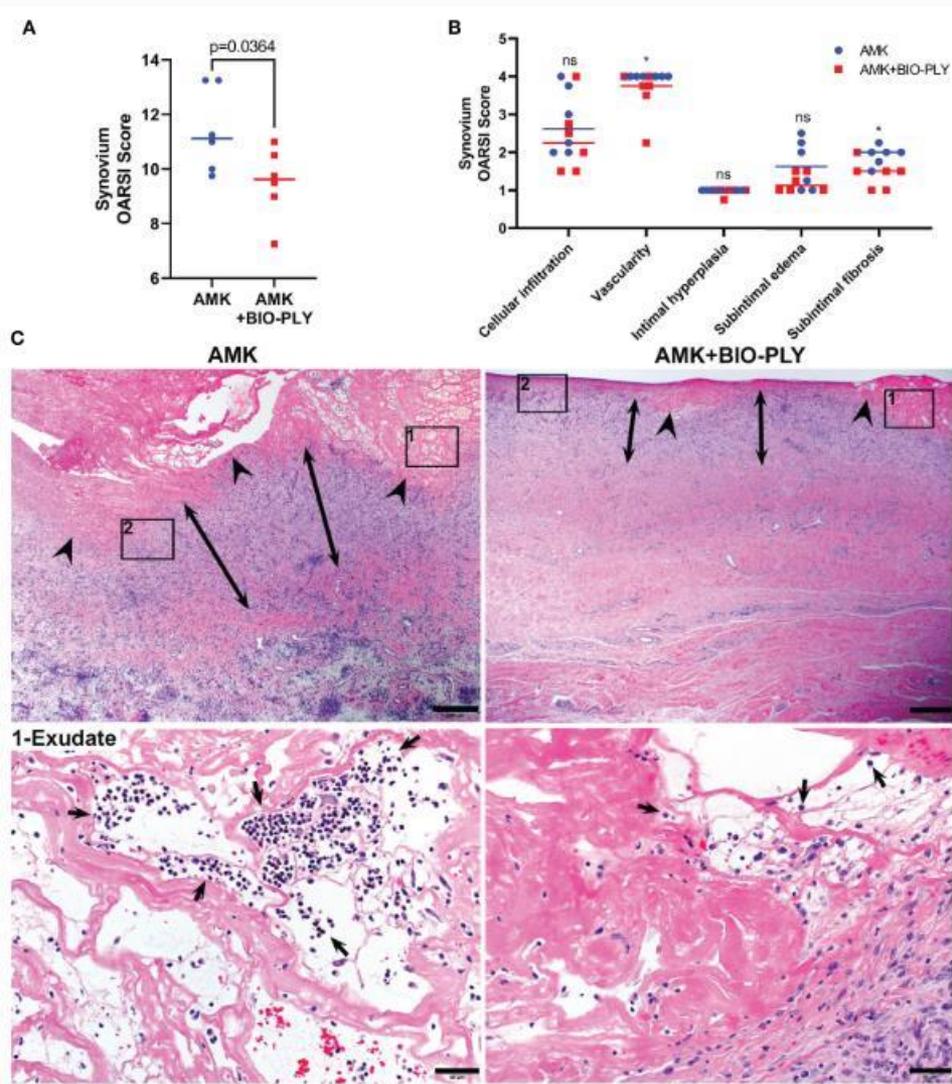


A Platelet-Rich Plasma-Derived Biologic Clears *Staphylococcus aureus* Biofilms While Mitigating Cartilage Degeneration and Joint Inflammation in a Clinically Relevant Large Animal Infectious Arthritis Model

Jessica M. Gilbertie^{1,2}, Thomas P. Schaefer^{3*†}, Julie B. Engiles^{3,4}, Gabriela S. Seiler⁵, Bennett L. Deddens⁵, Alicia G. Schubert¹, Megan E. Jacob^{2,6}, Darko Stefanovski³, Gordon Ruthel⁴, Noreen J. Hickok⁷, Devorah M. Stowe⁶, Alexa Frink¹ and Lauren V. Schnabel^{1,2*†}

Bio-PLY + ampicillin

Reduziu perda de proteoglicano cartilágineo associado à infecção





The effect of platelet rich plasma combined with microfractures on the treatment of chondral defects: an experimental study in a sheep model

G. Milano †, E. Sanna Passino †, L. Deriu †, G. Careddu †, L. Manunta †, A. Manunta †, M.F. Saccomanno †, C. Fabbriciani †

PRP + microfraturas

15 ovelhas

Lesão condral 8 mm

Tratamento 12 m pós indução

4 perfurações (MF)

1.5 mm x 5 mm

Eutanásia 6 m pós tto

Grupos

G1: microfraturas (MF)

G2: MF + PRP gel + cola

G3: MF + PRP líquido

PRP

2.400 rpm/3 min

3.000 rpm/12 min

Gel

6 ml PRP

2 ml gluconato de Cálcio 10%

2 ml cola de fibrina

Concentração

$4 \times 1.415 \times 10^3$ plaquetas/ μ L

Avaliação macro

A (G1)

Tecido de reparação delgado

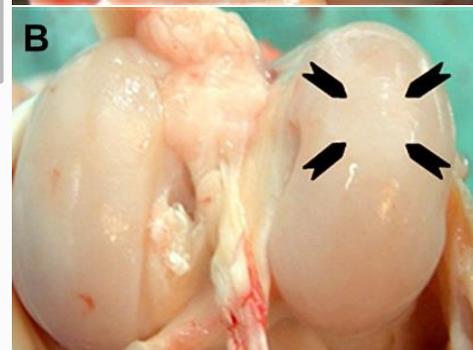
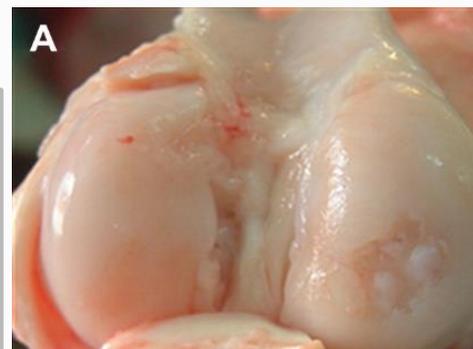
B (G2)

Similar à cartilagem sadia

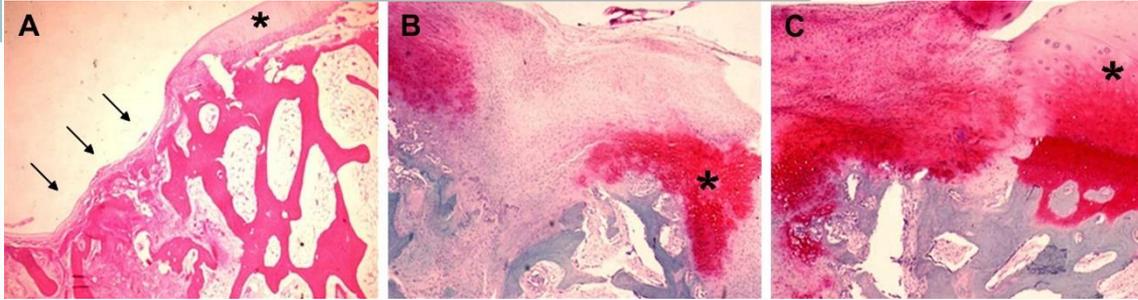
C (G3)

Cobre o defeito

Delgado e irregular

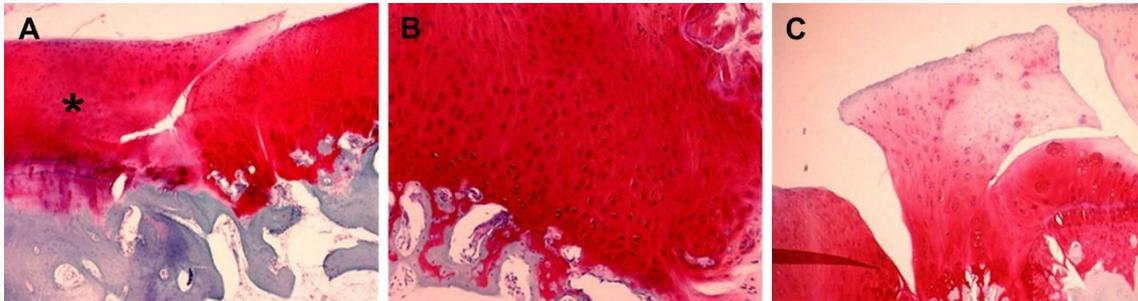


Avaliação histológica



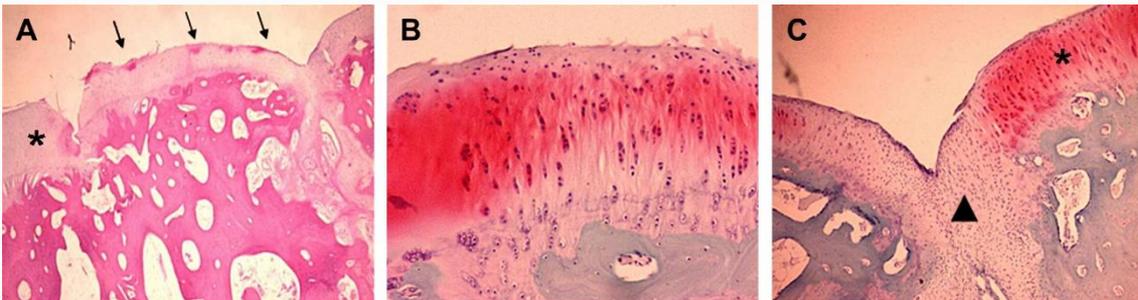
G1

A: tecido de reparo delgado
 B: MF com células arredondadas, MEC fibrosa e mal organizada
 C: tecido de reparo parcialmente integrado ao sadio



G2

A: espessura semelhante à sadio (*), zona de transição
 B: poucas células arredondadas, intensa coloração Safranina
 C: aglomerados de células tipo condrócitos em zona de transição



G3

A: fina camada, cobre o defeito
 B: células tipo condrócitos organizadas em colunas
 C: tecido fibrocartilaginoso (seta), cartilagem normal com intensa proliferação celular (*)



The effect of platelet rich plasma combined with microfractures on the treatment of chondral defects: an experimental study in a sheep model

G. Milano †✉, E. Sanna Passino †, L. Deriu †, G. Careddu †, L. Manunta †, A. Manunta †, M.F. Saccomanno †, C. Fabbriciani †

Considerações dos autores

Potencial eficácia no reparo de defeito condral

TGF-β1, BMP-2, FGF

Sem inflamação ou reação imune

Não houve formação de tecido hialino

Pouca Safranina

Atividade celular limitada

MEC de baixa qualidade

PRP

Gel + cola de fibrina

Melhores resultados

Não comparou efeitos isolados

Propriedades histológicas,
macroscópicas e mecânicas:

MF < PRP líquido < PRP gel

Treatment of canine osteoarthritis with allogeneic platelet-rich plasma: review of five cases

José Catarino,¹ Pedro Carvalho,^{2,3} Sara Santos,¹ Ângela Martins,^{1,4} and João Requicha^{1,5,*}

| Animal | Breed | Age (years) | Joint |
|--------|--------------------|-------------|-------------|
| 1 | Mixed breed | 12 | Knee |
| 2 | Labrador Retriever | 10 | Tibiotarsal |
| 3 | Labrador Retriever | 13 | Knee |
| 4 | German Shepherd | 14 | Elbow |
| 5 | German Pointer | 6 | Carpal |

OA

Diagnóstico imagem

Refratários a tratamentos

Medicamentosos

Fisioterápicos

6 meses sem tratamento prévio

PRP

Alogênico

Dupla centrifugação

Sem detalhes do protocolo

1×10^6 plaquetas/ μL

Treatment of canine osteoarthritis with allogeneic platelet-rich plasma: review of five cases

José Catarino,¹ Pedro Carvalho,^{2,3} Sara Santos,¹ Ângela Martins,^{1,4} and João Requicha^{1,5,*}

| Animal | Breed | Age (years) | Joint |
|--------|--------------------|-------------|-------------|
| 1 | Mixed breed | 12 | Knee |
| 2 | Labrador Retriever | 10 | Tibiotarsal |
| 3 | Labrador Retriever | 13 | Knee |
| 4 | German Shepherd | 14 | Elbow |
| 5 | German Pointer | 6 | Carpal |

Grau de claudicação

| Animal | Day 0 | Day 30 | Day 60 | Day 90 |
|--------|-----------|-----------|----------|-------------|
| 1 | Grade III | Grade II | Grade II | Grade I |
| 2 | Grade II | Grade II | Grade I | No lameness |
| 3 | Grade IV | Grade III | Grade II | Grade II |
| 4 | Grade III | Grade III | Grade II | Grade II |
| 5 | Grade II | Grade II | Grade I | No lameness |

Escore de dor

| Animal | Day 0 | Day 30 | Day 60 | Day 90 |
|--------|----------|----------|--------|----------|
| 1 | Intense | Moderate | Mild | Painless |
| 2 | Moderate | Mild | Mild | Painless |
| 3 | Intense | Moderate | Mild | Painless |
| 4 | Severe | Intense | Mild | Mild |
| 5 | Moderate | Mild | Mild | Mild |

The background features abstract, organic shapes in shades of orange and teal. A large orange shape is at the top left, and a teal shape is at the bottom right. A white space separates them, containing the text. A small orange oval is positioned at the bottom center, overlapping the teal shape.

Evidências
científicas

The Role of Platelet-Rich Plasma in Cartilage Pathology: An Updated Systematic Review of the Basic Science Evidence

Michael P. Fice, B.A., J. Chance Miller, B.A., Robert Christian, M.D., Charles P. Hannon, M.D., Niall Smyth, M.D., Christopher D. Murawski, B.S., Brian J. Cole, M.D., M.B.A., and John G. Kennedy, M.D., F.R.C.S.



Critérios de inclusão

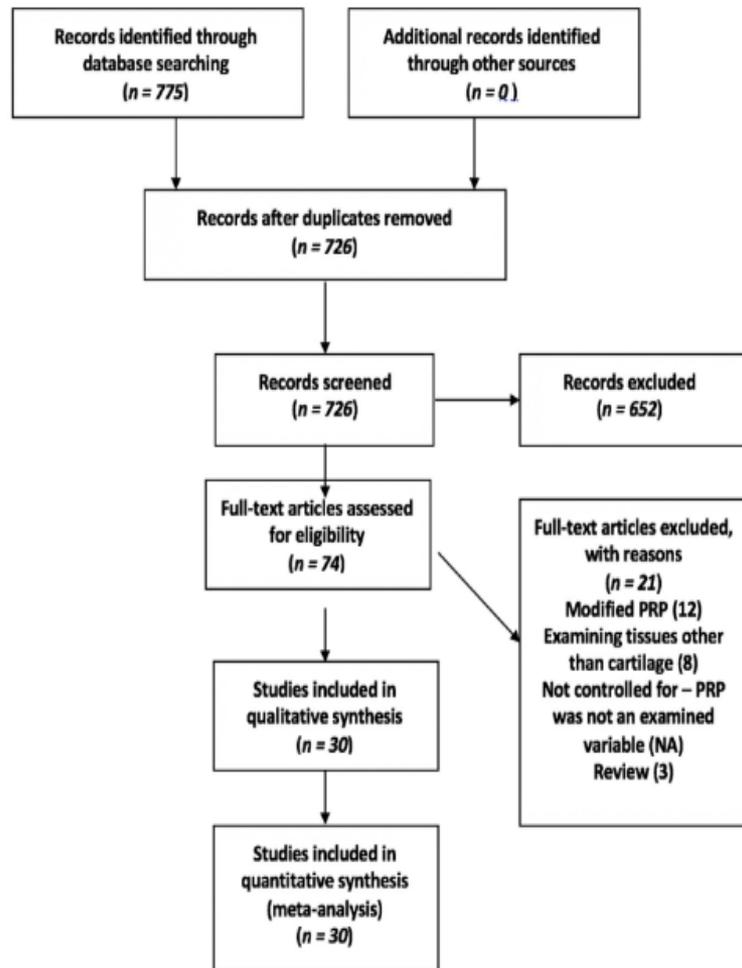
PRP em condrócitos e cartilagem
Excetuou discos intervertebrais e meniscos
PRP sozinho, sem reagente ou material
Revisão por pares e em inglês
Grupo controle

Identification

Screening

Eligibility

Included



The Role of Platelet-Rich Plasma in Cartilage Pathology: An Updated Systematic Review of the Basic Science Evidence

Michael P. Fice, B.A., J. Chance Miller, B.A., Robert Christian, M.D., Charles P. Hannon, M.D., Niall Smyth, M.D., Christopher D. Murawski, B.S., Brian J. Cole, M.D., M.B.A., and John G. Kennedy, M.D., F.R.C.S.



Diminuição inflamação *in vitro*
 Melhora histologia *in vivo*
 Melhora produção de MEC
 Proliferação de condrócitos
 Reduz MMP-3 e 13

Ação em processo inflamatório pouco elucidada
 Metodologia
 Parâmetros avaliados
 Padronização de técnica
 Caracterização do PRP



Table 3. Platelet-Rich Plasma Cytology Reporting in Basic Science Studies on Cartilage Repair Published Since 2011

| Component | Reported Studies, n (%) | Studies Not Reporting, n (%) |
|--|-------------------------|------------------------------|
| Platelet count | 21 (70.0) | 9 (30.0) |
| WBC count | 9 (30.0) | 21 (70.0) |
| RBC count | 2 (6.7) | 28 (93.3) |
| Platelet + WBC + RBC count | 2 (6.7) | 28 (93.3) |
| Platelet + WBC count | 9 (30.0) | 21 (70.0) |
| Platelet count without WBCs or RBCs | 12 (40.0) | 18 (60.0) |
| No reference to Platelet, WBC or RBC count | 9 (30.0) | 21 (70.0) |

RBC, red blood cell; WBC, white blood cell.

Platelet-rich plasma versus hyaluronic acid in the treatment of knee osteoarthritis

A meta-analysis

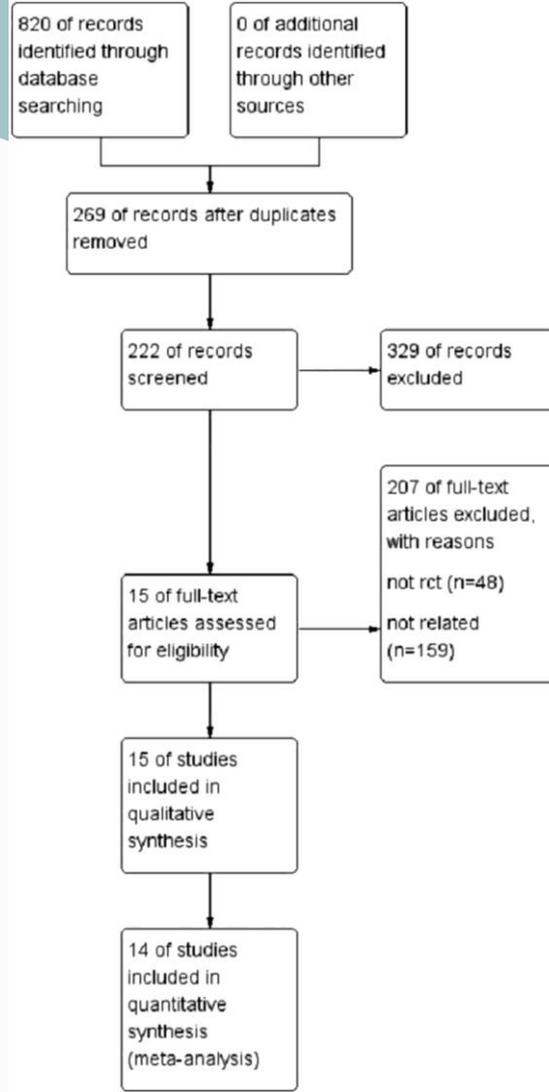
Zehan Chen, BS^a, Chang Wang, MD^a, Di You, MD^b, Shishun Zhao, PhD^a, Zhe Zhu, PhD^c, Meng Xu, MD^{d,*}

Critérios de inclusão

- OA de joelho,
- PRP (GT) HA (GC),
- RCTs,
- Citar estudos envolvendo indicadores VAS, IKDC, WOMAC, KOOS
- Citar efeitos adversos

Critérios de exclusão

- Uso de animal ou cadáver,
- Impossibilidade de extrair ou converter dados válidos,
- Estudos retrospectivos, revisões ou papers de conferência



OPEN

Platelet-rich plasma versus hyaluronic acid in the treatment of knee osteoarthritis

A meta-analysis

Zehan Chen, BS^a, Chang Wang, MD^a, Di You, MD^b, Shishun Zhao, PhD^a, Zhe Zhu, PhD^c, Meng Xu, MD^{d,*}

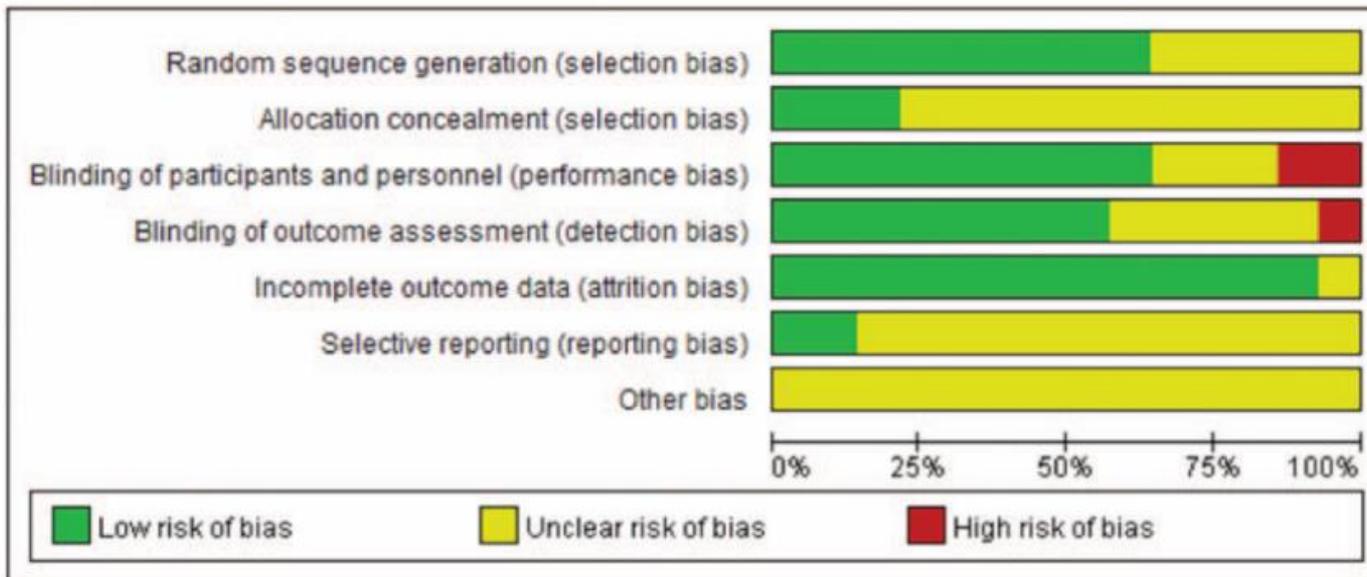
Conclusão

PRP > AH

Redução da dor

Melhora da função articular

Tratamento conservativo da OA



RESEARCH ARTICLE

Open Access

Platelet-rich plasma in orthopedic therapy: a comparative systematic review of clinical and experimental data in equine and human musculoskeletal lesions

Patrícia M Brossi, Juliana J Moreira, Thaís SL Machado and Raquel YA Baccharin*

| Preparation method | | In vivo models or clinical studies | |
|--------------------|---|--|---|
| Commercial | System used | Injection | Location (intra-articular, intra-lesional, peri-lesional, etc.) |
| | Detail any modifications to manufacturer protocol | | Volume injected |
| | Detail any manufacturer options (if any) and option selected (i.e. final volume of PRP) | | Ultrasound guidance (yes or no) |
| Manual | Volume of blood collected | | Timing of injection relative to injury or surgery |
| | Type and final concentration of anticoagulant | | Re-dosing interval |
| | Centrifugation speed in gravitational (<i>g</i>) force; (rpm are not appropriate—results in variable <i>g</i> force depending on centrifuge radius) | | Post-injection rehabilitation |
| | Centrifugation time | | Prior or concurrent treatments |
| | Number of spin cycles | Complications | Describe major and minor |
| | Final volume of PRP | | Detail number affected |
| | | | Duration post-treatment |
| | Outcome measures | As appropriate to tissue/injury of study | |

| Characterization of PRP | |
|-------------------------|--|
| Hematology | Platelet, leukocyte, and red blood cell concentration in blood and PRP Consider reporting fibrinogen concentration |
| Growth factors | Consider reporting, particularly for new protocols that have not been validated to increase growth factor concentration |
| Storage | Fresh or frozen-thawed |
| Activation | |
| Yes | Agent (i.e. calcium chloride, autologous thrombin, bovine thrombin, etc.) Agent concentration Time to clot Releasate only or entire clot used |
| No | |

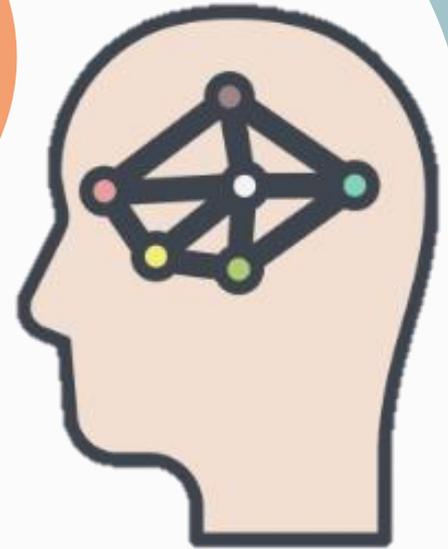
Perspectivas futuras

Explorar possibilidades hemocomponentes

- Vesículas extracelulares
- Terapias acelulares

Associação de terapias regenerativas

Otimizar efeitos



Obrigada!

Grupo de Pesquisa Aplicada em Medicina Equina

Group of Applied Research in Equine Medicine



@equiscience.usp



@equiscience_usp



@equiscience.usp



equiscience



sarahseidel@usp.br



@mv.sarahseidel

