

# Use of Subepithelial Connective Tissue Graft as a Biological Barrier: A Human Clinical and Histologic Case Report

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The aim of the present study was to develop a method to study the healing process after gingival grafting and to observe the histologic results after use of the modified edentulous ridge expansion technique. A 47-year-old nonsmoking woman with a noncontributory past medical history affected by edentulism associated with a horizontal alveolar ridge defect was referred to the authors for surgical correction of the deficit to improve implant support and the final esthetics of an implant-borne prosthesis. At the 4-month follow-up visit, a biopsy was performed by a punch technique in the same sites of healing abutment connection. The tissue was elevated from the attached gingival. Clinically, the grafted tissues seemed to be attached to the bone surfaces. The histologic findings revealed dense grafted tissues, providing long-term stability to the area. No ligament or bone, characteristic for periodontal regeneration, were observed. The presence of thick attached keratinized tissue around implants may constitute a protective factor against marginal inflammation or trauma.

**Key Words:** *connective tissue, epithelium, osteotome technique, endosseous dental implantation, biopsy*

## INTRODUCTION

The modified edentulous ridge expansion (MERE) technique corrects the atrophic ridge with a reduced number of surgical procedures and a reduced healing time. This technique uses connective tissue graft as a biological barrier to cover an immediate implant for improved hard and soft tissue regeneration. This approach restores proper placement and continuity to the mucogingival junction, increases the quantity of keratinized tissue, and deepens the fornix.

To our knowledge, no studies have obtained the

biopsies of grafted tissue from humans in vivo. The aim of the present study was to develop a method to study the healing process after gingival grafting and to observe the histologic results after use of the MERE technique.

## CASE REPORT

### Subject

A 47-year-old nonsmoking woman with a noncontributory past medical history affected by edentulism associated with a horizontal alveolar ridge defect was referred to the authors for surgical correction of the deficit to improve implant support and the final esthetics of an implant-borne prosthesis. Patient exclusion criteria were an extremely atrophic ridge with no interposition of cancellous

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DOI: 10.1563/AAID-JOI-D-11-00109



**FIGURES 1–3.** **FIGURE 1.** An autogenous connective tissue graft was layered over the bony wound. The connective tissue graft, about 1.5 mm thick and 27 mm wide, was harvested from the palate. **FIGURE 2.** Integration of the grafted tissue in the surrounding tissues was recorded. **FIGURE 3.** The healing donor site was recorded (the donor site was selected from the palate).

bone between the buccal and palatal plates and concomitant vertical defect.

### MATERIALS AND METHODS

Details regarding the MERE technique were recently described.<sup>1</sup> In brief, a full-thickness flap was dissected only in the alveolar crest. A sagittal osteotomy was outlined in the bone, and vertical bone-releasing osteotomies were also carried out mesially and distally 2 mm away from the adjacent teeth. The final implant site was prepared to the final depth with the osteotome technique. Tapered internal implants with a laser microgrooved coronal design (Biohorizons, Birmingham, Ala) were then tapped into position.

An autogenous connective tissue graft was layered over the bony wound and used as a biological barrier for better hard and soft tissue regeneration. The connective tissue graft was harvested from the palate. The donor site was selected from the palate, 2 mm below the gingival crestal margin. The connective tissue was placed over the implants as a biological barrier and inserted under the vestibular and palatal residual keratinized mucosa. Sutures were placed from the vestibular to the palatal site, stabilizing the connective tissue graft. A connective tissue graft covering the bony wound was used to augment the keratinized mucosa and avoid bone graft infection. Periosteal sutures, using 4/0 Trofilorc sutures (LorcaMarín, SA, Murcia, Spain), were used to intentionally position the flap buccally and palatally for healing by secondary intention.

At the 4-month follow-up visit, a biopsy was performed by a punch technique in the same sites of healing abutment connection. The tissue was elevated from the attached gingival. The biopsy specimen was fixed in 10% neutral buffered formalin solution at room temperature for 1 day.

After fixation, the biopsy was processed in Leica ASP 300S (Leica, Nussloch, Germany) tissue processor (formalin 1 for 30 minutes, formalin 2 for 30 minutes, ethanol 70% for 1 hour, ethanol 80% for 1.5 hours, ethanol 96% for 1.5 hours, ethanol absolute for 1 hour, ethanol absolute for 1.5 hours, ethanol absolute for 1.5 hours, xylene 1 for 1.5 hours, xylene 2 for 1.5 hours, paraffin 1 for 1 hour, paraffin 2 for 2 hours, paraffin 3 for 3 hours). The sample was embedded in paraffin, and sections 4  $\mu$ m thick were obtained from the specimen with Leica RM2125RT Microtome (Leica). The sections were stained with hematoxylin and eosin in a Leica Autostainer ST5020 (Leica).

### CLINICAL OBSERVATIONS

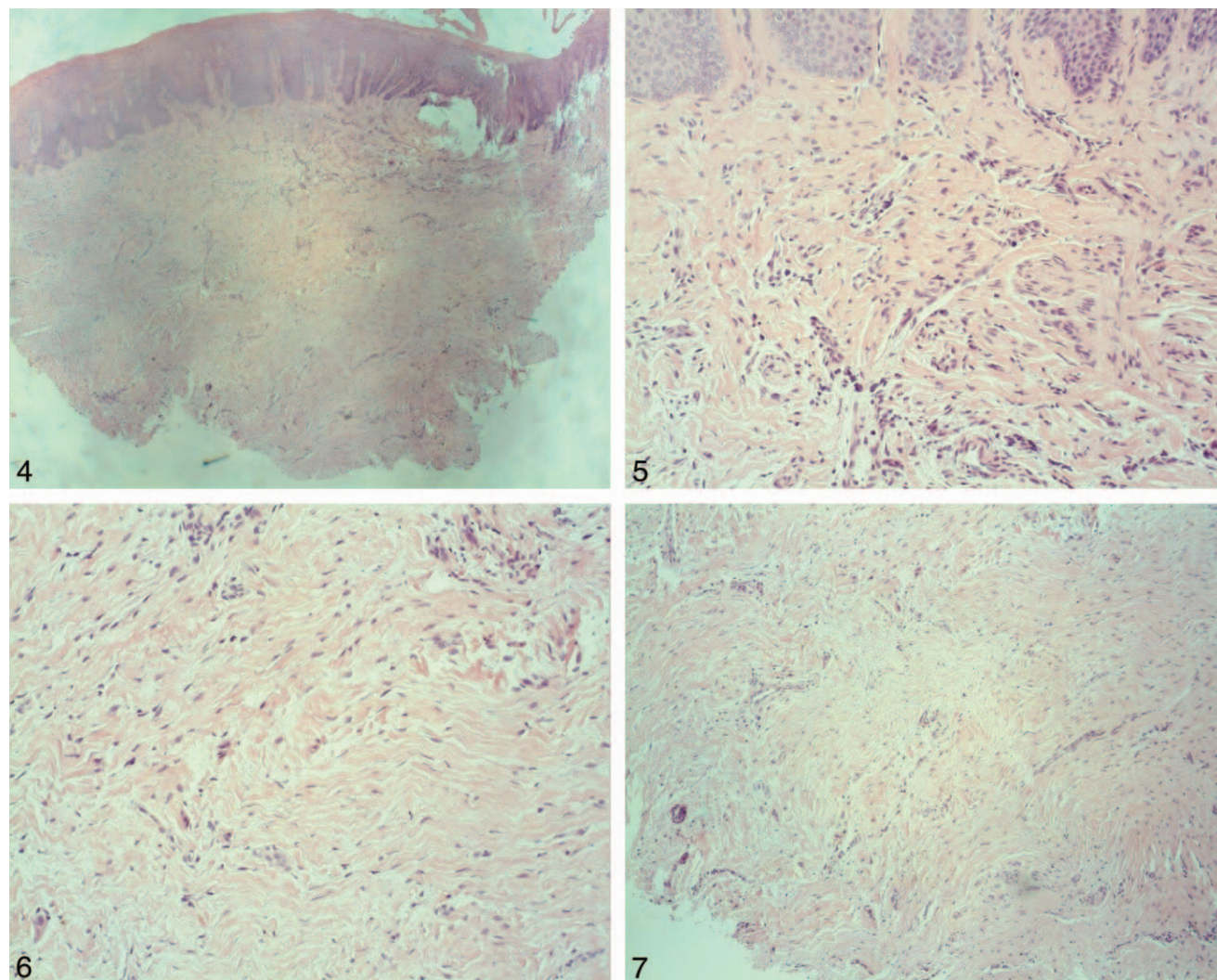
Wound healing was uneventful. The integration of the grafted tissue in the surrounding tissues and healing donor site were recorded (Figures 1 through 3).

### HISTOLOGIC OBSERVATIONS

The epithelium was slightly thickened (1.5 mm) and parakeratinized. Rete ridges were elongated, projected, and anastomosed into the gingival connective tissue. The connective tissue subjacent to the epithelium was composed of collagen fibers of varying thicknesses, with fusiform cells and many blood vessels. Inflammatory infiltrate was absent. No bone or osteoid were observed (Figures 4 through 7).

### DISCUSSION

Soft tissue augmentation with autogenous grafts is a widely used procedure in a variety of disciplines in dentistry. It is indicated in partially and fully



**FIGURES 4–7.** **FIGURE 4.** Panoramic view from the top: mucosa over connective tissue. The epithelium was slightly thickened and parakeratinized (hematoxylin and eosin [HE] stain,  $\times 4$ ). **FIGURE 5.** Border area between the mucosa and subepithelial connective showing the many vessels (HE stain,  $\times 25$ ). **FIGURE 6.** Middle part: young tissue with numerous fibroblasts with spindle-shaped nuclei and intense angiogenesis. The connective tissue subjacent to the epithelium was composed of collagen fibers of varying thicknesses, with fusiform cells and many blood vessels. Inflammatory infiltrate was absent. No bone or osteoid were observed (HE stain,  $\times 20$ ). **FIGURE 7.** Deep margin: mature connective tissue with fusiform cells and rare blood vessels. Inflammatory infiltrate was absent. No bone or osteoid were observed (HE stain,  $\times 10$ ).

edentulous patients to augment areas with a lack of or a reduced width of keratinized tissue, as well as to increase soft tissue volume. Various studies have suggested associations between an adequate width of keratinized tissue, higher survival rates of dental implants, health of the peri-implant mucosa, and improved esthetic outcome.<sup>2–4</sup>

The MERE technique exploits the healing processes to regenerate soft and hard tissues at the treated site. This approach restores proper placement and continuity to the mucogingival junction, increases the quantity of keratinized tissue, and

deepens the fornix. The technique appears to be relatively simple: it reduces the biological cost and corrects the atrophic ridge with a reduced number of surgical procedures and a reduced healing time. The clinical and patient-centered outcomes in this case were excellent. No scars resulting in esthetically displeasing appearance were observed.

The histologic findings revealed dense grafted tissues, providing long-term stability of the area. The rete ridges hyperplasia observed provided mechanical resistance to external irritations. No ligament or bone, characteristic for periodontal

regeneration, were observed. This indicated that the healing that occurred in our case was a long connective tissue attachment, which has been shown to be stable over time.<sup>5</sup> In conclusion, the presence of thick attached keratinized tissue around implants, may constitute a protective factor against marginal inflammation or trauma.

#### ABBREVIATION

MERE: modified edentulous ridge expansion

#### ACKNOWLEDGMENT

The authors wish to thank Dr Adolfo Apicella of the Anatomy and Histopathology Department "Luciano

Armanni," Second University of Naples, Naples, Italy, for his valuable contribution to the present study.

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