



Evidence Based
Healthcare Group

Evidence Based Review

Efficacy of Autologous Platelet Rich Plasma in bone healing

Reviewers	Tanya Skaler: principal reviewer, literature searching Amanda Bowens: literature searching
Date review completed	June 2007

Important Note:

The purpose of this evidence based review is to summarise information on the effectiveness and safety of Autologous Platelet Rich Plasma. It is not intended to replace clinical judgement or to be used as a clinical protocol. A reasonable attempt has been made to find and appraise papers relevant to the focus of this review; however, it does not claim to be exhaustive.

The review was developed by staff of ACC's Evidence Based Healthcare Team.

However, the content does not necessarily present the official view of ACC or represent ACC policy. The review is based upon information supplied up to the end of May 2007.

EFFICACY OF AUTOLOGOUS PLATELET RICH PLASMA USE IN BONE HEALING –

EVIDENCE-BASED REVIEW

Executive summary

This review examines the evidence from the published clinical studies related to use of autologous Platelet Rich Plasma (PRP) in surgical procedures involving bone healing.

A high concentration of platelets in the platelet rich concentrate is thought to have osteo-generating properties, thus contributing to osseous healing. Platelet rich concentrate is obtained from patient's blood using commercially available devices and then mixed with thrombin, forming a gel that is either directly applied to the surgical surfaces or mixed with cancellous chips added to grafts.

Augmentation with autologous platelet rich gel is used in a variety of surgical procedures. In orthopaedic surgery it was initially intended for patients undergoing total knee arthroplasty (20). Later its use has been extended to cosmetic and plastic surgery, ophthalmology, general and ENT surgery and other surgical disciplines. This evidence based review is limited to the use of autologous platelet rich gel in orthopaedic, spinal, dental, periodontal and maxillofacial surgery.

The overall impression from the evidence based review is that the published literature appears to be lacking robust scientific evidence to either verify or refute the ability of PRP to enhance osseous healing.

A majority of the existing studies used low score non-analytic methods, such as case reports and case series, with a high risk of the results being unreliable (2-4, 7-9, 11, 14-19, 21-24, 26-31, 35-47, 49, 50). A common conclusion of these studies appears to be that further research in a form of randomised clinical trials was required for determining the efficacy of Autologous Growth Factors (AGF) and PRP in surgical procedures.

Several articles on systematic review of the studies on the effectiveness of autologous platelet rich plasma cited mixed and conflicting results. Cochrane Collaboration review of the bone augmentation techniques for dental implant treatment concluded that no reliable evidence was found in support of PRP use in this type of dental procedures (18).

A review of new treatment methods in orthopaedic practice asserts that albeit the US Food and Drug Administration approved marketing of autologous blood concentrates, very little clinical evidence existed in support of the use of this product for orthopaedic surgery (40).

To summarise the EBH findings, the existing evidence in support of autologous PRP and AGF use in surgical procedures appears to be conflicting, inconclusive and the quality and amount of evidence are not entirely convincing.

Background

The Evidence Based Healthcare (EBH) group completed a brief report on Autologous Growth Factors (AGF) and bone healing in November 2002. The conclusion of the literature review at that point was that little evidence of effectiveness of AGF in bone healing was found. It was recommended to review the literature when new research in this area becomes available.

Dr Margaret Mackey asked the EBH researchers to carry out an updated evidence-based review. This report supplements the EBH literature review completed in November 2002.

Search strategy

The following databases and cites have been searched for relevant literature:

- Ovid Medline, including Ovid Medline(R) In-Process & Other Non-Indexed Citations and Ovid Medline(R)
- EMBASE
- EBM reviews – ACP Journal Club
- EBM reviews – Cochrane Central Register of Controlled Trials
- EBM reviews – Database of Abstracts of Reviews of Effects
- EBM reviews Full Text – Cochrane DSR, ACP Journal Club, and DARE
- All EBM reviews – Cochrane DSR, ACP Journal Club, DARE, and CCTR
- Cochrane Oral Health Group¹
- Proquest
- References to relevant research listed in the referenced articles
- Handsearch of the following publications: Journal of Evidence Based Dental Practice and Journal of Oral and Maxillofacial Surgery : Official Journal of the American Association of Oral and Maxillofacial Surgeons, Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology

The websites related to the manufactures of the platelet rich products, such as Symphony™²(10) and ReGen Lab (48), have been perused for the information on the methods of the product preparation and references to the studies.

The website of the insurance company Cigna³ was viewed for its coverage position in respect to the use of platelet rich products in surgical treatment.

The websites of the Australian Therapeutic Goods Authority, European Agency for the Evaluation of Medicinal Products (EMA) and US FDA were searched for relevant information.

The WHO International Clinical Trials Registry⁴ was searched for Randomised Controlled Clinical studies. No studies have been found in respect to use of PRP in bone healing.

¹ <http://www.ohg.cochrane.org/trials.html>

² <http://www.carolinabloodmgt.com/main.htm>

³ <http://www.cigna.com>

⁴ <http://www.who.int/ictrp/en>

Google and Google Scholar search engines were used for supplementary search.

A Swiss company ReGen Lab⁵ was contacted and provided an information pack that the company is currently distributing to the orthopaedic surgeons in New Zealand.

Interpore International⁶ was contacted with the request to provide pertaining scientific articles but no response was received from the company.

The website for the Australian and New Zealand Horizon Scanning Network was scanned for the relevant material.⁷

Electronic search was conducted in March-May 2007, with the last search carried out on 14 May 2007.

Marx and Garg discussed a terminology confusion as references to PRP have been made under a variety of names (32). Although platelet rich plasma is the most accurate term to describe the product used in clinical practice. Other terms used in the literature have been included in the search with the key words used as: Autologous Growth Factors, AGF, Platelet derived growth factor, autologous platelet concentrate, PDGF, Platelet Rich Plasma, protein growth factors, osseous healing, fracture healing.

Selection criteria

Due to paucity of clinical studies on effectiveness of autologous platelet rich products, all papers published from 2002 to 2007 involving use of platelet rich plasma in orthopaedic, spinal, dental, periodontal and maxillofacial surgery were selected for this review. The publications were limited to human studies.

The research articles related to use of autologous PRP in soft tissue healing, plastic surgery, ophthalmology, paediatrics and cardiothoracic surgery were viewed but not analysed. The studies involving synthetic platelet-derived growth factors, such as Bone Morphogenic Proteins were not included in this review.

⁵ <http://www.regenlab.com>

⁶ <http://www.interpore.com>

⁷ <http://www.horizonscanning.gov.au/internet/horizon/publishing.nsf/Content/search-results-2?OpenDocument&FREETEXT=autologous+platelet+rich+plasma&BOOLEAN=any>

Investigation

All the relevant studies were assessed using the SIGN grading of evidence scoring system (Appendix 1).

This evidence-based review examined the study results in three main areas of PRP use in bone repair: orthopaedic surgery, spinal surgery, and dental, periodontal and maxillofacial surgery.

The reviewed papers are summarised in the tables below.

Table 1. Studies on the PRP use in orthopaedic surgery

Reference	Design score	Study design	N & age in treatment group	N & age in control group	AGF preparation and application technique	Key results	Study conclusions and/or recommendations
Bibbo et al (3)	3	Case series of patients with a high risk of non-union of foot and ankle surgery	62 (16-76)	0	PRP was obtained using the Symphony™ I & II platelet concentrating system (DePuy Acromed™, Raynham, MA) and combined with calcium chloride and thrombin. APC product was applied intra-operatively after preparation of bony surfaces.	Plain radiographs were taken every 2 weeks post-operatively until radiographic union was achieved. The mean time for union for the PRP alone (no bone graft use) group was 40 days; for autograft with PRP the mean time was 31 days; for allograft with PRP the mean time to union was 45 days.	Patients at high risk of non-union undergoing elective foot and ankle surgery may benefit from the use of PRP with or without the use of allograft or autograft.
Franchini et al (21)	3	Prospective case series of PRP use in treatment of impaired fracture repair or in reconstructive surgery	19 patients (15-69)	0	Platelet gel was used in a combination with hydroxyapatite. The blood was obtained and centrifuged a day prior to surgery. Platelet poor plasma was frozen overnight, then thawed and mixed with platelet concentrate before the procedure.	Radiographs and clinical outcome were evaluated every 3 months. No peri- or postoperative surgical complications were reported, and the graft consolidation was described as complete.	Long-term follow up studies, including histological verification of osteoinductive properties of platelet rich gel, are required to confirm the safety and efficacy of this treatment.
Coetzee et al (12)	2+	A prospective study of patients undergoing syndesmosis fusion in the agility total ankle replacement	66 (41-79)	114 (40-83) (retrospective cohort)	Symphony PCS (DePuy, Warsaw, Indiana) was used to obtain a platelet concentrate	Syndesmosis fusion was evaluated on radiographs by an	AGF application appears to improve the syndesmosis union rate

					from patient's blood. The concentrate was combined with thrombin and the gel sprayed directly onto the prepared bone surfaces.	independent radiologist at 8 weeks, 12 weeks, 16 weeks and 12 months after the operation. Complete fusion rate was recorded at 8 weeks 76%, at 12 weeks 93.9% and at 6 months at 96.9%. In comparison, in the historic cohort corresponding fusion rates were 61.4%, 73.6% and 85.1%. Non-unions and delayed unions were recorded at 3% and 6% in the AGF group and 15% and 26% in the control group.	in the Agility total ankle replacements.
Barrow et al (2)	3	Case series preliminary assessing syndesmotic fusion rates in total ankle arthroplasty	20 (40-80)	Historical controls	PRP was obtained by using Symphony™ PCS (DePuy Orthopaedics, Warsaw, Indiana)	Fusion rate observed at 6-month was estimated at 100%.	Authors suggest that the improved rate of distal tibio-fibular fusion <u>may</u> be attributable to the application of autologous platelet concentrate.
Bielecki & Gazdzik (4)	3	A case study of a patient with non-union of a diaphyseal humerus fracture	1 (50)	0	Patient's blood was centrifuged at 3200 rpm, and the PRP component was mixed with 10% calcium	The non-union was treated with percutaneous autologous platelet-	Percutaneous PRP could be effective in treatment of non-union.

					chloride and bovine thrombin.	rich gel injection into the non-union site. To assess bone consolidation clinical assessment, x-rays and DEXA examinations were carried out up to 24 weeks after the procedure. Remodelling of the union and increase in bone mineral density was reported.	
Trawick (47)	3	A case study of a patient with non-union of tibia and fibula fractures	1 (38)	0	PRP was prepared by using Symphony™ PCS ⁸ (DePuy Orthopaedics, Warsaw, Indiana)	Plain radiographs and clinical outcome up to 10 months post-operatively suggested complete healing of the non-union	Augmentation of bone grafts with PRP has significant potential for treatment of fracture non-unions
Watson (49)	3	Case series evaluating bone healing following augmentation of demineralised bone matrix for treatment of recalcitrant non-united fractures of long bones	6 (25-82)	0	PRP was obtained by using Symphony™ PCS (DePuy Orthopaedics, Warsaw, Indiana)	Post-operative radiographs and clinical assessment were reported as indicative that bone consolidation was achieved in all cases.	Demineralised bone powder in a combination with platelet concentrate may provide a bone graft substitute.
Schmidt (45)	3	Case series describing the use of PRP in tibial bone grafting	3 (34-39)	0	PRP was obtained by using Symphony™ PCS (DePuy Orthopaedics, Warsaw, Indiana) Platelet concentrate was mixed with cancellous chips.	Radiographs taken at 6 months post-operatively showed complete healing of non-union sites.	Platelet concentrate assisted bone consolidation in non-union of the tibial shaft and the proximal tibia, and aided healing of

⁸ ***Platelet Concentrate System***

							osteomyelitis of the distal tibial metaphysis.
Grant et al (24)	3	Retrospective case series comparing the fusion rates in Charcot's foot reconstruction surgeries among patients with diabetes. The comparison was made between 2 methods of PRP preparation	50	0	PRP was obtained by either use of Interpore Cross AGF system or Symphony™ PCS	Assessment of post-operative radiographs of patients with foot reconstruction surgeries suggested a better fusion rate for AGF cases compared with augmentation with Symphony products. The difference was not considered to be statistically significant.	No recommendations
Rodeo et al (40)	2++	Review of new developments in orthopaedic research					Very little evidence of effectiveness of autologous PRP use in bone healing exists at this time.

Table 2. Studies on the PRP use in spinal surgery

Reference	Design score	Study design	N & age in treatment group	N & age in control group	AGF preparation and application technique	Key results	Study conclusions and/or recommendations
Hee et al (26)	2-	A prospective clinical study of patients who underwent transforaminal lumbar interbody spinal fusion (TLIF) with application of AGF	23 (mean age 44.3)	111 - Historical cohort without AGF application	Blood processed through a 2-stage platelet sequestration protocol and concentrated using Ultraconcentrator, Interpore Cross. AGF concentrate was mixed with thrombin. AGF gel then was mixed with bone grafts for application to the fusion sites. 489% increase in the platelet count from the baseline to AGF concentrate stage.	Results suggest faster bony healing in the treatment group: at 4 months post-operatively 70% of patients showed radiographic evidence of bony consolidation, compared with 36% in the control group. At six months follow up 96% in the treatment group had bony consolidation on radiographs, compared with 64% in the control group. No significant difference between the groups in pseudoarthritis rates (4% for the AGF treatment group vs 6% for the historical cohort). The non-union rate for multiple-level fusions was slightly higher for	Use of AGF in TLIF procedures <u>may</u> expedite bone consolidation, however it does not result in overall increase in spinal fusion rates. Further improvements in AGF application technique are needed before this method can be used routinely.

						the treatment group (10% vs 8%).	
Jenis et al (27)	2+	A prospective clinical study of treatment outcome for patients with standard anterior-posterior interbody fusion. Treatment group underwent fusion with iliac crest bone autograft; control group had allograft combined with AGF	15 (40.3)	22 (41.4)	The buffy coat was sequestered from whole blood and placed into an ultraconcentrator for eventual formation of a platelet-rich solution. Obtained AGF was added to thrombin and mixed with crushed cancellous autograft chips.	CT scan and radiographs evaluation at 12 and 24 months showed no significant difference between fusion rates. Clinical assessment of back pain and functional outcome had similar results for both groups.	Allograft augmented with AGF is a reasonable alternative to autograft in one- or two-level lumbar interbody fusion. Future research is required to refine this treatment technique.
Weiner & Walker (50)	2+	A retrospective study of two groups of patients who underwent lumbar fusion with autologous graft with and without application of AGF concentrate	32 (32-84)	27 (31-80)	Following blood centrifugation a platelet concentrate was obtained from the buffy coat by using Intepore Cross International equipment. The AGF concentrate was combined with thrombin and applied to iliac crest autogenous cancellous bone graft.	At 1 year post-operatively in the treatment group 62% of patients achieved solid arthrodesis, 36% developed clear pseudoarthrosis, with 2 fusions failed at 2 years. At 1 year post-surgery in the control group 91% of patients achieved solid fusion, 1 patient developed clear pseudoarthrosis, and 2 fusions failed.	Lumbar intertransverse fusion rates were inferior for the treatment group. Possible explanation is that AGF inhibit osseogenerative role of bone morphogenic proteins.
Bose & Balzarini (7)	3	A prospective clinical study of patients who underwent posterior-lateral inter-tranverse spine fusion surgery	60	0	AGF were obtained by using an ultraconcentrator (Intepore Cross International, Irvine, Calif). AGF concentrate mixed with thrombin into AGF gel.	Radiographs were assessed in respect to fusion healing at regular intervals: at 1, 3, 6, 9, 12 and 24 months. 58 out of 60	No adverse effects attributable to AGF were detected.

						patients had solid or maturing fusions at 12 months.	
--	--	--	--	--	--	--	--

Table 3. Studies on the PRP use in dental, periodontal and maxillofacial surgery

Reference	Design score	Study design	N & age in treatment group	N & age in control group	AGF preparation and application technique	Key results	Study conclusions and/or recommendations
Raghoobar et al (38)	2++	Split-mouth study evaluating effects of PRP on remodelling of autologous bone grafts used for augmentation of maxillary sinus floor	5 (57-62)	0	PRP was made using commercial Platelet Concentration Collection System kit (Palm Beach Gardens, Florida, USA). PRP solution was mixed with 10% calcium chloride solution and the patient's serum as source of autologous thrombin. The resulting gel was combined with a bone graft.	PRP enhanced graft was used in bilateral maxillary sinus floor grafting for patients with severe resorption of sinus floor. Histological, clinical and radiological evaluation did not show any significant differences between the patients treated with bone graft only and bone graft combined with PRP.	No beneficial effect of PRP use was noted.
Froum et al (23)	2++	Split-mouth study testing efficacy of PRP in bilateral maxillary sinus grafts with bovine allograft	3 (35-69)	0	Patients' blood was centrifuged with a Sequestra 1000 gradient density cell separator (Medtronic). Platelet concentrate was mixed with bovine thrombin to form gel, which was added to graft material.	Test implants were biopsied and assessed by a blinded histologic and histomorphometric evaluation. The variables assessed comprised the total volume of calcified material, percentage of	Augmentation with PRP made no difference in bone production or in interfacial bone contact.

						vital bone, percentage connective tissue, percentage residual graft material, and percentage of bone at the implant-bone interface.	
Consolo et al (13)	2++	Split-mouth study comparing bone maxillary sinus floor regeneration using autologous graft with and without application of PRP	16 (37-57)	0	Patient's blood was centrifuged using RC3C, Sorvall, Thermo Electron Corporation, Waltham, MA, USA. Platelet concentrate was then mixed with autologous thrombin to form gel.	Radiological follow up, densitometry and core biopsy from the site of implant were analysed. Clinical performance showed no difference between treatment and control sites. Densitometric values were initially higher at treatment sites but the values converged at 8 month post-surgery. Histology results indicated higher osteoinductive activity at 4 month after the treatment, declining by 7 months.	No statistical difference between treatment and control sites were found.
Mannai (31)	2-	Prospective study of maxilla bone grafting with xenograft, autograft and PRP in patients with maxilla atrophy	97 (16 to 87)	0	Autologous platelet gel was obtained by using SmartPrep II (Harvest Technologies, Plymouth, MA and Munich, Germany) which provides concentration up to 700% of baseline.	Evaluation of x-ray and CT scan results at 3 months post-procedure showed 'excellent' bone maturation. In 97.8% of cases healing was reported as 'uneventful'.	Clinical healing time was reduced and the amount of autologous bone required for grafting was less, thus potentially reducing morbidity associated with graft harvesting.

Oyama et al (36)	2-	Prospective case-control study of patients who underwent alveolar bone grafting for alveolar cleft. Cancellous iliac bone was used as a graft. PRP was mixed with the particles of cancellous bone chips and packed into the alveolar cleft. Control group received cancellous iliac bone graft with human fibrin glue but with no PRP added to the graft.	7 (average age 16.1)	5 (average age 16.4)	PRP was obtained by centrifugation. Human fibrin glue (Beriplast P) was added to the PRP concentrate.	Evaluation of CT scan before and at 5 or 6 months after the operation suggested enhanced bone regeneration in the treatment group. The measurement used was percentage of volume of regenerated bone of volume of alveolar cleft. The higher percentage indicated greater osteoregeneration. In the treatment group average VRB/VAC was 80.19%, in the control group the value was estimated at 63.67%), which suggested better osteoregeneration in the treatment group.	Incorporation of PRP into autograft could enhance the osteogenesis of alveolar bone grafting.
Curi et al (15)	3	Case study of 3 patients with avascular non-healing osteonecrosis of the mandible following dental extraction. Avascular necrosis developed following cancer treatment with bisphosphonate therapy. Bone resection of the affected bone was combined with topical application of autologous platelet-rich plasma (PRP) to the entire bone cavity bed as an	3	0	Automated autologous platelet concentrate system (Smart PRP Harvest Technologies, Plymouth, MA)	The post-operative follow up showed: Case 1 – at 6 months complete healing of the surgical site Case 2 – at 6 months partial healing of the surgical site Case 3 – at 8 months healing of the oral mucosa and alveolar	Findings of the study were reported as not conclusive. Larger randomised prospective studies are recommended.

		adjunctive therapy.				bone at the surgical site	
Camargo et al (8)	2++	Split-mouth study examining the role of a combination of PRP, bovine porous bone mineral and guided tissue regeneration in treatment of intrabony defects secondary to periodontal disease	18 (39)	0	Patients' blood was mixed with 10% trisodium citrate as an anticoagulant, and then centrifuged at 5600 rpm for 6 min. The red blood cell fraction was mixed with 10% calcium chloride and 100U/ml of bovine thrombin into a sticky gel. The gel was mixed with cancellous bovine porous bone mineral granules.	Changes in pocket depth were measured at 6 months after the surgical treatment. A greater decrease in pocket depth, suggesting a greater clinical gain, was reported for defects treated with a combination of PRP/BPBM.	The results suggested that PRP and bovine porous bone mineral enhanced regenerative effect of guided tissue regeneration in patients with severe periodontitis.
Christgau et al (11)	2++	Prospective split-mouth study assessing outcome of autologous PRP augmentation in osseous regeneration following guided tissue regeneration therapy in deep intrabony periodontal defects	25	0	Platelet concentrate was obtained by using an apheresis technique. The concentrate was mixed with 10% calcium chloride. A median plasma enrichment factor was estimated at 7.9.	Clinical and radiological measurements were statistically evaluated. Radiological evaluation indicated a greater bone density gain at test sites compared with control sites at 3 and 6 months. In contrast, after 12 months control sites suggested a better bone density gain.	PRP use did not appear to enhance clinical and radiological outcomes.
Ouyang et al (35)	2++	Split-mouth study comparing treatment of periodontal intrabony defects with bovine porous bone mineral (BPBM) graft only and with application of PRP as an adjunct to BPBM.	10 patients with 17 intrabony defects (test group – 9 defects,	0	PRP obtained by blood centrifugation and then mixed with a saline solution and human thrombin.	Periapical radiographs 12 months after the procedure showed regeneration of periodontal tissues in both groups.	Significantly favourable clinical outcome from a combination of PRP with BPBM. Further studies are needed to

			control group – 8 defects) (27-45 years old)			Comparison of clinical parameters suggests a favourable outcome for the treatment group. The clinical outcome measured at 1 year post-surgery indicated statistically significant improvement in probing depth reduction, attachment gain, bone probing reduction and defect bone fill for the treatment group. Digital subtraction radiographs at 1 year post-operatively showed significantly more radiographic gains in alveolar bone mass in the treatment group.	assessing long-term effectiveness of PRP.
Sammartino et al (42)	2++	Split-mouth study looking into efficacy of PRP in bone regeneration for treatment of periodontal defects at distal root of the mandibular second molar following surgical extraction of deeply impacted mandibular third molar.	18 (21-26 years old)	0	Patients' blood was mixed with a 10% trisodium citrate anticoagulant solution. Pre-operatively the mixture was centrifuged at 1200 rpm and PRP concentrate was extracted. Intra-operatively the PRP concentrate was mixed with batroxobine and 10% gluconate of calcium. The mixture was shaken up	Orthopantomogramme and intraoral radiogrammes at 12 and 18 weeks post-operatively were evaluated. In cases treated with PRP, osseous biopsy was taken at 1 2 weeks to assess bone regeneration.	PRP is effective in enhancing bone regeneration for treatment of periodontal defects.

					for 30 seconds. The gel was applied to the bone defect wall and to the root surface of the second molar.		
Papli et al (37)	2	Prospective case study compared the effects of two treatment options for intrabony periodontal defects: an intralesional graft of autologous platelet concentrate with guided periodontal regeneration using a bioabsorbable barrier membrane (MEM)	5 (33)	0	Platelet concentrate was obtained by using platelet concentrate collection system (PCCS, 3i Implant Innovations, Palm Beach Gardens, FL)	Radiographs were assessed at 8, 26 and 52 weeks suggested an increase in bone formation in case and control sites.	Results did not demonstrate benefits of APC application as the clinical outcome was similar for both treatment options. Further clinical trials are needed.
Ribiony et al (39)	3	Case series of severe atrophic mandible restoration with the use of a combination of autologous bone graft with autologous PRP	5 (48-63)	0	Patients' blood underwent a 2-stage centrifugation with a platelet concentrate combined with 10% calcium chloride and the patients' serum as source of autologous thrombin. The mix was added to the autologous iliac crest bony fragments.	Non infection or complications were reported. Radiographic results showed considerable enhancement of bone regeneration at 6 months.	This method appears to be an effective way in treatment of severe atrophic mandible.
Hanna et al (25)	1-	Randomised split-mouth clinical trial comparing the clinical outcome for sites treated with allograft alone and with allograft augmented with PRP in treatment of periodontal intrabony defects	13 (37-74)	0	Patient's blood was processed through a centrifuge (SmartPreP, Harvest Technologies Corp., Plymouth, MA). Platelet rich concentrate was then mixed with 10% calcium chloride and topical thrombin	Probing depth, clinical attachment level and recession were measured at the baseline examination and at 6 months post-surgery. Both treatment and control sites showed significant clinical improvement on the measured parameters. Clinical periodontal	Augmentation of allograft with PRP improved periodontal response in comparison with using allograft alone.

						response was deemed to be better in the PRP group.	
--	--	--	--	--	--	--	--

Discussion

The enhancement of bone formation by using augmentation with PRP is a growing area of research. A number of papers have been published on studies examining the role of PRP and AGF in bone healing. However no studies with a strong degree of evidence from high quality randomised controlled trials have been identified from the thorough literature search.

A number of the published studies used low score non-analytic design, such as case reports and case series and, with a high risk of the results being unreliable ((2-4, 12, 15, 21, 24, 39, 45, 47, 49). Most studies recommend further research in a form of randomised clinical trials for determining effectiveness of the use of AGF and PRP in bone repair.

Reports on systematic review of the studies on effectiveness of autologous platelet rich plasma indicate that available evidence in support of the PRP use is weak (18, 40, 43).

In-depth review of the publications on autologous platelet rich products available in the USA describes a number of methods of obtaining autologous PRP (41). The authors conclude that autologous platelet rich plasma is a safe and effective treatment modality.

Methods of obtaining PRP vary, with most of the studies describing centrifugation for obtaining a high platelet concentration and mixing the concentrate with human fibrin to achieve a gel consistency. Depending on the sequestering technique platelet concentration in the final product has considerable variations. Autologous Growth Factors technique, a proprietary platelet concentration system, claims more advanced ultrafiltration methods resulting in superconcentration plasma up to 8-10 times of the whole blood. Other methods of PRP preparation report platelet concentration to be from 4-6x (Symphony II) to 8x (GPS II) (41).

Orthopaedic surgery

A limited number of studies on use of autologous PRP in orthopaedic surgery have been published up to date. Reports on PRP use in the accelerating of bone repair include augmentation with platelet rich gel for enhancing fracture healing, in particular in patients at risk of non-union, and treatment of existing non-union.

Nine articles on use of PRP in orthopaedic surgery were identified. All the articles represent descriptive studies, namely case studies and case series. Out of these reports, several articles describing effectiveness of PRP use were found on the Symphony website. Thus interpretation of the results is to be cautious due to potential involvement of the product manufacturer in these studies (45, 47, 49).

All the reported studies indicated a degree of benefit arising from PRP augmentation in the orthopaedic procedures. Bibbo et al (3) assessed the results of adjuvant use of autologous platelet concentrate (APC) in a group of patients at a high risk of non-union. Sixty two patients underwent 123 surgical procedures on foot and ankle. Out of these procedures 56 fusion sites required grafting, which was done either by allograft or autograft combined with APC. Other fusion sites were treated with the application of autologous platelet concentrate only. The

study achieved 94% overall union, with the mean time to union for all study patients at 41 days. The overall improved union rate was attributed to the use of the APC. The authors concluded that the adjunctive use of APC in patients at high risk of non-union in elective foot and ankle surgery may assist with achieving acceptable time to union.

Franchini et al (21) reported an osteoinductive effect of autologous platelet concentrate use in reconstruction surgery and treatment for impaired fracture repair. In the case series of 19 patients undergoing reconstructive bone surgical procedures the clinical and radiological outcome was described as complete osteointegration of the graft. The study used radiographs and clinical assessment for individual patients as a means of evaluating the results. The authors interpreted the study results as suggestive of PRP efficacy in bone regeneration and of a PRP favourable effect in reconstructive surgery.

A significant benefit from using AGF augmented graft was reported by Coetzee et al (12). The study used AGF application for the Agility total ankle replacement in 66 patients. A comparison of the radiographic results with the historic cohort of 114 ankle replacement procedures without AGF application suggested an improved syndesmosis union rate. The arthrodesis rate was estimated at 94% for the AGF augmented group compared with 73% union rate for the historic controls. The authors concluded that the AGF use significantly improved arthrodesis rate for this procedure.

In a preliminary report on the above study, its co-authors reported a greater arthrodesis rate in total ankle arthroplasty (2). The authors cautioned that the improved syndesmotic fusion rates may be attributable to variables other than the use of PRP, such as a different operative technique and a modification in the post-operative protocol.

Several case studies reported successful results from the use of PRP for treatment of non-union (4, 45, 47). The common conclusion of these studies was that albeit the results were promising, further clinical studies into efficacy and safety of PRP application were required.

A retrospective study of the PRP use in treatment of complex neuropathic fractures in the diabetic population by Grant et al made a comparison of fusion rates between two groups of patients treated with application of either Interpore Cross AGF system or the Symphony PRP concentration system (24). The overall fusion rate was higher for the foot reconstruction procedures augmented with Interpore Cross AGF. However the difference was reported as not being statistically significant. The authors reported that the overall fusion rates with the use of either technique were greater than the results found in the literature.

A conclusion on the need for further clinical trials into effectiveness of PRP augmentation was a common theme of a majority of the reviewed articles (3, 4, 21, 24, 30, 45).

The published studies seem to support use of PRP in some types of orthopaedic surgery. However it appears that the level of evidence these studies provide is at a low grading level. This impression is reinforced by two articles on critical analysis of PRP use in orthopaedic surgery (16, 40).

A critical overview of bone grafts and bone graft substitutes in orthopaedic trauma surgery analysed the studies published up to date on effectiveness of various osteoinductive strategies (16). The authors acknowledged that intuitive impression was that use of a platelet concentrate would contribute to bone healing. Hence a number of strategies have been developed for augmenting bone graft with the substance. However the authors pointed out that the indication for PRP use was based on low level evidence. The overview concluded that scientific evidence in respect to clinical effectiveness of autologous platelets use was insufficient, and that more robust design clinical trials were needed.

Similarly, an overview of advances in orthopaedic research acknowledged that albeit autologous blood concentrates have been approved for marketing by the US FDA, very little clinical evidence in support of efficacy of PRP in bone healing exists (40).

To sum up, albeit the papers on the use of autologous PRP gel in some orthopaedic surgical procedures present beneficial effect of the PRP augmentation, these studies provide low grade evidence that has not been corroborated by randomised clinical trials.

Spinal surgery

Four papers on platelet rich gel use in spinal surgery were found, suggesting that research on autologous PRP application in the spinal fusion procedures was limited. The results of the studies on efficacy of PRP application were conflicting.

Two studies reported beneficial outcome from the application of autologous platelet products in lumbar interbody fusion (26, 27). In contrast to these studies was a paper by Weiner & Walker (50) reporting possible negative impact from PRP augmentation and generating fierce debate in the orthopaedic community (1, 5, 34).

A prospective clinical study by Hee et al (26) evaluated results of transforaminal interbody spinal fusion (TLIF) augmented by AGF. The outcome was compared with a historical cohort of patients who underwent similar procedures by the same surgeons without the use of AGF. The results of the radiological review suggested that AGF application expedited bony consolidation but did not lead to increase in the spinal fusion rates. The authors indicated that outcome of AGF application may depend on the AGF preparation techniques, and suggested that further AGF refinement in the AGF application method was required before the procedure can be used routinely in spinal fusions. The study included a small sample of patients with the factors, such as previous spinal surgery and pseudoarthrosis, which may have impacted on the fusion rates.

In another prospective clinical study Jenis et al (27) compared radiological and clinical outcomes for 37 patients who underwent standard posterior-anterior lumbar fusion with autograft and allograft enhanced with AGF. Radiographic analysis of CT scans at 6 months after the procedure indicated fusion incorporation at 56% for both groups. At 24 months fusion rate for the treatment group was at 89%, for the control group at 85%. The study concluded that allograft augmented with AGF was an acceptable alternative to autograft. The

authors suggested that use of allograft combined with AGF for this procedure could eliminate morbidity associated with autograft harvesting.

Contrary to the above mentioned reports on successful use of PRP and AGF is the Weiner's (50) study, and its hypothesis that AGF concentrate may hinder bone consolidation.

The conclusion of Weiner's study was that the use of AGF resulted in inferior lumbar fusion rates when compared with using autograft non-augmented with AGF.

A retrospective study compared a control group that underwent a single level spinal fusion with iliac crest bone graft with a test group that used autograft augmented with AGF. The radiography results evaluated at one and two years showed an inferior fusion rate in the test group. The fusion rate in the group with conventional autograft was estimated at 91%, while the fusion rate in the group with autograft augmented with AGF was calculated at 62%.

The authors hypothesized that AGF in a concentrated form may inhibit healing properties of bone morphogenic proteins, hence having a negative impact on bone healing.

Subsequent to the publication of this research a number of orthopaedic surgeons expressed reservations regarding the study conclusions. Arm (1), Nucci (34), Birch (5) and Boden (6) pointed out the deficiencies of the study design and questioned validity of the study results.

A retrospective study by Bose (7) reported lumbar spine solid fusion in 58 out of 60 patients who underwent spinal fusion with autologous graft augmented with AGF. No adverse effects from the use of AGF were noted. The purpose of the study was to describe a particular method for obtaining and applying AGF, and it did not compare fusion rate against other spinal fusion techniques.

Overall, paucity of published studies on spine fusion does not provide convincing evidence of either detrimental or beneficial effects of autologous PRP for this procedure.

Dental, periodontal and maxillofacial surgery

Fifteen published studies and two papers on systematic review on the use of PRP in dental, periodontal and maxillofacial surgery have been identified. Conclusions of the studies appear to be conflicting and ranging from the papers that indicate significant benefits to some treatment outcomes of the PRP application (8, 15, 31, 35, 36, 39, 42) to the studies that suggest no additional value in using this treatment method in comparison with conventional surgical techniques (11, 23, 37, 38, 44). Both papers on systematic review comment on lack of scientific evidence to support the use of PRP (18, 43).

Impregnation with PRP is currently used in a variety of surgical procedures, such as alveolar bone grafting, maxillary sinus floor augmentation, facial reconstructive surgery and treatment of periodontal intrabony defects. Not all these applications of PRP are relevant to claims given cover under the ACC scheme. PRP use in

reconstructive dental and maxillofacial surgery is pertinent to the use of the product for injury-related surgical procedures. Augmentation with PRP of a sinus graft is used as a method for accelerating enhancement of maxilla bone. This treatment may be indicated for patients with maxilla insufficient for dental implant that could be required as a result of dental injury.

Five papers on application of PRP in periodontal surgery relate to intrabony defects secondary to periodontal disease, a gradual process which is of little relevance to the dental problems covered by ACC (8, 11, 25, 35, 37). However for completeness of this report all these papers have been included in the review as the studies were indicative of PRP role in bone healing.

Few reports on the effects of the PRP use for the maxillary sinus floor augmentation did not suggest beneficial outcome from this therapeutic approach.

Raghoobar et al in a split-mouth study on remodelling of autologous bone grafts used for augmentation of the maxillary sinus floor reported no benefits in the use of PRP (38). Patients with a severely resorbed maxilla were randomly allocated into two groups: treated with augmentation with autologous graft taken from the iliac crest and treated with graft combined with PRP. No difference in clinical assessment was noted, and the microradiological and histomorphological examination of the biopsies demonstrated no difference in the results. The study reported no benefit from using PRP for maxillary sinus floor augmentation.

The conclusion of Raghoobar's study corresponds with the findings of another split-mouth study of bilateral maxillary sinus floor grafts. Froum et al assessed efficacy of PRP in enhancing allografts in three patients (23). The study used histological and histomorphometric evaluation as a means of evaluation. The authors concluded that the addition of PRP to allografts did not make a significant difference in bone production or in interfacial bone contact in the test implants. The paper appears to indicate that PRP application may be beneficial in treatment of relatively small periodontal defects and in larger bone defects grafted with autologous bone. But its effectiveness has not been shown in treatment of larger allografts.

The results of Consolo et al study on effectiveness of PRP in maxillary sinus floor augmentation are in line with the previous studies (13). A split-mouth study of 16 patients randomised for allograft with or without impregnation with PRP showed no statistical difference between the treatment and control sites.

Several studies reported some beneficial effects in osseous healing from the use of PRP in oral and maxillofacial procedures.

A prospective study by Mannai of 97 patients who had dental implants in background of severe maxillary atrophy showed 97.8% healing rate (31). The study assessed 314 implants placed simultaneously with bone grafts in the maxilla. Bone grafts were composed of 25-30% of autologous bone and 70-75% of xenogenous bone (bovine). The patients had autologous platelet concentrate added to grafts and implants. The author suggested the benefits of the APC use in this procedure were in reduced healing time and in reduction of amount of autologous bone required for grafting. The former assertion was compared against 6 to 9 months healing time when APC was not used, as opposite to 3 month with the use of APC. The latter indicated a substantial reduction in the autologous graft

size, hence reducing the morbidity associated with graft harvesting. While the study reported a high success rate, there was no convincing comparison with other treatment options of the results this method achieved.

Oyama et al reported that grafting of alveolar cleft with application of PRP to the graft significantly enhanced osteoregeneration within 5-6 months after the operation (36). However there was no follow up after 6 months, and the authors cautioned that PRP long term bone-regeneration effects and their impact on success of tooth implantation subsequent to grafting were not known.

A unique case study on PRP use for treatment of avascular necrosis of the mandible in oncology patient reported inconclusive results (15). Three patients selected for this procedure had avascular necrosis which was associated with treatment with bisphosphonates, the drugs that were known to inhibit normal osteoclastic function. Hence this condition could potentially relate to ACC cover under the treatment injury provisions. Treatment of the cases consisted of surgical resection of the necrotic tissues with subsequent application of autologous PRP. In two cases of the complete healing of the surgical sites was noted at 6 and 8 months post-operatively. One patient had a partial healing at the resection site at 6 months. The authors indicated that this treatment modality showed promising results and warranted further clinical trials.

As discussed above, bone loss secondary to periodontal disease does not relate to injuries covered by ACC, and the papers on the use of PRP in periodontal treatment have been reviewed for completeness of the impression on PRP role in osseous healing. One of the characteristics of periodontal disease is loss of alveolar bone around the affected teeth. Conventional treatment methods of advanced periodontal disease include grafting and Guided Tissue Regeneration (GTR). The studies compared outcome of treatment with PRP augmentation against these conventional therapies.

A split-mouth study by Camargo et al (8) assessed clinical effectiveness of PRP in treatment of periodontal intrabony defects. The study selected eighteen systematically healthy patients with advanced periodontitis for treatment with Guided Tissue Regeneration (GTR) technique only, and with a triple therapy consisting of a combination of PRP and Bovine Porous Bone Mineral (BPBM) with GTR. Intrabony defects were randomised to be treated by either of these modalities. The clinical outcome was measured in respect to pocket depth, clinical attachment level and defect fill. The results indicated a greater decrease in pocket depth and a significantly better gain in clinical attachment in the triple therapy group. The authors concluded that the treatment with a combination of PRP, BPBM and GTR was more effective than treatment with GTR alone, and that the combination of PRP with BPBM enhanced osteoregenerative effect of GTR.

In another split-mouth study of 25 patients with intrabony periodontal defects Christgau et al treated randomly allocated sites using surgical procedure with and without application of autologous platelet concentrate (APC) (11). Comparison of bone regeneration between test and control sites found no clinical or radiological indicators of effectiveness of APC. Radiological evaluation at 3 and 6 months post-operatively suggested a better bone density gain in the sites treated with the platelet concentrate. However at 12 month post-surgery bone density gain in the control sites appeared to be greater. Overall conclusion of the study was the value of the APC for periodontal regeneration was questionable.

A case-control study by Ouyang et al reported a greater alveolar gain and a better clinical outcome for patients who underwent grafting for intrabony periodontal defects with an adjunct application of autologous PRP to bovine porous bone mineral graft (35). Albeit the study design was robust and its findings statistically significant, the sample size was small and the follow up was limited to 12 months post-procedure. Likewise, a comparison of clinical periodontal responses between bone-derived allograft alone and PRP-augmented allograft showed a better clinical outcome from the application of autologous PRP to the bovine graft (25).

Correspondingly to the conclusions of the primary studies, the articles published on systematic review of the studies came to conflicting conclusions.

Freymler reviewed the literature evaluating the effectiveness of PRP when used alone and in a combination with various types of grafts (22). The results of the studies assessing the effect of PRP augmentation on autologous graft varied from a greater degree of graft maturation to no osteoinductive effect. A similar conclusion was reached from the review of the studies on augmentation of anorganic bone mineral with PRP. The studies reported a range of results, from some extent of improved bone regeneration to no benefit with the addition of the platelet gel. The paper refers to the data on effectiveness of use of autologous PRP as '**conflicting**' and did not corroborate this conclusion by quantitative data from the study.

Analogously, the use of PRP in a combination with organic bone noted mixed results, with most studies detecting no bone growth stimulating effect from adding PRP to organic graft. When PRP was used alone, few in vivo studies showed slight increase in osseointegration or no benefit from using the gel. Freymiller suggested that possible explanation of the conflicting results is that mechanism of animal healing may be different from human, and that the different methods of platelet gel preparation may contribute to the variations in the bone healing process. The author suggested that the PRP may have minimal effect on bone healing, and pointed out that in view of paucity of scientific evidence on PRP effectiveness its use in surgical practice was inordinate.

Review of the scientific articles by Carlson et al indicated that PRP had proven effectiveness in periodontal and oral surgery applications (9). The review, however, included very few original studies, and the studies it reviewed were published between 1995 and 2000, making the results of this review obsolete.

Conversely, Esposito et al in the Cochrane Database of Systematic Reviews evaluated clinical trials involving bone augmentation techniques for dental implant treatment (18). The authors tested the null hypothesis of no difference in clinical outcome for different bone augmentation methods. The conclusion of the review was that no reliable evidence was found in support of efficacy of platelet rich plasma in conjunction with implant treatment.

Likewise, the literature review of bone growth factors in maxillofacial skeletal reconstruction by Schliephake found little scientific evidence on the effectiveness of PRP application (44). The review concluded that it was unlikely bone healing or regeneration would be significantly enhanced by the application of PRP alone.

Similarly to Schliephake's conclusions, a later review by Sanchez et al. summed up that in the absence of robust clinical trials, the level of evidence in support of PRP use was low (43). It was suggested that albeit PRP may have a significant osteoregenerative potential associated with the growth factors, no controlled studies had been carried out to verify efficacy of this treatment modality.

In the *Clinical Controversies in Oral and Maxillofacial Surgery* Marx unequivocally advocated for the use of PRP, and responded to the criticism of its application (33). Marx argued that the studies concluding little or no benefit from PRP augmentation involved PRP of poor therapeutic quality obtained through use of inadequate equipment. The paper refers to several studies that reported a successful application of PRP in various surgical procedures. Out of the mentioned studies very few were related to bone regeneration, a few reported results in soft tissue healing and other studies were outside the scope of this review.

Common criticism of the published studies can be summarised as following:

- a) Manufacturing processes for obtaining autologous PRP are diverse and inconsistent. Inadequate devices for obtaining PRP may affect viability of platelets and hence inhibit role of the bioactive growth factors (33).
- b) A number of studies involved a small research sample and thus were unable to produce statistically significant results.
- c) The majority of studies did not have control groups. Case studies and case series in the absence of a comparative sample provide a low degree of evidence of effectiveness.
- d) The studies used imprecise means of determining bone healing (ie. Using radiographs rather than direct visualisation or biopsy of the treatment sites).
- e) The period of observation of the effects of PRP application was short, and long-term consequences of its use have not been studied.

Conclusion

Overall impression from the reviewed literature is that lack of robust scientific studies does not allow the reviewer to either verify or refute the ability of PRP to enhance osseous healing.

Further to the EBH brief report of November 2002, a few clinical studies have been found that would support effectiveness of autologous Platelet Rich Plasma in treatment of impaired bone fracture healing. A majority of the studies suggested that further clinical trials needed to be carried out in order to determine efficacy of platelet rich plasma in bone healing.

Several articles on systematic review of the studies on effectiveness of autologous platelet rich plasma in orthopaedic and dental surgery do not support PRP use (16, 18, 40). The consensus of these articles appears to be that the current use of PRP is not based on strong scientific evidence.

The authors of several studies made unequivocal conclusions on the benefit of autologous PRP use in dentistry (8, 25, 35). However quality, size and validity of these studies do not appear to substantiate these conclusions. These studies do not

demonstrate robust evidence of effectiveness. Reported effectiveness of treatment modality derived from low quality trials is likely to be biased.

The current concepts review on the role of growth factors in the repair of bone discussed the in vitro and animal studies on the role of platelet-derived growth factors (PDGF) in fracture healing and bone repair (30). The review concluded that such studies did not determine clearly effectiveness of the PDGF in osseous healing. Lieberman et al also discussed clinical applications of commercial products containing growth factors in fracture healing and spinal fusion. In regard to fracture healing the authors summed up that the available preclinical data on PDGF in the treatment of non-union was insufficient to predict clinical effectiveness of growth factors applications. In respect to spinal fusion, the role of PDGF was unclear and required further analysis.

Orthopaedic surgery

Several case reports demonstrated accelerated bone healing with the use of PRP rich plasma. Augmentation with PRP reported as beneficial in cases of non-union and impaired fracture healing. However these reports are confined to case studies and case series, hence the level of evidence is not regarded as scientifically robust.

Spinal surgery

Few reports on the use of PRP have been identified. The conclusions of the reports are in conflict, and the overall impression is that superiority of the PRP use over conventional fusion has not been established.

Dental, periodontal and maxillofacial surgery

A few reports on the clinical benefits of autologous PRP augmentation need to be balanced against the reports on insufficient or absent clinical response from this treatment modality. The papers on systematic review of the existing studies do not appear to indicate endorsement for this therapy.

To sum up, the studies reported up to date provided no conclusive scientific evidence to determine that use of autologous PRP has significant benefits for osseous healing. Grading of evidence by SIGN scoring system suggests that a vast majority of studies are graded at a low level of evidence (3 or 2-), with a significant risk or moderate probability that the relationship is not causal.

Recommendations

Overall impression of the review of the existing publications is that evidence on the efficacy of autologous PRP use is contradictory, inconclusive and the quality and amount of evidence are not entirely convincing.

Although this therapeutic modality is deemed to be potentially promising, paucity of high quality evidence in a way of randomised controlled trials does not appear to conclusively support its use.

References

1. Arm D. Letters to the Editor. *Spine* 29 (8): 946-48, 2004.
2. Barrow C, Pomeroy C. Enhancement of syndesmotoc fusion rates in total ankle arthroplasty with the use of autologous platelet concentrate. *Foot & Ankle International* 26 (6): 458-61, 2005.
3. Bibbo C, Bono C, Lin S. Union rates using autologous platelet concentrate alone and with bone graft at high-risk foot and ankle surgery patients. *Journal of Surgical Orthopaedic Advances* 14 (1): 17-21, 2005.
4. Bielecki T, Gazdzik T. Percutaneous Injection of autogenous growth factors in patient with nonunion of the humerus. A case report. *Journal of Orthopaedics* 3 (3): e15, 2006.
5. Birch N. Letters to the Editor. *Spine* 29 (10): 1162-63, 2004.
6. Boden S. Efficacy of autologous growth factors in lumbar intratransverse fusions: point of view. *Spine* 28 (17): 1971, 2003.
7. Bose B, Balzarini M. Bone graft gel: autologous growth factors used with autograft bone for lumbar spine fusions. *Advances in Therapy* 19 (4): 170-75, 2002.
8. Camargo P, Lecovic V, Weinlaender M, Vasilic N, Madzarevic M, Kenney E. Platelet-rich plasma and bovine porous bone mineral combined with guided tissue regeneration in the treatment of intrabony defects in humans. *Journal of Periodontal Research* 37: 300-306, 2002.
9. Carlson N, Roach R. Platelet-rich plasma. Clinical applications in dentistry. *Journal of American Dental Association* 133: 1383-86, 2002.
10. Carolina Blood Management I. Speeding the healing process: Carolina Blood Management
11. Christgau M, Moder D, Wagner J, Glabl M, Hiller K, Wenzel A, Schmalz G. Influence of autologous platelet concentrate on healing in intra-bony defects following guided tissue regeneration therapy: a randomised prospective clinical split-mouth study. *Journal of Clinical Periodontology* 33: 908-21, 2006.
12. Coetzee JC, Pomeroy GC, Watts JD, Barrow C. The use of autologous concentrated growth factors to promote syndesmosis fusion in the Agility total ankle replacement. A preliminary study. *Foot & Ankle International / American Orthopaedic Foot And Ankle Society [And] Swiss Foot And Ankle Society* 26 (10): 840-846, 2005.
13. Consolo U, Zaffe D, Bertoldi C, Ceccherelli G. Platelet-rich plasma activity on maxillary sinus floor augmentation by autologous bone. *Clinical Oral Implants Research* 18 (2): 252-262, 2007.
14. Corporation CH. Cigna HealthCare Coverage Position. Wound healing: tissue engineered skin substitutes and growth factors. Cigna HealthCare Coverage position, 2006.
15. Curi M, Cossolin G, Koga D, Araujo S, Feher O, Oliveira D, Zardetto C. Treatment of avascular osteonecrosis of the mandible in cancer patients with a history of bisphosphonate therapy by combining bone resection and autologous platelet-rich plasma: report of 3 cases. *Journal of Oral & Maxillofacial Surgery* 65: 349-55, 2007.
16. De Long W, Einhorn T, Koval K, McKee M, Smith W, Sanders R, Watson T. Bone grafts and bone grafts substitutes in orthopaedic trauma surgery. A critical analysis. *The Journal of Bone and Joint Surgery* 89 (3): 649-73, 2007.

17. Dugrillon A, Eichler S, Kern S, Kluter H. Autologous concentrated platelet-rich plasma (cPRP) for local application in bone regeneration *International Journal of Oral & Maxillofacial Surgery* 31: 615-19, 2002.
18. Esposito M, Grusovin M, Worthington H, Coulthard P. Interventions for replacing missing teeth: bone augmentation techniques for dental implant treatment. *The Cochrane Database of Systematic Reviews*. The Cochrane Collaboration, volume 1 ed: The Cochrane Library, 2007.
19. Everts P, Mahoney C, Hoffmann J, Schonberger J, Box H, van Zundert A, Knape J. Platelet-rich plasma preparation using three devices: implications for platelet activation and platelet growth factor release. *Growth Factors* 24 (3): 165-71, 2006.
20. Floryan K, Berghoff W. Intraoperative use of autologous platelet-rich and platelet-poor plasma for orthopaedic surgery patients. *Association of Operating Room Nurses. AORN Journal* 80 (4): 668-674, 2004.
21. Franchini M, Dupplicato P, Ferro I, Gironcoli M, R A. Efficacy of platelet gel in reconstructive bone surgery. *Orthopaedics* 58 (2): 161-63, 2005.
22. Freymiller E, Aghaloo T. Platelet-rich plasma: ready or not? *Journal of Oral & Maxillofacial Surgery* 62: 484-88, 2004.
23. Froum S, Wallace S, Tarnow D, Cho S. Effect of platelet-rich plasma on bone growth and osseointegration in human maxillary sinus grafts: three bilateral case reports. *The International Journal of Periodontics & Restorative Dentistry* 22 (1): 45-53, 2002.
24. Grant W, Jerlin E, Pietrzak W, Tam H. The utilization of autologous growth factors in facilitation of fusion in complex neuropathic fractures in the diabetic population. *Clinics in Podiatric Medicine and Surgery* 22: 561-84, 2005.
25. Hanna R, Trejo P, Weltman R. Treatment of intrabony defects with bovine-derived xenograft alone and in combination with platelet-rich plasma: a randomised clinical trial. *Journal of Periodontal Research* 75: 1668-77, 2004.
26. Hee H, Majd M, Holt R, Myers L. Do autologous growth factors enhance transforaminal lumbar interbody fusion? *European Spine Journal* 12: 400-7, 2003.
27. Jenis L, Banco R, Kwon B. A prospective study of Autologous Growth Factors (AGF) in lumbar interbody fusion. *The Spine Journal* 6: 14-20, 2006.
28. Kurica K, Booton K, Giuffre J. Autologous growth factors and resorbable porous ceramic without bone graft for instrumented posterolateral lumbar fusion. *WorldSpine Meeting*. Chicago, Illinois: Carolina Blood Management, Inc, 2003.
29. Leitner G, Gruber R, Neumuller J, Wagner A, Kloimstein P, Hocker P, Kormoczi G, Buchta C. Platelet content and growth factor release in platelet-rich plasma: a comparison of four different systems. *Vox Sanguinis* 91: 135-39, 2006.
30. Lieberman J, Daluiski A, Einhorn T. Current concepts review: the role of growth factors in the repair of bone. *Biology and clinical applications. Journal of Bone and Joint Surgery* 84 (6): 1032-44, 2002.
31. Mannai C. Early implant loading in severely resorbed maxilla using xenograft, autograft, and platelet-rich plasma in 97 patients. *Journal of Oral & Maxillofacial Surgery* 64: 1420-26, 2006.
32. Marx P, Garg A. *Dental and craniofacial applications of platelet-rich plasma: Quintessence Publishing Co, Inc, 2005.*

33. Marx R. Platelet-rich plasma: evidence to support its use. *Journal of Maxillofacial Surgery* 62: 489-96, 2004.
34. Nucci R. Letters to the Editor. *Spine* 29 (10): 1163, 2004.
35. Ouyang X, Qiao J. Effect of platelet-rich plasma in the treatment of periodontal intrabony defects in humans. *Chinese Medical Journal* 119 (18): 1511-21, 2006.
36. Oyama T, Nishimoto S, Tsugawa T, Shimizu F. Efficacy of platelet-rich plasma in alveolar bone grafting. *Journal of Oral & Maxillofacial Surgery* 62: 555-58, 2004.
37. Papli R, Chen S. Surgical treatment of intrabony defects with autologous platelet concentrate or bioabsorbable barrier membrane: a prospective case series. *Journal of periodontology* 78: 185-93, 2007.
38. Raghoobar G, Schortinghuis J, Liem R, Ruben J, van der Wal J, Vissink A. Does platelet-rich plasma promote remodelling of autologous bone grafts used for augmentation of the maxillary sinus floor? *Clinical Oral Implants Research* 16: 349-56, 2005.
39. Ribony M, Polini F, Costa F, Politi M. Osteogenesis distraction and platelet-rich plasma for bone restoration of the severely atrophic mandible: preliminary results. *Journal of Oral & Maxillofacial Surgery* 60: 630-35, 2002.
40. Rodeo S, Hidaka C, Maher S. What's new in orthopaedic research. *Journal of Bone and Joint Surgery (American volume)* 87 (10): 2356-67, 2005.
41. Roukis T, Zgonis T, Tiernan B. Autologous platelet-rich plasma for wound and osseous healing: a review of the literature and commercially available products. *Advances in Therapy* 23 (2): 218-36, 2006.
42. Sammartino G, Tia M, Marenzi G, di Lauro A, D'Agostino E, Claudio P. Use of autologous platelet-rich plasma (PRP) in periodontal defect treatment after extraction of impacted mandibular third molars. *Journal of Oral & Maxillofacial Surgery* 63: 766-70, 2005.
43. Sanchez A, Sheridan P, Kupp L. Is platelet-rich plasma the perfect enhancement factor? A current review. *The International Journal of Oral & Maxillofacial Implants* 18: 93-103, 2003.
44. Schliephake H. Bone growth factors in maxillofacial skeletal reconstruction. *International Journal of Oral & Maxillofacial Surgery* 31: 469-84, 2002.
45. Schmidt A. Symphony™ PCS (Platelet Concentrate System) in tibial bone grafting: a report of three cases. In: Symphony, ed: Carolina Blood Management, Inc, 2003.
46. Tischler M. Platelet rich plasma. The use of autologous growth factors to enhance bone and soft tissue grafts. *New York State Dental Journal* 68 (3): 22-24, 2002.
47. Trawick R. Resolving a distal tibia non-union: a case study on the use of Growth Factors from Autologous Platelet Rich Plasma (PRP). In: Symphony, ed: Carolina Blood Management, Inc, Not stated.
48. Unknown. ReGen Lab: products for tissue repair. In: Lab R, ed: ReGen Lab
49. Watson T. Autologous Growth Factors combined with demineralised bone matrix used as an alternative to autograft for the treatment of recalcitrant nonunited fractures of long bones: a report of six cases: Carolina Blood Management, Inc, 2002.
50. Weiner B, Walker M. Efficacy of autologous growth factors in lumbar intertransverse fusions. *Spine* 28 (17): 1968-71, 2003.