Implant surgery: treatment with a fluid gel compound with hyaluronic acid and piperacillin plus tazobactam

Aim of the Work
Evaluation of applicability, clinical benefits and tolerability of a biomaterial as an organic scaffold with hyaluronic acid and piperacillin + tazobactam used alone or in combination with bone allograft and resorbable collagen membrane in the restoration of bone defects as well as in prevention and treatment of peri-implant infections.

Materials and Methods
A group of 43 patients with peri-implantitis and peri-implant bone defects or requiring sinus lift and extractive surgery were treated using the product in addition to standard procedures, the product was also used to wash sockets and applied on the implant before its placement in order to prevent early infections.

Results and Conclusions
The results of this preliminary study on selected patients, showed a good applicability of the product in surgical cases, with higher benefits and an excellent clinical tolerability. The biomaterial helped the processes of tissue repair creating a favourable environment for healing through the prevention of bacterial infections, owing to the presence of piperacillin and tazobactam, an antibiotic with broad spectrum activity against Gram + and Gram−.
INTRODUCTION

The aim of this study is to verify whether the association of high molecular weight hyaluronic acid and piperacillin + tazobactam may find useful application in bone regenerative therapy. This possibility is suggested by the protective action of hyaluronic acid and its effectiveness in promoting tissue restoration as well as its anti-inflammatory activity in a broader sense (antioxidant properties, reduced production of eicosanoids, etc.) together with its capacity to inhibit the adhesion and growth of bacteria. The latter phenomenon is associated with the antibacterial action of piperacillin and tazobactam. The antibacterial action of piperacillin + tazobactam together with the anti-adhesive properties, with respect to bacteria, of hyaluronic acid can reduce the formation of bacterial biofilm, which is so important for implantation. This product, mixed with bone of synthetic/heterologous/bone bank origin, could therefore be used with benefit for oral surgery.

Several publications have recently reported that a coating of hyaluronic acid on various medical devices has an anti-adhesive capacity with respect to cells and bacteria (1-3, 4-7).

Earlier studies suggested that hyaluronic acid can prevent bacterial adhesion to implants, thus exerting a bacteriostatic effect. Many of the biological processes mediated by hyaluronic acid are essential for tissue restoration and wound healing and regenerating tissues are particularly rich in this acid (8). Hyaluronic acid contributes to a wide range of cell functions that are essential for tissue restoration: inflammation response (9-11), cell migration (12-15), cell proliferation (16) and extracellular matrix organisation (17-21). Hyaluronic acid also plays an important role in angiogenic processes.

More recently, researchers have begun to study the effects of hyaluronic acid on the healing processes of post-extractive wounds (22) and wounds caused by the application of implants (23). In this regard, it has been observed that exposure to high molecular weight hyaluronic acid (900-2300 kDa), in appropriate dosages (0.5, 1.0 and 2.0 mg/ml), on osteoblast cultures (24) increases cell proliferation to a statistically significant extent. This study also evaluated, with positive results, the effect of adding hyaluronic acid on osteoblast differentiation and therefore bone formation. These results have led the authors to conclude that high molecular weight hyaluronic acid, with its stimulatory effects on osteoblasts, can increase the osteogenic and osteoconductive properties of implants and bone graft substitutes.

Further confirmation of the possible effectiveness of high molecular weight hyaluronic acid in oral pathologies emerges from a split-mouth study conducted on rabbits after extraction of the first upper and lower molars, in which it is noted that exposure to a high molecular weight hyaluronic acid gel promotes the post-extractive healing of the alveolus in comparison with a simple suture in the control sites (25). The study conducted over time, to be exact at 3, 7, 13, 20 and 30 days after the extraction, shows that hyaluronic acid improves all the parameters considered and, more precisely, the total alveolar area, the area of bone neoformation inside the alveoli, and the percentage of the alveolus recolonised by the bone tissue. This appears to depend on the capacity of hyaluronic acid to promote and accelerate the replacement of the clot with granulation tissue. This is because the hydrophilicity of hyaluronic acid makes the fibrin clot much softer and easier to colonise by the cells participating in the construction of the tissue being formed (13, 26). Further confirmation comes from a study on rats undertaken to investigate the possibility that hyaluronic acid may enhance the early differentiation of granulation tissue in osteogenic mesenchymal tissue. The authors, using histological techniques, studied the effects of a gel based on hyaluronic acid...
on bone neoformation without an implant in surgical wounds induced in the nasal sinus cavity and the calvaria of rats (27). The results confirmed the enhancement of osteogenic activity in wounds treated with hyaluronic acid. This finding confirms and extends the data obtained in other sites, such as the femur and the tibia. In particular, the introduction of hyaluronic acid in experimentally induced cavities in the femur of rats produces, after just four days from the application, evident signs of intense osteoinductive activity, contrary to what is observed in control sites without hyaluronic acid (28). Similar results are obtained on the tibia of rabbits where, in the cavities exposed to hyaluronic acid, better healing can be observed at all the time intervals studied (20, 30 and 40 days) in comparison with control sites (cavities filled with cancellous bone grafts only) (29).

The above data show that exposure to high molecular weight hyaluronic acid improves wound healing processes regardless of the application site.

Clinical evidence has demonstrated an acceleration of bone deposition and its remodelling in post-extraction sites treated with high molecular weight hyaluronic acid associated with autologous bone in comparison with the control sites treated with autologous bone only. The result is faster bone regeneration (30). In another study conducted on humans, high molecular weight hyaluronic acid was used in conjunction with autologous + heterologous bone and resorbable membrane for vertical regeneration of the alveolar ridge to allow subsequent insertion of the implant. Based on the results of this study, it has been possible to measure, through a biopsy, a vertical increase of 4 mm of bone tissue after 12 months (31). Further clinical evidence on the use of a product based on high molecular weight hyaluronic acid has demonstrated fast reossification of post-extraction sites, cavities secondary to complex extractions of included teeth, cystic cavities, insertion of implants, etc. (assessed on the basis of radiological evidence after 2 months). This is probably secondary to rapid neocollagenogenesis (32).

Finally, a recent study, which evaluated the correction of intrabone defects with hyaluronic acid in conjunction with autologous bone in 9 patients, has shown, in defects with an average depth of 8.3 mm, a gain of clinical attachment of 2.6 mm, with stability of the individual teeth, even after 9 months of treatment (33). As mentioned above, piperacillin has a broad spectrum of in-vitro activity, but has demonstrated an even broader spectrum of activity and, in some cases, has proved to be even more powerful against Gram- organisms than other antibiotics belonging to the penicilligoupe (34). Piperacillin with a concentration of 8 mg/l inhibits 90% of all anaerobic bacteria (34). Some studies have evaluated the activity of piperacillin in infections of the mouth, in particular in those associated with surgical treatment of the oral cavity. Post-surgical infections of the oral cavity have responded to treatment with piperacillin administered systemically (1-4 g/day for a period of 12 days) with excellent results in an average of 74% of all cases (34-38). The bactericidal activity of piperacillin against the characteristic strains of the above infections was found to be excellent or good in almost all cases (34).

Postoperative prophylactic therapy with piperacillin for the prevention of surgical infections of the oral cavity was found to be effective in all the patients studied (36, 38). The antimicrobial activity of piperacillin is enhanced by its association with tazobactam (39) against both Gram- and Gram+ anaerobic bacteria. In particular, the association of tazobactam + piperacillin produces an antimicrobial agent which is also active against bacteria that produce β-lactamase. As tazobactam is an inhibitor of bacterial β-lactamase, it extends the antibacterial activity of piperacillin also to those bacteria that are resistant to penicillins and cephalosporins because they produce β-lactamase (penicillinase or cephalosporinase).

**MATERIALS AND METHODS**

The product is a fluid gel and its components are 0.5% high molecular weight sodium hyaluronate (1.5·10⁶ Da) and sodium piperacillin + sodium tazobactam. The product is sterile and is packaged in glass bottles. In addition, sterile disposable syringes with a perforating needle were used for preparing the gel and blunt tip needles (replacing the perforating needles) for application directly on the surgical site or first on the heterologous bone and the membrane before in-site placement. The study has allowed an evaluation of the application characteristics of the product in selected surgical situations, as well as its clinical benefits and possible side effects. A total of 43 patients aged between 35 and 78 years, consisting of 19 men...
and 24 women, were treated. Specifically, 6 for surgical treatment of peri-implantitis, 6 for treatment of peri-implant bone defects (during the placement phase for immediate implantation), 14 in extraction surgery for delayed implantation placement with a bone graft in the alveolus, 3 for the large lift of the maxillary sinus, 1 for bone defects caused by enucleation of mandibular cysts, and 13 during implantation surgery with washing of the site and application of the solution on an implant before its insertion (for prophylaxis of precocious infections). At least one month before surgery the patients received professional oral hygiene treatment, with demonstrated adequate plaque control.

All patients took systemic antibiotics for coverage against post-surgical infections.

The patients started taking the antibiotics the evening before surgery and continued for the next 6 months. Amoxicillin + clavulanic acid (825 mg + 175 mg) or, if the patient is allergic to penicillins, clindamycin (600 mg), was administered every 12 hours. The patients also used a 0.2% chlorhexidine-based mouthwash twice a day, starting 2 days before surgery and continuing for the next 8 days. Patients with pockets > 6 mm and bone loss > 4 mm were included in the study for the treatment of peri-implantitis. Implants with > 80% bone loss, pain or mobility were excluded. The product being studied was used alone or in association with autologous and heterologous bone (Bioss, Geistlich) and resorbable collagen membrane (BioGide, Geistlich) for the surgical treatment of peri-implantitis. In particular, after local anaesthesia, removal of any prostheses, incision and dissection of the full thickness of the flap with its extension, after careful bone curettage and removal of any inflammatory tissue, to the healthy tissue beyond the infected area, a 6-step implant decontamination protocol was applied:

- milling of the implant surface with a sterile cutter;
- application of a 37% orthophosphoric acid-based gel around the implant left in situ for approximately 2 minutes, followed by aspiration of the gel using a sterile cannula and washing with a sterile physiological solution;
- application of a compress consisting of strips of sterile gauze soaked with piperacillin + tazobactam-based fluid gel around the implant, in the defect and under the mucoperiostal flap, left in situ for 5 minutes;
- graft of heterologous bone rehydrated with the piperacillin + tazobactam-based solution;
- coating of the bone graft with resorbable collagen membrane rehydrated with the piperacillin + tazobactam-based solution;
- suturing of the flap after inserting the screw cap.

The hyaluronic acid and piperacillin + tazobactam-based fluid gel was used for the regenerative treatment of the peri-implant bone defects of the alveolar ridge and in extractive surgery (in particular for delayed implantation placement), in sinus floor lifting (side window or crestal access technique), in bone defects resulting from enucleation of mandibular cysts. In particular, the therapeutic protocol was applied in 3 steps:

- mixing of the piperacillin + tazobactam-based fluid gel with bone particulate until a homogeneous mixture is obtained and removal of the excess solution with sterile gauze;
- application of the product, modelled using a suitable instrument, in the bone defect;
- coating of the bone graft with resorbable collagen membrane rehydrated with the piperacillin + tazobactam-based solution;

Finally, the product being studied was used during the implant insertion surgery for washing the site and was applied on the implant itself before insertion (as prophylaxis of precocious infections). In particular, the implant site was washed by placing the blunt needle tip near the base of the site and injecting the product until the solution flowed out of the top edge. A few drops of the product were then dripped onto the implant immediately prior to its insertion into the site to ensure that the entire surface was wet.

The evaluation criteria were based on the recording of signs and symptoms and radiographic assessments.

Evaluations based on the physical examination of the surgical site (after 8 days, during the stitch removal session, and then after 3 and 6 months) were made on the basis of changes in the signs and symptoms of the soft tissues affected by the flap and the state of the suture, such as oedema, swelling, redness, oozing, pain and any lymphatic gland involvement. In particular, for peri-implantitis, the presence of suppuration, bleeding on probing (BOP), pocket depth (PD), and radiographic evidence of bone loss were assessed. BOP was recorded using a dichotomous index for presence or absence, after waiting for 15 seconds following a
In addition, radiographic assessments were made after 3-6 months to assess healing and any changes in the peri-implant bone tissue. The radiographs were analysed using a method that involved the digitising and standardising of measurements (41).

For all patients, the tolerability of the product being studied was then assessed, and the occurrence of any side effects, both local and systemic, was recorded.

The authors declare that this study as presented was conducted in accordance with the ethical standards laid down in the Declaration of Helsinki and that informed consent was obtained from all participants prior to their enrolment in the study.

**RESULTS**

The results of clinical assessment of signs and symptoms after 8 days were excellent in 51% of patients, good in 46% and poor in 3%. After 3-6 months, the results of clinical assessment were excellent in all the patients treated (Tab. 1).

In the 37 patients belonging to the 5 groups listed in Table 1, no local or systemic side effects were recorded, with the exception of 4 patients (3 in the extractions group and 1 in the peri-implant defects group) who reported a “bad taste” on the first day after surgery.

Table 2 summarises the pre- and post-treatment data for the three groups of patients on whom at least one implant was inserted.

The group suffering from severe peri-implantitis consisted of 6 patients with 6 implants. Table 3 summarises the pre- and post-treatment changes in soft tissues, and Table 4 the radiographic variations in bone levels. The results for the 6 implants with severe peri-implantitis were a gain of bone tissue around the implant, which varied from 50 to 80% (Fig. 1). No local or systemic side effects were detected.

**DISCUSSION**

The results achieved by applying the treatment protocol described in this study for the prevention and treatment of peri-implantitis

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**tab. 1** Signs and symptoms of soft tissues affected by the flap and state of the suture.

<table>
<thead>
<tr>
<th>Days</th>
<th>Per-impl. def.</th>
<th>Extraction</th>
<th>Sinus lift</th>
<th>Cysts</th>
<th>Insertion of implants</th>
</tr>
</thead>
<tbody>
<tr>
<td>8 days</td>
<td>+++</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>+++</td>
</tr>
<tr>
<td>3 months</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
</tr>
<tr>
<td>6 months</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
</tr>
</tbody>
</table>

Complete remission of signs and symptoms of inflammation +++ (excellent); remission of symptoms with reduction of signs ++ (good) partial reduction of symptoms and signs + (poor); little or no reduction of signs and symptoms - (nil)

**tab. 2** Changes in soft and hard tissues from baseline to follow-up.

<table>
<thead>
<tr>
<th>Group 1 Site washing</th>
<th>Group 2 Peri-implant defects</th>
<th>Group 3 Sinus lift</th>
<th>Sinus lift</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of subjects</td>
<td>13</td>
<td>6</td>
<td>3</td>
</tr>
<tr>
<td>No. of implants</td>
<td>16</td>
<td>6</td>
<td>4</td>
</tr>
<tr>
<td>No. of sites with pre-Rx: defects over time</td>
<td>0</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Rx: defects over time</td>
<td>0</td>
<td>2</td>
<td>0</td>
</tr>
</tbody>
</table>

**tab. 3** Patients with peri-implantitis: changes in soft tissues from baseline to follow-up (6 months).

<table>
<thead>
<tr>
<th></th>
<th>No. of subjects</th>
<th>No. of implants</th>
<th>Pre-treatment oedema</th>
<th>Pre-treatment BOP</th>
<th>Pre-treatment suppuration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bone loss in mm (Rx), pre-treatment</td>
<td>6</td>
<td>6</td>
<td>6</td>
<td>6.46</td>
<td>0</td>
</tr>
<tr>
<td>Bone gain in mm (Rx), post-treatment</td>
<td>6</td>
<td>6</td>
<td>0</td>
<td>85</td>
<td>0</td>
</tr>
</tbody>
</table>

**tab. 4** Patients with peri-implantitis: radiographic changes in bone levels from baseline to follow-up (6 months).

The results of clinical assessment of signs and symptoms after 8 days were excellent in 51% of patients, good in 46% and poor in 3%. After 3-6 months, the results of clinical assessment were excellent in all the patients treated (Tab. 1).
are encouraging. Research to evaluate a non-surgical approach has shown that this treatment methodology does not give predictable results.

In this study, the level of bone gain, based on radiographs, was found to be on average 3.85 mm. This bone gain remained stable until the follow-up examination after 10 months (Fig. 1).

A recent publication has described a method for the decontamination of the implant to remove bacterial plaque mechanically, by using abrasive bicarbonate powder with air, combined with local antibiotic treatment (tetracycline) (41). The idea of decontaminating the implant surface can also be applied to a surgical field, such as the mouth, and this procedure is possibly more important than the treatment received by patients with peri-implantitis. This underlines the importance of a surgical strategy to facilitate the direct viewing of the implant. In fact, any method of decontamination of the implant surface that does not allow a complete view of the surface may prove to be ineffective. So far, other methods of decontamination of the implant surface have not been shown...

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Fig. 1
Radiographs of bone levels before and after treatment (10 months) showing reduction of the defect.
A: time 0;
B: after 10 months;
C: insertion of the implant;
D: time 0;
E: after 10 months.
a significant improvement in the results of the treatment of peri-
implantitis, possibly because of the impossibility of viewing the implant
surface (42-45).
In the authors’ opinion, the use of hyaluronic acid associated with
the local antibiotic is extremely important. In fact, this biomaterial
has been shown to prevent bacterial adhesion to implants (5-7).
Moreover, many of the biological processes mediated by hyaluronic
acid are essential for tissue restoration and wound healing and
regenerating tissues are known to be particularly rich in this acid (8).
Hyaluronic acid contributes to a wide range of cell functions that are
essential for tissue restoration: inflammation response (9-11),
and extracellular matrix organisation (17-21). Moreover, hyaluronic acid
plays an important role in angiogenic processes and has
demonstrated osteogenic and osteoconductive activity, which is its
most important characteristic (28-30).
The decision to use, in the decontamination protocol, a bone
substitute and a collagen membrane was based on a study that reported 3
years of stability of the regenerated bone in cases where there were
lesions related to peri-implantitis (46).
The procedures and materials contained in this study should now
be clinically evaluated in a randomised controlled study on a
larger number of patients and with a longer follow-up period.

CONCLUSIONS

The application of a protocol for the decontamination of the implant
surface that includes, in addition to the
aggressive mechanical removal of the biofilm present (by milling), a
local antibiotic treatment (with pipercillin + tazobactam) should be
considered in relation to the clinical results achieved, also keeping in
mind that the alternative of removing an implant with peri-
implantitis exposes the patient to the risks of bone damage and possible
damage to the nervous tissue and also to individual teeth.
The encouraging results of this study, although observed for
different indications and in a limited number of cases, support the
applicability of local antibiotic treatment combined with hyaluronic
acid in these selected cases of implant surgery.

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