Application of Concentrated Growth Factors (CGF) and Mphi Laser to Treat Defects in the Oral and Maxillofacial Region. A two - case report.

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ABSTRACT

The application of Concentrated Growth Factors (CGF) in oral and maxillofacial surgery (OMS) and, in general, in regenerative medicine is steadily increasing. The purpose of this study is to present a review and case reporting on the use of CGF for tissue regeneration in the oral maxillofacial region.

MATERIALS AND METHODS

Literature search was carried out using Medline search and manual search using the keywords: "concentrated", "growth factors", "tissue engineering", "regenerative medicine" and "blood". The review followed the method recommended by PRISMA and included clinical studies with adequate information. Papers with lack of data were excluded. Additionally, authors' experience on this topic was reported with the description of two relevant cases. In both patients, bony defects were filled with autologous fibrin rich CGF and synthetic alloplastic materials, and then treated with Mphi laser. Results: There were no published data on the combined use of Mphi laser and CG Fin clinical applications in the OMS region. Two case reports on the surgical regenerative management of oral lesions CGF and Mphi laser were described. Postoperative recovery was uneventful. Laser was effective in reducing postoperative pain, swelling, bleeding, speech impairment, analgesic use, trismus and wound healing. There was no difference in wound healing after one and 3 months. Conclusion: The innovative application of Concentrated Growth Factors (CGF) in combination with Mphi laser in Oral and Maxillofacial Region defectsproduced rapid improvement and minimized complications. The treatment is fairly simple and cost effective.

I. INTRODUCTION

Growth factors are molecules capable of facilitating several biological activities, such as cell proliferation, differentiation and repair. The use of concentrated growth factors (CGF) in dentistry is a relatively new concept. Since its inception, CGF have been used to enhance regeneration and healing for a variety of procedures, such implant placement, socket preservation, bony reconstruction and tissue regeneration [1]. The use of Multiwave-Locked System laser devices, such as Mphi laser [2], represents an innovative adjunctive therapy for the enhancement of wound healing after CGF treatment. A unique feature of MLS® Laser Therapy is the patented wave technology involving the use of two different and synchronised emissions,one with continuous/frequenced mode and 808 nm wavelength, the other with pulsed mode and 905 nm wavelength, that makes it one of the most efficient lasers for improving wound healing. Mphi laser has many therapeutic indications: sprains, muscle tears, tendinitis, brachial neuralgia, craniofacial pain, bursitis, lumbago, arthritis, articular pain, edema, hematoma. MLS® Laser Therapy produces its effects through anti-inflammatory and analgesic properties [3]. These effects are beneficial in the enhancement of wound healing and management of complications that can occur in surgical procedures. A previous study on the use of Mphi laser on treatment of craniofacial pain yielded promising results [5]. Research on laser therapy is widespread: up to the present time more than 28,200 articles are listed on PubMed, of these over 14,400 papers discussed pain associated with oral surgery. A systematic review on the application of LLLT on pain management [4] showed mixed results due to multiple inconsistencies in classification of wavelengths, outcome measures, study methods, recording techniques, degree of difficulty in oral surgery and duration of surgery.

Throughout typical wound healing, the fibrin substance is imperative in hemostasis, and forms the primary framework for the new extracellular matrix [6]. Fibrin permits the attachment of cells (for example, platelets and white blood cells - WBCs) and proteins to the bone tissue, fibroblasts and osteoblasts, endothelial cells, and smooth muscle cells. Also, keratinocytes attach to fibrin. Fibrin subsequently encourages wound healing by acting as conveying locations for the attachment of cytokine, growth factors and cell adhesion molecules [1]. In animal studies, fibrin accumulated in hypodermal tissue and revealed to be a significant factor in angiogenesis [7]. Additionally, numerous reports showed that wound healing is led by fibrin structure (i.e. concentration, quantity of division points, porosity and permeability). The fibrin substantial configurations are determined by several factors comprising clotting rate, Factor XIII concentration, thrombin, chloride ions, pH, etc [1]. Enhancing these environments is one of the goals of the CGF procedure. Pathological modifications of these fibrin regulators occur in some disorders, for example diabetes, and this undoubtedly tips to instabilities in wound healing. Accordingly, patients affected by these diseases are the subjects who benefit the most from the CGF procedure. Furthermore, not only the use of PRP and PRF have been reported to promote faster healing, but there are findings indicating that fibrin glue by itself can be utilised to improve wound healing [8]. With no anticoagulants used, the platelets start to be stimulated intuitively together with the coagulation cascade. The subsequent matrix/membrane complex has a high fibrin content and functions synergistically with growth factors [8]. In addition to CGF, employed as an autologous store of growth factors, synthetic alloplastic or xenoplastic materials can be used. The course of bone regeneration depends on various factors including, but not limited to, the size, the type and extent of the deficiency, the patient medical status, demographic variables and smoking status, provided that coagulum formation process is not impaired [8]. The typical healing time of oral bony defects is normally up to 12 months for small lesions, two years for medium-size lesions, and five years for larger cystic lesions [9]. Once the lesions were removed, bone defect was totally filled with blood clot/coagulum. A series of events occur after the preliminary blood clot formation, which include clot contraction and serum elimination, resulting in marginal serum-filled gaps amongst bony partition and coagulum exterior [9, 10]. This may notably affect angiogenesis and wound healing. Furthermore, the area created by the dental lesion elimination may originate a favourable environment for microbial proliferation and increase the possibility of infection. Thus, it is essential to stabilize the newly formed blood clot to facilitate wound healing [8]. Numerous methods of clot preservation have been documented including, but not limited to, guided bone regeneration and guided tissue regeneration using autografts, allografts and xenografts. Recently, the innovative application of autologous concentrated growth factors (CGF) has been well received [11]. Growth factors are naturally occurring substances, such as steroids or proteins, capable of promoting cellular growth, proliferation, healing, differentiation, and angiogenesis [12]. Blood growth factors are principally found in blood plasma and platelets. Of these, the most important are: platelet derived growth factor (PDGF), transforming growth factor (TGF), vascular endothelial growth factor (VEGF), epidermal growth factor (EGF) and insulin like growth factor 1 (IGF 1) [13, 14]. Historically, the initial production of platelet-rich plasma (PRP) [15] was presented in 1998 and the second, platelet rich fibrin (PRF) [16], in 2000. Blood obtained through patient venepuncture was used to generate fibrin-rich gel PRF [16–18]. CGF was originally, discovered by Sacco and co-workers [19]. CGF revealed a superior tensile strength, extra growth factor content, better viscosity and greater adhesive strength than PRF [20]. The usage of autologous fibrin provides many advantages including safety, no side effect, easy technique, cheap, and efficacy for the patients [20, 21].

II. MATERIALS AND METHODS

This study attempted to carry out literature review and cases report on the use of Mphi laser in conjunction with CGF applications in the oral and maxillofacial region. Literature search was carried out using Medline search and manual search using the keywords: "concentrated", "growth factors", "tissue engineering", "regenerative medicine" and "blood". The review followed the method recommended by PRISMA and included clinical studies with adequate information. Papers with lack of data were excluded. No publication was found on this topic. The authors presented their experience with the above mentioned technique by describing two cases on the application of Mphi laser in conjunction with CGF in the regeneration of bony patients with large cystic lesions in the upper and lower jaw, respectively. CGF was prepared (Figure 1) according to the procedure using patient's blood through venepuncture (total blood collected 40 ml), which was allocated into two red and two white sterilised 10 ml Vacutainer® tubes. These four tubes were instantly centrifuged with Medifuge MF 200 (Silfradent, Italy) following the manufacturer's recommendation. After spinning, sedimentation of the Vacutainer's content was allowed for 20 min until further processing. The top 2ml layer comprised the platelets poor plasma (PPP) containing serum, the next 2ml layer was platelets rich plasma (PRP), the third 0.7ml was the stem cells/white layer, while the fibrin-rich mass with CGF was in the central part of the tube constituting the CGF clot, the remaining 4ml and 1ml precipitated portions were red blood cells and sedimentary blood fragments, respectively. The final CGF mixture was collected by tipping off the top PPP and meticulously collecting the clot layers. Using a sterile Petri dish, the CGF clot

was detached from the red blood portion using scissors (12). Fabrications of CGF sticky bone and membrane were followed by the manufacturer's protocol. For sticky bone, Geistlich Bio-Oss® (bovine demineralized freeze dried bone, Geistlich Biomaterials) and Osteon (hydroxyapatite and calcium triphosphate, Genoss®) were used instead of calcium triphosphate. Digital radiographic and clinical assessments were performed at one, three and six month review. The patients were monitored and were asked to report post operative pain, swelling, bleeding, speech impairment, analgesic use, trismus and wound healing.

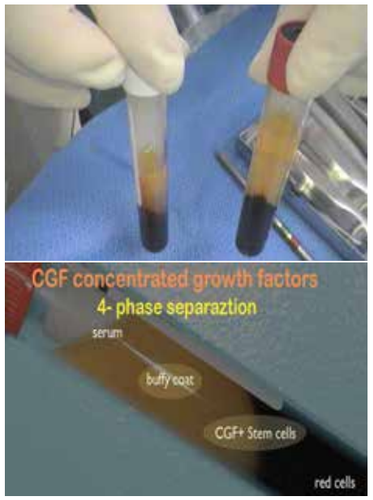

Figure 1: White and Red Vacutainer® tubes showed different colours and layers after spinning, used for CGF preparation of sticky bone and membrane (Adapted from Dr Ezio Gheno, 2015)

Ideally, there should be a control treatment site on a contra-lateral side to compare the effectiveness of Mphi laser and CGF treatment to the sham/control side. However, due to lack of suitable cases for a split mouth study, these two cases were treated as clinical audit

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III. RESULTS

The search found only six CGF related articles that, however, did not meet completely the inclusion criteria. These six articles illustrated the application of CGF in the oral and maxillofacial region (OMR) including: sinus lift, ridge augmentation, gingival recession, implantology, maxillofacial reconstruction.

The cases reported below showed good results, indicating that the innovative clinical application of CGF in OMR is promising. No post-operative complications associated with CGF procedures were reported in the two cases. Mphi laser was effective in reducing postoperative pain, swelling, bleeding, speech impairment, analgesic use, trismus and wound healing. Apparently, there was no difference in wound healing after one and three months.

CASE REPORT

Case 1

A female patient, 55 year old, was presented to a private specialist Oral Surgery clinic for a complaint of pain and swelling in the anterior lower jaw region. Her medical history was unremarkable. Clinical assessment indicated inflammation in the lower vestibule, tender and fluctuant to touching in the lower right lateral incisor and first premolar area.

As the involved teeth were splinted together in a fixed porcelain fused to metal bridge, no mobility was detected (Figure 2). After clinical assessment, cone beam computed tomography (CBCT) scans were acquired, displaying two well outlined round cystic defects (diameter ~ 3 cm), located in the right anterior area of the mandible (Figure 3).

Figure 2: Intra-oral photo of lower right lateral and first premolar splinted together by porcelain fused to metal bridge

and demonstrating a fair amount of bone loss at the lower right lateral incisor and first premolar, with intact lingual bony plate.

The surgical management involved total elimination of two cystic defects (lower right lateral incisor and second premolar), apicoectomy of the involved teeth and concealing the deficiency with CGF mixture of fibrin rich sticky bone and membrane. After CGF clot preparation, surgical process was carried out under local anaesthesia. Subsequently, a mucoperiosteal buccal flap was raised between lower left first incisor and lower right second premolar. An interchanging dulled and piercing division was used to detach and eradicate the two cystic lesions from bone. After enucleation/ cystectomy, apicoectomies of the involved teeth were performed. The resultant bony deficiencies were rebuilt by packing of CGF fibrin-rich clot sticky bone reinforced with Geistlich Bio-Oss® and Osteon, which totally covered the cystic defects (Figure 4). Ultimately, the flap was sutured in place with resorbable chromic sutures. At the end of the treatment, Mphi laser was applied at the surgical site following the manufacturer's protocol. The total CPW energy used was 0.637 Joules at an energy density of 1.27 J/cm^2, frequency of 1500 Hz, lasing time per application 0.04 second at 25% intensity. Two laser applications, at the apical and coronal half, were employed at each implant on buccal and lingual side. Post-operatively, the patient was given thorough oral hygienic instruction and dietary program. Prescription

Figure 3: Cone beam computed tomography (CBCT) and 3D reconstruction of patient's oral and maxillofacial region shows defects at the lower right anterior section of the mandible.

Figure 4: Outline of treatment for two dental cysts using CGF sticky bone and membrane.

of antibiotics (Amoxicillin/Clavulanic and metronidazole) and analgesics (combined paracetamol and ibuprofen) were given. Postoperative follow-ups were uneventful and were done at first day, first week, one month, three months and six months. No post-operative complications associated with CGF treatment were noted. Digital radiographic and clinical assessments were performed at one, three and six month review. The patient was monitored for postoperative pain, swelling, bleeding, speech impairment, analgesic use, trismus and wound healing. Mphi laser helped to reduce postoperative pain, swelling, bleeding, speech impairment, analgesic use, trismus and wound healing. There was no difference in wound healing after one and 3 months. Through the ensuing six months, an even and stable building of the deficiencies by freshly produced bone was noted.

Case 2

A healthy 81 years old lady with no significant medical history, presented to a specialist Oral Surgery Clinic for surgical management of perio-implantitis of her two existing implants in the upper first premolar and molar area. Though the patient did not complain of any particular troubles, digital imaging indicated areas of bone loss surrounding the coronal third of her two implants spreading bucco-lingually.

Clinical evaluation indicated slight gingival inflammation in the buccal aspect of the two implants though the sulcular mucosa appeared normal. A digital peri-apical imaging of the implants showed moderate bone loss in their coronal thirds (Figure 5). Treatment options for the defect encompass conservative and radical surgical management. Conservative treatment includes debridement followed either by traditional or CGF bone and tissue regeneration (GBR and GTR).

Radical treatment involved surgical removal of the two mentioned implants. The patient

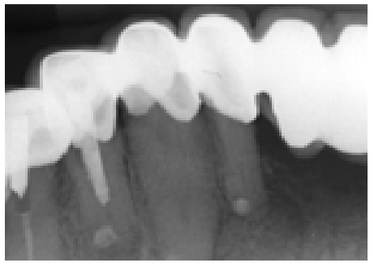

Figure 5: Post-operative peri-apical radiography showed cystic defects were filled CGF containing sticky bone and membranes.

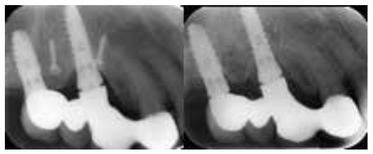

Figure 6: Left x-ray showed perio-implantitis defect at coronal third of upper first premolar and molar. Right x-ray illustrated defects after CGF treatment.

chose conservative CGF GBR and GTR.

This treatment comprised of raising a bucco-palatal muco-periosteal flap followed by a total removal of soft granulated peri-implantitis tissue, debridement of the infected bone, removal of two remaining GBR screws and smoothing of exposed implant titanium threads. The exposed implant surfaces were conditioned with CGF stem cells and PRP prior to application of CGF sticky bone and membrane to restore the entire peri-implantitis defects (Figure 6).

The CGF sticky bone and membrane were prepared instantaneously, prior to operation, as explained in case 1, using four 10 ml Vacutainer® tubes. Once the defects were completely filled with sticky bone and membrane, the flap was closed using chromic resorbable sutures then coated with PPP to create a fibrin seal to enhance wound healing (Figure 7). At the conclusion of the procedure, Mphi laser was used to irradiate the surgical wound following the manufacturer's protocol. The total CPW energy used was 0.637 Joules at an energy density of 1.27 J/cm^2, frequency of 1500 Hz, lasing time per application 0.04 second at 25% intensity. Two laser applications, at the apical and coronal half, were employed at each implant on buccal and lingual side. Similar prescription antibiotic regimen (Amoxicillin/ Clavulanic and metronidazole) and analgesics (combined paracetamol and ibuprofen) and post surgical oral hygiene instruction were given. Postoperative follow-up were uneventful and were carried at first day, first week, one month, three months and six months. The patient was observed and reported for post operative pain, swelling, bleeding, speech impairment, analgesic use, trismus and wound healing. No post-operative complications associated with CGF therapy were recorded.

Digital x-ray assessments were performed at one, three and six month review. Mphi laser helped to reduce postoperative pain, swelling, bleeding, speech impairment, analgesic use, trismus and wound healing. There was no difference in wound healing after one and 3 months.

Figure 7: Outline of treatment for peri-implantitis for upper right first premolar and first molar using CGF sticky bone and membrane.

DISCUSSION

Traditionally, regeneration of bony defects following eradicating big cystic defects in the oral and maxillofacial region, such as maxilla and mandible, may, at times, be coupled with complications. For example, shrinkage of blood clot, serum exudation, and development of lifeless gaps, as well as a risk of minor infection, considerably affect the regenerative courses of the jawbones.

The above issues have brought attention to the medical scientific community through research and publications [1,8]. Conventionally, it is not uncommon to have a total eradication of cystic defects and covering the finishing bony lesion with primary wound suturing. The main dilemma of the surgeon is to find the best way for bone defect reconstruction. According to the available literature [8], large bony defects are commonly filled and reconstructed with autotransplants obtained from the iliac ridge, ribs or donor sites in the oral cavity. Application of autotransplants enables primary wound healing, preservation of bone contours and fast regeneration. However, a drawback of this approach is the need for additional surgical procedure, highly specialized personnel, general anaesthesia and very high expenses [8,9]. Application of growth factors in guided bone regeneration procedure has been well-known for an extended period of time. This technique is applicable in implantology, specially due to its versatility in various augmentation techniques, and in unfavourable anatomic situations (horizontal and vertical augmentation, sinus lift etc.) [14]. CGF can be applied alone or mixed with bone autotransplants or other bone graft substitutes. The above-mentioned indications exemplify small bony defects that can be easily restored.

The good results obtained in the two clinical case reports indicated that the innovative clinical application of Mphi laser and CGF in OMR is promising, though the exact mechanism of action is not discussed as it is beyond the scope of this paper. The application of Mphi laser and CGF in the reconstruction of large cystic defects has not been reported yet. The presented cases are a pioneering attempt of reconstructing and restoring bone defects of the upper and lower jaws in combination with the use of synthetic alloplastic bone substitutes, secondary surgical procedures and chemical additives. The only method that is somewhat comparable with the presented cases is a lateral sinus lift procedure with filling dead spaces between the sinus mucosa and bony palate with pure CGF blocks [8, 20]. The published data addressing this topic suggested that newly formed bone of acceptable quality (density) and quantity was formed within 3–6 months, and had less postoperative complications. Moreover, the procedure is economically acceptable to the patient [20, 21].

Application of CGF sticky bone and membrane is one of the most up-to-date methods for reconstruction of bone defects in the OMR [8, 20]. Concentrated growth factors are applicable alone or mixed with bone graft material. The two cases presented in this article demonstrate the efficiency of CGF in significantly shortening bone-healing time, particularly in massive bone defects, reducing the incidence of postoperative complications, and enabling better restoration of surrounding periodontium. The method is relatively simple, with minimal risk of infection and allergic reaction, and economically feasible.

In an ideal scientific study, there should be a control group. However, the current cases were not suitable for a split mouth study and were treated as clinical audit. More research should be carried out, particularly, prospective split mouth study to confirm CGF and Mphi laser effectiveness and efficacy in tissue regeneration for the oral and maxillofacial region.

CONCLUSION

Majority of current regenerative technologies in OMR still faced with three main issues: compatibility, failure and cost. The use of autologous materials such as CGF may represent a potential solution to these dilemma. The combination with Mphi laser application may represent a valuable aid in managing post operative discomfort. Further research should be conducted, especially, prospective split mouth study to validate the effectiveness of the CGF and Mphi laser combined treatment in tissue regeneration for the oral and maxillofacial region.

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