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# Experimental

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## The Temporal Sequence of Periosteal Attachment after Elevation

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This study investigated the adherence of periosteum to bone after elevation to document the temporal sequence of healing at the periosteal/bone interface. There has been a lack of consensus among surgeons as to the time required for healing at this interface; some believe that the healing achieves significant strength in a few days, whereas others believe that the periosteum does not adhere to the bone for many weeks. The aim of this study was to document the time course for healing, completeness of the reattachment, and structural characteristics of the union of bone and periosteum.

To test the hypothesis, scalp flaps were elevated in a subperiosteal plane and were reattached in 40 adult guinea pigs and controls. The individual groups were studied at 3, 6, 12, 30, and 90 days postoperatively. Postmortem study consisted of analysis of the mechanical and histologic findings. Strength of adherence was documented by measuring the force required for reverse avulsion of the flaps with an Instron Mini 44 tensiometer. The specimens were also submitted for electron microscopic examination. The mean tension recorded in the plateau phase of avulsion of the flaps was as follows: controls, 78 g; experimental at 3 and 6 days, not applicable (weak adherence not permitting exposure for reverse avulsion); 12 days, 39 g ( $p = 0.0001$ ); 30 days, 58 g ( $p = 0.0012$ ), and 90 days, 63 g ( $p = 0.0229$ ). There was a significant difference between all groups and the controls. Electron microscopic study showed collagen deposition at the bone periosteal interface, which became progressively more organized in the groups studied at 30 and 90 days, with decreasing amounts of inflammation and inflammatory cells.

This study demonstrated that healing at the bone/periosteal interface progresses at a rate consistent with healing of most other wounds, dispelling many widespread beliefs that the adherence at this interface was accelerated. The temporal sequence of healing at the periosteal bone interface should be considered in the various procedures in which periosteal flaps are elevated. For example, there is clinical relevance in subperiosteal brow lift procedures, in which the periosteum should be reattached by a fixation technique that will remain stable for a minimum of 30 days to allow adequate adherence

between the bone and periosteum at the postoperative elevated brow position. (*Plast. Reconstr. Surg.* 111: 1942, 2003.)

Periosteal elevation is often performed in many fields of surgery, including craniofacial, periodontal, hand, orthopedic, and aesthetic surgery.<sup>1-5</sup> In 1965, Skoog described the role of periosteal flaps in craniofacial surgery. He referred to these flaps as “boneless bone grafting” because of the formation of bone under the flaps.<sup>1</sup> Often in orthopedic surgical procedures, periosteal flaps are elevated before the placement of hardware, and the flaps are used to cover the hardware. Letts et al.<sup>3</sup> described the use of periosteal flaps to improve coverage in children with acetabular dysplasia, and Manske<sup>4</sup> described the use of periosteal flaps in the treatment of duplicated thumbs. Periosteal flaps are also useful during osteotomies, in which it is believed that the flap may expedite healing because the periosteum serves as an important source of blood supply for the underlying bone.<sup>6,7</sup>

Periosteal flaps are often used in cosmetic surgery (e.g., subperiosteal brow lift). The procedure's popularity stems from its ability to decrease the likelihood of injury to the supraorbital nerve. In addition, if performed endoscopically, the size of the incision (and scar) is reduced. Favorable results have been reported with special emphasis on the safety and efficiency of the procedure.<sup>8,9</sup> Troilius reported a mean increase of 7 mm in vertical height of the brow 1 year after endoscopic

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subperiosteal brow lift, a finding highlighting the longevity of the procedure.<sup>5</sup>

Despite the importance of this subject, little experimental work has examined the healing process at the periosteal/bone interface. Surgeons use several different fixation techniques in subperiosteal brow lifts. These range from simple chromic sutures (which last only 1 to 2 weeks) to Mytek anchors (which provide a longer period of fixation).

Clinical studies of the long-term effectiveness of the subperiosteal endoscopic brow lift have shown that favorable results do endure and benefit patients.<sup>8</sup> In the only relevant animal study, subgaleal and subperiosteal brow lifts were performed on rats and compared for adhesion strength only on postoperative days 2 through 10.<sup>10</sup> By day 10, the force required to elevate the subperiosteal flaps was significantly greater than that required for the subgaleal flaps. Microscopic examination with hematoxylin and eosin stain showed a maximum inflammatory response on day 6. This study did not, however, answer many questions about the healing at the periosteal/bone interface. Its fundamental flaws were a failure to isolate the healing at the periosteal bone interface from that at the skin and soft-tissue level and to extend the investigation longitudinally to draw conclusions of long-term results. Moreover, other problems were the failure to compare experimental measurements with those of controls and to calculate tension with reliability and accuracy.

The purposes of this study were to document the healing of periosteum to bone after elevation of the periosteal flap; to address the temporal sequence of healing at the periosteal/bone interface, including the completeness of healing; and to document the structural characteristics of the union between bone and periosteum.

The relevance of these data applies to any procedure in which a periosteal flap is elevated. The most direct relevance can be appreciated in the subperiosteal brow lift. The elevated and repositioned flap can be fixated by a variety of different techniques, including absorbable sutures, permanent sutures, or screw anchors. The method of fixation should, however, be chosen on the basis of the time course of periosteal–bone healing to allow for controlled fixation at a level determined by the surgeon during surgery. It is hypothesized that firm attachment of the periosteum to the un-

derlying bone should allow the subperiosteal brow lift to endure for a longer period of time.

#### MATERIALS AND METHODS

Forty adult guinea pigs (1 to 1.2 kg) were randomly divided into control and experimental groups to be studied at 3, 6, 12, 30, and 90 days postoperatively. Eight animals were assigned to the control group and six animals to the experimental groups (there were two operative deaths). Guinea pigs were chosen as the model because the size of the animal allowed surgical elevation of a sufficient area of the periosteum with minimal operative mortality. The size of the animals also allowed for reliable mechanical measurement with a tensiometer. The animals were treated in accordance with the guidelines of the New York University Berg Animal facility, under direct scrutiny of the New York University School of Medicine Institutional Animal Care and Use Committee.

The animals were given ketamine 40 mg/kg and xylazine 5 mg/kg intramuscularly just before the procedure. Lidocaine with epinephrine was injected locally with a 27-gauge needle along the incision and area of dissection for both anesthesia and hemostasis. Anesthesia was maintained by using various concentrations of ketamine, titrated to clinical effect. After an adequate level of anesthesia was achieved, the cranium of the animals was shaved, scrubbed with surgical soap, and coated with Betadine (Purdue Frederick Co., Norwalk, Conn.). A no. 15 blade was used to make a coronal incision across the coronal vertex from ear to ear. Subperiosteal dissection was commenced at the level of the incision and was carried rostrally beyond the anterior orbits and laterally over the supraorbital rim (Fig. 1). The periosteal layer was closed with four interrupted 4-0 nylon sutures under no tension. The skin and galeal layer was closed with a running 4-0 nylon suture and a dressing was applied. The experimental groups were studied at 3, 6, 12, 30, and 90 days postoperatively.

Biomechanical and histologic studies comprised the evaluation techniques. To measure the mechanical strength of the adherence of the periosteum to the underlying bone, the entire skin and subcutaneous tissues were resected in a subgaleal plane. The rostral-most portion of the periosteum was elevated to the level of the anterior orbit. A 1-cm central strip of periosteum was isolated with a scalpel, with care taken not to disturb the central periosteal

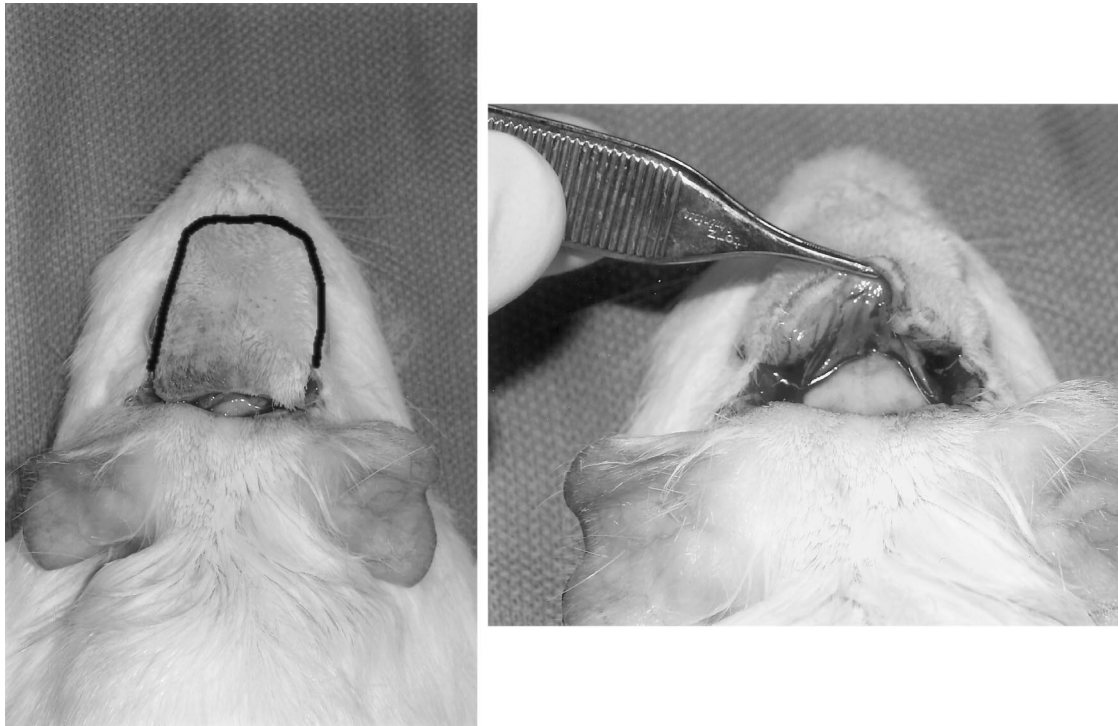


FIG. 1. (Left) The extent of the skin incision and the area of subperiosteal dissection. (Right) Subperiosteal dissection/elevation.

strip. The central strip was then connected by means of a clamp to a microtensiometer (Instron Mini 44, Instron Co., Canton, Mass.). The force needed to avulse the periosteum from the bone was measured and recorded. The control animals consisted of six animals not operated on but studied in the same manner. Histologic study consisted of electron microscopic examination of the periosteal/bone interface. The specimens were placed in glutaraldehyde and embedded in resin. They were stained with silver, sectioned with a diamond blade at  $1\ \mu$ , and examined at  $\times 10,000$ .

## RESULTS

### Biomechanical

The force required to avulse the periosteum from bone was measured for both controls and experimental groups. As expected, control animals required the most force, with a mean of 78 g (SD  $\pm 9$  g). At 3 and 6 days, the adherence was too weak to allow for subgaleal dissection; therefore, avulsion tension could not be measured. The group studied at 12 days had an avulsion tension of 39 g (SD  $\pm 6$  g). The group studied at 30 days had an intermediate level of adherence, with avulsion tension measuring 57 g (SD  $\pm 7$  g). The experimental group

requiring the greatest amount of tension was that examined at 90 days, with a mean of 62 g (SD  $\pm 11$  g) (Fig. 2).

A one-way analysis of variance was calculated on the tension force required to avulse the periosteal flaps; the analysis showed significance,  $F(5,30) = 141$ ,  $p < 0.0001$  ( $R^2 = 0.959$ ). Independent  $t$  tests used to compare the experimental groups showed significant differences among all groups ( $p < 0.001$ ), with the exception of the comparison of the groups studied at 30 and 90 days ( $p = 0.373$ ).

### Microscopic

One specimen from each group was submitted for electron microscopic examination. Because of the method of fixation of these specimens in a hard resin and sectioning with a diamond blade, there was minimal distortion of the bone/periosteal interface. Control animals showed a normal bone/periosteal interface (Fig. 3, above, left). Specimens at day 30 showed collagen deposition with minimal cellularity, consistent with an organized scar (Fig. 3, below, left). Specimens at day 90 showed a well-organized layer of collagen with minimal cellularity, findings also characteristic of an organized scar (Fig. 3, right)



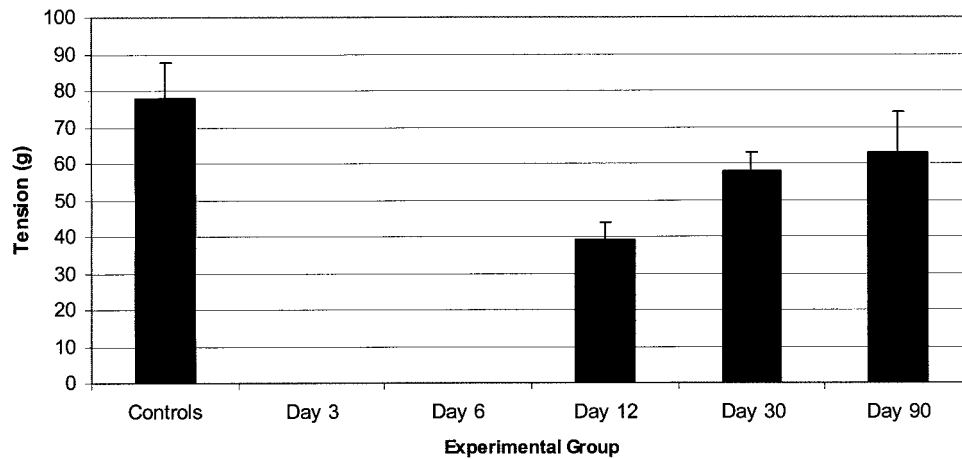


FIG. 2. Mean tension required to avulse periosteal flaps at designated intervals. *T-bars* represent standard deviations.

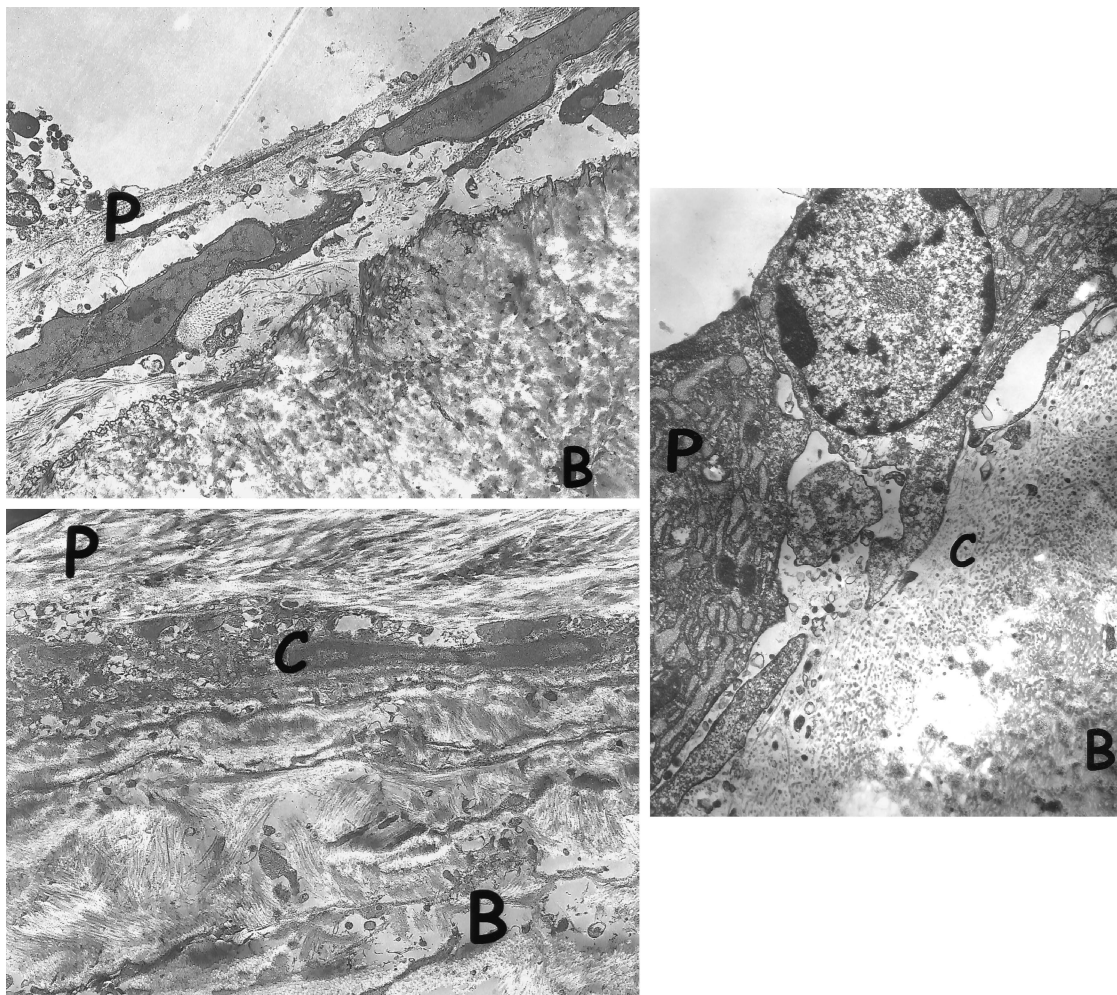


FIG. 3. Electron microscopic results. (*Above, left*) Results from a control animal demonstrating a normal bone/periosteal interface. Note the sparse collagen between the periosteum and underlying bone ( $\times 10,000$ ). (*Below, left*) Results from the group studied at 30 days with collagen deposition and only minimal cellularity, a finding consistent with newly formed scar tissue ( $\times 10,000$ ). (*Right*) Results from the group studied at 90 days with a well-organized layer of collagen between the bone and overlying periosteum ( $\times 10,000$ ). *P*, periosteum; *B*, bone; *C*, collagen.

## DISCUSSION

In this study, healing at the periosteal/bone interface achieves only minimal strength until approximately 12 days, with greater strength achieved at approximately 30 days, a finding similar to that observed in approximate healing times observed in other types of wound healing. Studies examining the bursting strength of surgically created skin wounds demonstrated that these wounds have little strength until approximately 14 days.<sup>11-13</sup> All of the studies showed that the wounds gained strength at approximately the same rate during the first 2 to 3 weeks, with a plateau of tensile strength achieved at approximately 60 days.<sup>11</sup> These studies also showed that the tensile strength of a wound does not reach that of the original, unwounded skin; rather, it levels off at about 80 percent.<sup>12</sup> In addition, the authors also found that because of technical reasons (as also reported in this study), measurable breaking strength could not be accurately determined for at least 5 days.<sup>13</sup>

In the current study, microscopic examination showed that adherence between the periosteum and bone is a result of scar deposition. The specimens showed a constant evolution of scar tissue, as previously reported in all types of healing wounds. This finding is also consistent with healing between any two separated layers of tissue. In almost every situation, when tissues are separated, they heal by deposition of scar tissue. Scarring, wherever it is located, follows an orderly process by which strength of adherence is attained. Healing at the bone/periosteal interface is no different.

For example, the senior author (J.G.M.) observed, after exploring the surgical results of a patient recently treated by cranial vault remodeling, that the subperiosteal plane was extremely easy to open; indeed, this often requires only a simple finger sweep to elevate the scalp if exploration is performed within the first 10 days postoperatively. In such procedures, in which the major focus is on the bony skeleton, there are often minimal shearing forces between the periosteum and the underlying bone. In the subperiosteal brow lift procedure, however, firm reattachment of the periosteum to the bone is critical to the overall success of the operation. The forehead-scalp flap can have a significant shearing vector directing the flap to its preoperative anatomic position.

Several years after re-elevation of a subperiosteal flap, craniofacial surgeons report that the bone/periosteal interface has a different appearance than that originally found in the patient before the initial surgery. The reported difference is the layer of scar tissue attaching the previously elevated periosteum to the bone. It is this scar layer that attaches the periosteal flap to the bone. This knowledge and the understanding of the orderly progression of scar formation and maturation leads to one obvious conclusion: To reliably allow scar tissue to form between the periosteum and bone such that the scar will hold the elevated flap at the desired position, there must be secure fixation so that the force vectors are taken out of play during the initial healing and scar maturation process. Our findings show that this initial healing interval is approximately 30 days in our guinea pig model, and this may likely correlate closely in human subjects. In addition to this known force vector, there are several unknown variables. Postoperatively, patients are unpredictable. They may test the strength of their new brow lift operation or be involved in unexpected trauma. These variables may affect the final outcome. A long-lasting method of fixation will remove unneeded variables and possibly avoid potential complications, thus providing the best possible result for the patient.

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