

## An integrative approach for comparing microcirculation between normal and alveolar cleft gingiva in children scheduled for secondary bone grafting procedures

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**Objective.** The aim of this study was to compare microcirculatory parameters in normal versus alveolar cleft gingiva in children selected for secondary bone grafting procedures.

**Study Design.** This study included 11 consecutive patients with complete unilateral alveolar clefts who required secondary bone grafting procedures. In a split-mouth design, noninvasive real-time simultaneous measurements among tissue oxygen saturation (StO<sub>2</sub>), hemoglobin level (rHb), and blood flow parameters were obtained from normal and alveolar cleft gingiva using spectrophotometry and laser Doppler flowmetry. Subsequent noninvasive capillary density measurements and tissue microangiarchitecture were assessed using sidestream dark-field imaging.

**Results.** There were no significant differences in StO<sub>2</sub> and rHb between normal and alveolar cleft gingiva. Blood flow, blood flow velocity, and capillary density were significantly decreased in alveolar cleft gingiva ( $P < 0.05$ ).

**Conclusions.** Alveolar cleft reconstructions alter gingival microperfusion properties, and microvascular changes adapt to conserve peak oxygen saturation. (Oral Surg Oral Med Oral Pathol Oral Radiol 2013;115:304-309)

Cleft lip with or without palate constitutes one of the most common congenital anomalies of the craniofacial region.<sup>1</sup> Corrective surgery requires a planned series of procedures in a sequential manner at the proper time over several years, from infancy to adulthood, for restoration of speech, feeding, overall facial esthetics, and adequate growth and development of the facial skeleton. Inspection of the many different surgical techniques that have been used to repair orofacial clefts, iatrogenic consequences such as wound contraction, and scar tissue formation have resulted in detrimental growth disturbances of the midface.<sup>2</sup> As a result, the important role of the mucosal tissues covering the un-

derlying maxillofacial bones has been highlighted. Repetitive surgical reconstructions displace tissue components such as tissue cells, blood vessels, and bone. This results in fibrous connective tissue formation with substantial accumulation of tissue scarring. Serial surgeries with incisions through preexisting scar tissue further displace the regionally scattered viable tissues, which prolong ischemia and hypoxia. This subsequently alters the temporal course of wound healing. Moreover, the inadvertent propensity for wound dehiscence and tissue necrosis is significantly elevated.

The gold standard for evaluating wound status is visual appraisal by the surgeon. There are instruments readily available for determining the health and vitality of oral tissues such as laser Doppler flowmetry (LDF) for perfusion parameters<sup>3-8</sup> and tissue reflectance spectrophotometry and near-infrared spectroscopy for hemoglobin level and oxygen saturation parameters.<sup>9,10</sup> Thus, a closer examination of the tissue of interest can provide detailed information on the status of wound metabolic physiology and regeneration. This study

### Conflict of Interest

In addition to his listed affiliation, Can Ince is inventor of the sidestream dark-field imaging technique and, therefore, holds patents and stock in this technology. The remaining authors indicate they have no personal financial relationships or potential conflicts of interest to declare.

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### Statement of Clinical Relevance

Alveolar cleft reconstructions alter gingival microcirculatory properties, which could prolong tissue hypoxia and delay wound healing. Direct microcirculatory inspection could provide a way of planning subsequent surgical approaches for maintaining peak tissue oxygen and perfusion levels.

presents the application of a monitoring system, the Oxygen-to-See (O2C), for simultaneous measurements of gingival oxygenation and microperfusion parameters using reflectance spectrophotometry and LDF. The O2C has been extensively used in human tissues in various medical and surgical research areas, including the gastrointestinal tract,<sup>11,12</sup> peripheral arterial occlusive disease,<sup>13,14</sup> neurosurgery,<sup>15,16</sup> transplantation surgery,<sup>17,18</sup> and maxillofacial surgery.<sup>19,20</sup> Recent gingival microcirculation research using optical spectroscopic-based microvascular imaging techniques, such as orthogonal polarization spectral imaging<sup>21,22</sup> and more recently sidestream dark-field (SDF) imaging,<sup>23</sup> has demonstrated reliable anatomically derived measurements of capillary density, a tissue microvascular diffusion-related parameter. SDF imaging provides a platform for real-time, noninvasive observations of tissue microvascular architecture and angiomorphology, which is necessary for evaluating the nature of vascular adaptations resulting from periodontal surgery or maxillofacial reconstructions involving the periodontal mucosa.

We hypothesized that the microvasculature of serially operated alveolar cleft gingiva follows a course of angiogenesis that alters vascular morphology and spatial distribution of the microcirculation to compensate for the damage by adequately establishing reperfusion to the surgically reconstructed alveolar arch mucosa. Therefore, the aim of this study was to compare microcirculatory parameters in normal versus alveolar cleft gingiva in children selected for secondary bone grafting procedures.

## MATERIAL AND METHODS

This study was reviewed and approved by the institutional Medical Ethics Committee of the Academic Medical Center of the University of Amsterdam. All participating parties were well informed about the study guidelines and procedures, and written informed consent was obtained from the parents of the participating patients. This study was performed in compliance with the principles established in the Helsinki Declaration.

### Patients

This was a single-center observational study. Patients referred to the Department of Oral and Maxillofacial Surgery for secondary bone grafting procedures for maxillary arch reconstruction of alveolar clefts were eligible to participate in this investigation. Eleven consecutive children, 4 females and 7 males, with a mean age of  $10 \pm 1.4$  years (range 7-12 years) and either a left or a right complete unilateral alveolar cleft (AC), were selected for this study. Important determinants for inclusion were the type of cleft (i.e., complete), the orientation of the cleft (unilateral), and the accessibility

of the location for measurements using the O2C apparatus and SDF imaging techniques.

### O2C apparatus, measurement principles, and probe description

The tissue oxygen saturation and perfusion analysis system O2C (Medizintechnik GmbH, Giessen, Germany) consists of a computer with built-in laser (Laser Device Class 3B, Protective Class 1, CE Mark) and light-emitting diodes and a fiber-optic measurement probe. The O2C transmits continuous wave laser light at 830 nm (<30 mW) and visible (white) light at 500 to 800 nm (1-nm resolution, <30 mW) to tissue, where it is scattered and collected on the surface of glass fiber elements in the probe. Oxygen saturation (StO<sub>2</sub>) is determined by the color of blood; hence, for spectrophotometry measurements, the tissue was illuminated with white light and the spectrum of the backscattered light was collected and analyzed to calculate the tissue optical absorption spectrum. The O2C apparatus determines StO<sub>2</sub> based on the differentiating absorption spectra of oxygenated and deoxygenated hemoglobin (Hb). Oxygenated Hb has 2 absorption peaks in the visible spectrum centered at 542 and 577 nm and deoxygenated Hb has 1 absorption peak centered at 556 nm; hence, by scaling the measured absorption spectrum between the known absorption spectra of oxygenated and deoxygenated Hb, StO<sub>2</sub> can be determined. The total optical absorption is used to reflect the tissue Hb content (rHb).

Gingival blood flow (BF) and BF velocity (BFv) were determined by LDF. Movements of red blood cells (RBCs) cause a Doppler shift of the illuminated laser light, which is detected, analyzed, and displayed as BFv. The detected laser signal also correlates with the number of moving RBCs in tissue. The relative BF was calculated by multiplying the number of moving RBCs by the velocity of each RBC.

The O2C probe contains 2 illuminating and detecting glass fiber elements enclosed with epoxy casting resin (nontoxic to human tissue). The measurement depth is dependent on the distance between the illuminating and detecting elements in the fiber-optic probe and cannot be set by the operator; thus, the measurement depth is defined by the probe and was estimated as half the space between the illumination fiber and the detection fiber. For the present study, a single probe (Flat Probe LF-2; probe head: width 12 mm, height 5.5 mm, length 44.5 mm; probe length 300 cm), designed for cutaneous applications, was applied on the normal (control) and alveolar cleft gingival areas, which had measured depths of approximately 1 to 2 mm. The O2C probe must be used with a sterile cover because the manufacturer does not permit gas sterilization.

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