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THE EFFECT OF HYALURONIC ACID ON PERIODONTAL ATTACHMENT GAIN DURING GUIDED BONE REGENERATION

ВЛИЈАНИЕТО НА ХИЈАЛУРОНСКАТА КИСЕЛИНА ВРЗ ПОЗИЦИЈАТА НА ПРИПОЈНИОТ ЕПИТЕЛ ВО ТЕК НА ВОДЕНАТА ТКИВНА РЕГЕНЕРАЦИЈА

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Abstract

Introduction: The management of periodontal defects has been an ongoing challenge in clinical periodontics. This is mainly a result of the fact that tissues, which comprise the periodontium, the periodontal ligament, the cementum and alveolar bone, represent three unique tissues in their own right. Thus, reconstruction of the periodontium is not just a simple matter of regenerating one tissue but involves at least three quite diverse and unique tissues.More recently, attention has been paid on regenerative and reconstructive therapies, rather on resective therapies. Among the many mediators used in periodontal regeneration ishyaluronic acid. In the field of dentistry, hyaluronic acid has shown anti-inflammatory and anti-bacterial effects in the treatment of periodontal diseases. The main aim of our investigation was to follow up the effect of hyaluronic acid on periodontal attachment. **Material and method:** 30 patients took part in the study. Patients were selected according to the following criteria: aged between 20 - 45, without anamnestic data for general disease,non-smokers, similar type of periodontal destruction, both in volume and type, on one contralateral side. Prior to the intervention, all patients were given advice on proper oral hygiene. Modified Widmann method was applied for all patients. BioOss beef boneapplied to each patient on one side (control group) and BioOss with 16% BDDE hyaluronic acid from Stylage-Vivacy Paris in a ratio of 2:1, was applied on thecontralateral side, enough until a thick, sticky bone ratio is obtained - (examined group). Results were monitored by CBCT (computed tomography in which X-rays are divergent to form a cone beam) and measurements were made before and after 12 (twelve) months. **Results:** The obtained data showedthat regenerative approach using hyaloss in combination with guided tissue regeneration (GTR) for the treatment of human infrabony defects resulted inbenefit in terms of Clinical attachment (CAL) gains, periodontal probing depth (PPD) reductions and radiographic defect fill, as well as linear bone growth (LBG), compared to GTR alone. **Conclusion:** Therefore, we can conclude that hyaluronic acid has beneficial effects on periodontal tissues. **Key words:** hyaluronic acid, bone regeneration, periodontal regeneration and reconstructive therapies attachment gain.

Апстракт

Вовед: Управувањето со пародонталните дефекти е тековен предизвик во клиничката пародонтологија. Ова главно е резултат на фактот дека ткивата што го сочинуваат пародонтот, пародонталниот лигамент, цементот и алвеоларната коска, претставуваат три уникатни ткива сами по себе. Така, реконструкцијата на пародонтот не е само едноставна работа за регенерирање на едно ткиво, туку вклучува најмалку три сосема различни и уникатни ткива. Во поново време, вниманието е фокусирано на регенеративните и реконструктивните терапии, наместо на ресективните терапии. Меѓу многуте медијатори кои се користат во пародонталната регенерација е хијалуронската киселина. Во областа на стоматологијата, хијалуронската киселина покажа антиинфламаторно и антибактериско дејство во лекувањето на пародонталните заболувања. Главната цел на нашето истражување беше да се следи ефектот на хијалуронската киселина врз пародонталната приврзаност. **Материјал и метод:** 30 пациенти учествуваа во студијата. Пациентите беа избрани според следните критериуми: на возраст меѓу 20 - 45 години, без анамнестички податоци за општа болест, непушачи, сличен тип на пародонтална деструкција и по волумен и по тип на едната и контралатералната страна. Пред интервенцијата на сите пациенти им беа дадени совети за правилна орална хигиена. За сите пациенти беше применета модифицираната метода на Видман. BioOss говедска коска нанесена на секој пациент на едната страна (контролна група) и BioOss со 16% BDDE хијалуронска киселина од Stylage-VivacyParis во сооднос 2:1, беше нанесена на контралатералната страна, во доволна количина додека не се добие густ леплив коскен сооднос - (испитана група). Резултатите беа следени со CBCT (компјутерска томографија во која X-зраците се дивергентни за да формираат конусен зрак) и мерењата беа направени пред и по 12 (дванаесет) месеци. **Резултати:** Добиените податоци покажаа дека регенеративниот пристап со употреба на хијалос во комбинација со водена регенерација на ткиво (GTR) за третман на дефекти на човечки инфрабонски резултираше со придобивки во однос на придобивките од клиничкото прикачување (CAL), намалувањето на длабочината на пародонталното испитување (PPD) и радиографскиот дефект, пополнување, како и линеарен раст на коските (LBG), во споредба со само GTR. **Заклучок:** Затоа можеме да заклучиме дека хијалуронската киселина има поволни ефекти врз пародонталните ткива. **Клучни зборови:** хијалуронска киселина, регенерација на коските, пародонтална регенерација и придобивка од реконструктивни терапии.

Introduction

In the field of dentistry in the last 50 years, scientists have been focused on a number of important requirements of periodontology, of its etiopathogenesis, but theywere mostly focused on periodontal regenerative possibilities and therapy.

Hyaluronic acid has been identified in all periodontal tissues in varying amounts and is more pronounced in non-mineralized tissues, such as the gingival and periodontal ligaments, compared with mineralized tissues such as cement and alveolar bone. In addition, due to the high levels of hyaluronic acid in the circulating blood serum, it is constantly present in the gingival blood flow fluid (GCF) which is a factor in serum overload[1].

Hyaluronic acid is an anionic glycosaminoglycanic acid widely distributed throughout connective, epithelial, and nerve tissues. It is unique among glycosaminoglycans in that it is non-sulfated and forms in the plasma membrane instead of in the Golgi apparatus. The human synovial hyaluronic acid averages about 7 million daltons per molecule, or about twenty thousand disaccharide monomers, while other sources mention 3-4 million daltons. One of the major components of the extracellular matrix, hyaluronic acid, contributes significantly to cell proliferation and migration, and may also be involved in the progression of some malignancies. The average 70 kg person has approximately 15 grams of hyaluronic acid in the body, of which one third is degraded and synthesized every day[2]. Hyaluronic acid is also a component of group A streptococcal extracellular capsule, and is believed to play an important role in virulence. Hyaluronic acid is one of the most well-known hygroscopic molecules known in nature. When hyaluronic acid (HA) is incorporated in aqueous solution, hydrogen bonding occurs between adjacent carboxyl and N-acetyl groups; this feature allows hyaluronic acid to maintain conformational stiffness and retain water. One gram of hyaluronic acid can bind up to 6 L of water. As a physical material, it has functions in spatial filling, lubrication, shock absorption, and protein exclusion[3]. The viscoelastic properties of the material can slow down the penetration of viruses and bacteria, a feature of particular interest in the treatment of periodontal diseases. Hyaluronic acid, as a viscoelastic substance, helps in periodontal regenerative procedures by maintaining spaces and protecting surfaces[3]. By recognizing its hygroscopic and viscoelastic nature, hyaluronic acid can affect cell function by modifying surrounding cellular and extracellular micro and macro media. Hyaluronic acid has many structural and physiological functions within tissues, including extracellular and cellular interactions, the interaction between the growth factor and the regulation of osmotic pressure, and tissue lubrication, which helps maintain the structural and homeostatic integrity of tissues[4]. Xenografts are proper alternatives for bone repair and regeneration because of their similarity to the human bone[5]. Although, the available amount of autograft material is always limited, one can obtain as much xenograft as desired. Due to its hydroxylapatite structure, natural bovine bone is potentially a much better graft material than a synthetic bone substitute. However, unresorbed graft remains of bovine bone have been observed in histological analyses even after three years[6]. Because xenografts are osteoconductive rather than osteoinductive, it is important to identify methods to improve their effectiveness in vivo[7].

Considering the various beneficial effects of hyaluronic acid, we focused our interest on the effects of hyaluronic acid in combination with bone graft and their effect on periodontal attachment gain during Guided Bone Regeneration (GBR).

Materials and method

30 patients took partin the study. Patients were selected according to the following criteria:

- aged between 20 - 45
- without anamnestic data for any general diseases
- non-smokers
- finding of both sides, main andcontralateral similar type of periodontal destruction, both in volume and type

All patients hadclinical examination before the intervention and the followingfeatureswere measured:

- depth of the periodontal pocket,
- loss of attachment,
- recession,
- gingival bleeding and gingival inflammation (according to Silnes Loë gradient)

Prior to the intervention, all patients were given advice on proper oral hygiene. The modified Widmann method was applied toall patients.Afterwards,BioOss beef bonewasapplied to each patient on one side which was used as control. BioOss with 16% BDDE hyaluronic acid from Stylage, Vivacy- Paris in a ratio of 2:1 until a thick, sticky bone ratio was obtained,wasapplied on the contralateral side of each patient - which data served as examined group.

In all patients, the results were monitored by CBCT (computed tomography in which X-rays are divergent to form a cone beam) and measurements were made before and after 12 (twelve) months.

Table 1. Values for attachment loss for the control group before and 12 months after surgery

	Lost attachment X	Stand. Deviation	St.dev.	t	p
before operation	5,26	2,862			
after operation 12 months	3,26	1,509	1,26	1,26	0,00000

Table 2. Values for attachment loss for the examined group before and 12 months after surgery

	Lost attachment X	Stand. Deviation	St.dev.	t	p
before operation	6,012	2,254			
after operation 12 months	3,395	1,724	2,91	4,39	0,000121

Table 3. Difference in values forattachment gain for the examined group and control group12 months after surgery

	Attachment gain	Stand. Dev.	St.dev.	t	p
control group	3,26	1,96			
examined group	3,93	1,724	2,989	-0,124	0,834

Results

Table 1. shows the values for attachment loss before and after 12 months postoperatively. Main value before surgery was 5,26 mm and 3,26mm after 12 months. High statistical significance of the differences for the values can be established.

Table 2. shows the values for the test group where hyaluronic acid was used. Main value before surgery was 6,12 mm and 3,395 mm after 12 months. High statistical significance forthe differences for the values can be established.

Table 3. shows attachment gain post operatively in the test and in the control group, a significant benefit of attachment gainand reduction of pocket depthcan be seen in both groups, yet there was no statistical significance of the differences. However, when comparing the numbers, the result in the examined group is 0.6 mm better.

Discussion

Today, hyaluronic acid is widely used in many branches of medicine with interesting potential applications in dentistry for the treatment of acute and chronic inflammatory disease. Data obtained from the present review of 20 clinical studies demonstrates that, due to its positive action on tissue repair and wound healing, topical administration of hyaluronic acid could play a role not only in postoperative dental surgery, but also in the treatment of patients affected by gingivitis and periodontitis, with a significant improvement in their quality of life. Further, laboratory-based research and large-scale randomized controlled clinical trials on a larger scale are advisable to confirm these promising results. From the perspective of current research, hyaluronic acid-based bone regenerative scaffolds are more biocompatible and bioactive with biomimetic strategies. As a matrix component, hyaluronic acid, especially sulfated HA (hyaluronic acid) may trigger cell behavior modulation via several signaling pathways, leading to faster and more desirable bone formation. Scaffolds and carriers based on HA are shaped into either rigid forms or colloids. As a rigid scaffold material, when incorporated with other materials, hyaluronic acid may alter the scaffold morphology and improve mineralization, making it more desirable and more functional for bone regeneration. Moreover, hyaluronic acid is chemically versatile, with its properties changed via simple chemical modification and crosslinking. The viscidity,

rheological properties, pH, and charge properties of hyaluronic acid can be modulated into states suitable for gelation or delivery. This leads us to the carrier hyaluronic acid. Either by mixing, or by chemically or electrostatically encapsulating a diverse range of growth factors, drugs, mineralized components, or cells in hyaluronic acid-based carriers, bone formation can be markedly enhanced and accelerated. New bone formation could more closely resemble that of the original tissue. Some strategies can also perform superbly in osseointegration for implantation. Hyaluronic acid-based hydrogels and micro particles can covalently bind to metal implant surfaces and release bioactive components, resulting in better osteogenesis and osseointegration. However, the specific mechanisms behind the effects of hyaluronic acid on osteogenesis still require proper investigation-controlled delivery as well as biomimetic scaffold and carrier designs, not just hyaluronic acid-based forms.

More recently, cross-linked hyaluronic acid (HA) products were used as gel barriers to cover the osseous defects around the implants and implant recipient sites, thereby promoting guided bone regeneration(GBR)[8]. Claar performed a lateral coverage of the augmentation followed by use of cross-linked hyaluronic acid in gel form, which was developed especially for guided bone regeneration[9]. The principles of GBR applications[10,11] are as follows: - Cell exclusion: creating a barrier to prevent forming fibrous connective tissue by epithelial cells. - Tenting: new wound space beneath the membrane must be regenerated solely around soft tissues so that high quality of new tissue can be gained. - Scaffolding: at first, a fibrin clot is seen in this space which is a scaffold for progenitor cells. Adjacent hard tissues serve as storage for stem cells. - Stabilization: to gain successful healing, the defective area must be protected from environmental effects such as flap movement, bacterial invasion, exposure of region, etc. by fixing the membrane into position.

The findings of our investigation-clinical study indicate that the use of HA + Bioss, as a regenerative material, was found to be effective in improving the clinical parameters compared to bioss alone in guided tissue regeneration. After 12 months postsurgery, a greater amount of mean Clinical attachment gain (0.6mm) was observed in the HA with Bioss group when compared to the Bioss groupalone. The results obtained in the present study were compared with other studies reported on the use of esterified HA. CAL following regenerative therapy is the single most commonly used clinical outcome variable. Ballini et al.[12] reported the mean gain of 2.6 mm CAL following application of esterified HA in combination with autologous bone in treatment of infrabony defects. However, Vanden Bogaerde[8] reported a mean CAL gain of 3.3 mm at 12 months follow-up. The greater mean CAL gain in their study could be explained by the differences in initial PD. The clinical studies have demonstrated that the CAL gain following regenerative periodontal therapy is strongly dependent on the initial PD, the greater the initial depth, thehigher is the CAL gain[13].

In ourpresent study, significantly greater mean CAL gain of 0.6 mm observed in hyaluronic acid + GTR investigation group in comparison with Bioss andGTR alone group, could be related to molecular characteristics of hyaluronic acid, since HA is known to stimulate cell migration, cell proliferation and also act as a carrier for other molecules, such as BMPs-2.14,15. From clinical standpoint, it was more significant to observe that 100% of sites treatedwith hyaluronic acidin combination with GTR experienced CAL gain of more than 2,91 mm, while only 8.3% of sites treated with GTR alone showed CAL gain of nomore than 2 mm.

Reduction of pocket depth to limit the risk of local reinfection is a primary goal of periodontal therapy. Shallow pockets have a strong, negative predictive value for future disease progression while deep pockets in treated areas are risk indicators for periodontal disease progression. In our present study, pocket depth reductions were significantly greater in both test and control groups. Engström et al.[16] reported mean PPD reduction of 4.1 mm when hyaloss was used in combination with bioabsorbable polylactic acid (PLA) barrier. However, Vanden Bogaerde[5] reported mean PPD reduction of 5.8 mm (range: 0–10 mm) at 12 months following an application of HA. Greater reduction of PPD reported by Vanden Bogaerde[8] could be related to the inclusion of the initial probing pocket of variable depth, which may have possiblyinfluenced the treatment outcome. During the 12 months period, the infrabony lesions in this study responded well to hyaluronic acid combined with GTR treatment with regards to reduction in radiographic DD. It is the experience of the investigators that the most accurate means of determining osseous defect response (crestal changes as well as within the defect) is by direct visualization at re-entry surgery, but a major disadvantage of re-entry procedure is the need for a second surgical procedure to visualize the osseous defect[17]. To overcome this difficulty, radiographic monitoring of alveolar bone changes has been utilized with various degrees of success[18]. Radiographic bone measurement is a non-invasive, painless alternative to direct bone measurement. Therefore, in ourpresent study, radiographic monitoring of alveolar bone changes was carried out as end-point variable. The most reliable outcome variable for assessing periodontal regeneration is human histology. Due to ethical considerations and patient management limitations, no histological evidence was obtained to establish the proof of periodontal regeneration. The importance of wound

stability for bone and periodontal regeneration has been reported. Based on the histological evidence from human material, it may be assumed that the clinical improvements following esterified HA treatment may represent, at least to some extent, a real periodontal regeneration characterized by the increase of osteoblastic activity by stimulating differentiation and migration of mesenchymal cells[19]. Moreover, the physiochemical properties of HA help keep the growth factors responsible for tissue repair in situ[20].

Conclusion

The aim of our study was to evaluate the use of hyaluronic acid in periodontal surgery and its possibly beneficial effects on periodontal tissues and periodontal regeneration.

Although there was no statistical significance of attachment gain there was clinical attachment gain where hyaluronic acid was used.Therefore, we can recommend the use of hyaluronic acid for periodontal regeneration.

The results of this research are expected to contribute to the knowledge of the impact of hyaluronic acid on periodontal regeneration and its application in the daily life of periodontology.

The obtained research results, together with literature data, give us relevant knowledge for optimal scientifically supported planning and realization for successful periodontal treatment. The scientific contribution of this research is the optimism that the scientific findings from this research will inciteinterest and need for new research regarding this issue.

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