Semi-automated registration and segmentation for gingival tissue volume measurement on 3D OCT images

Geng Wang,1,2 Nhán Minh Lê,1 Xiaohui Hu,1 Yuxuan Cheng,1 Steven L. Jacques,1 Hrebesh Subhash,2 and Ruikang K. Wang1,*

1University of Washington, Department of Bioengineering, Seattle, WA 98195, USA
2Clinical Method Development - Oral Care, Colgate-Palmolive Company, Piscataway, NJ 08854, USA
*wangrk@uw.edu

Abstract: The change in gingival tissue volume may be used to indicate changes in gingival inflammation, which may be useful for the clinical assessment of gingival health. Properly quantifying gingival tissue volume requires a robust technique for accurate registration and segmentation of longitudinally captured 3-dimensional (3D) images. In this paper, a semi-automated registration and segmentation method for micrometer resolution measurement of gingival-tissue volume is proposed for 3D optical coherence tomography (OCT) imaging. For quantification, relative changes in gingiva tissue volume are measured based on changes in the gingiva surface height using the tooth surface as a reference. This report conducted repeatability tests on this method drawn from repeated scans in one patient, indicating an error of the point cloud registration method for oral OCT imaging is $63.08 \pm 4.52 \mu m (1\sigma)$, and the measurement error of the gingival tissue average thickness is $-3.40 \pm 21.85 \mu m (1\sigma)$.

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1. Introduction

Gingiva health is an important indicator to consider when evaluating oral health [1,2]. Gingivitis [3,4] is a reversible form of oral inflammation usually caused by adherence of mature biofilm to teeth. Gingivitis is antecedent to the more tissue destroying form of gum disease, periodontitis [5–7]. The first sign of gingivitis is a slight change in the color and texture of gingival tissue; whilst signs of advanced gingivitis can be identified by bleeding on probing and edema (by a clinician) or bleeding during oral hygiene (by the patient) [8,9]. The later stages are easier to discern, while the earlier signs require a trained dental professional to observe.

The current gold standard for the clinical assessment of gingivitis is based on measuring the gingival index (GI) [10]. GI methods are index-based, heavily relying on subjective visual inspections, and might not provide sufficient accuracy for evaluating the progression between different gingivitis stages. Probing of gingival tissue using a metal probe is also an invasive approach that often leads to patient discomfort and even pain [11,12]. In the past, color photography was utilized to analyze gingival inflammation as well as color characteristic analyses [8]; however, the limitation is that 2D digital images cannot be used to measure changes in gingival tissue volume. 3D camera-based time of flight and structured illumination techniques may solve this problem [13], but they are time consuming and limited by its inability to replicate in vivo conditions.

Optical coherence tomography (OCT) is a proven technology that provides in vivo, non-invasive, 3-dimensional structural and functional information with microscopic resolution [14–18]. It is a useful imaging modality for examining the oral cavity because of its capability to visualize the 3D morphology of the tooth and gingival tissue precisely [7,19–21]. However, unlike measuring volume changes in other tissues, it is difficult to measure the absolute volume of the gingiva with
OCT because there are not clear reference boundaries available to define the absolute volume. For this oral investigation, a relative volume is measured instead. We have previously reported the differences in tissue volume and blood-vessel density between inflamed and healthy gingiva [22] with an accuracy in the order of magnitude of tens of micrometers. To compare these differences, the use of 3D image registration and segmentation is essential [23]. We noticed that the registration and segmentation of 3D OCT images have been reported in the prior literature to address the research and clinical purposes in the applications of OCT in ophthalmology [24–26]. While the fundamentals of registration and segmentation are similar, our purpose here is to develop such method for the application in gingival tissue.

In this paper, we describe the development of a semi-automated registration and segmentation method using 3D OCT images to accurately measure gingival tissue volume. In order to demonstrate the capability of our method, 17 consecutive volumetric OCT images were captured from the same tooth and gingiva in a period of less than 15 min. The subject was asked to reposition his head for imaging between each volumetric scan. Then these 3D images were registered and analyzed. Image registration was realized using the tooth surface as the reference. The null hypothesis was that the average of the thickness measurements (from the 17 OCT cube) was not significantly different from zero, which means the thickness of gingiva did not change during the scan. The assumption of constant gingival thickness is reasonable since we measured the gingiva volume during a period of less than 15 minutes. We demonstrate experimentally that the accuracy of the gingival thickness measurement is within 50 µm and is feasible for applying to volume measurements in future clinical studies.

2. Registration of the 3D OCT images of gingival tissue

In this study, the volumetric OCT oral data was captured using a high speed swept source- OCT system with a 200kHz A-scan rate, 1310nm central wavelength and 110nm bandwidth. The axial pixel resolution (Z direction) in tissue was 5.2 µm approximately, and the lateral pixel resolution (X/Y direction) was 32 µm. For the scanning protocol of oral volumetric OCT image, we set a 16×12.8 mm² scanning field of view (FOV) with 500 A-scans in one B-scan and 400 locations in the low axis (Y direction). Meanwhile, a CMOS camera was integrated within the OCT probe to provide visual information to guide the OCT to image the gingiva, as shown in Fig. 1.

To measure the volumetric change in gingiva tissue accurately, the quantification should be performed at a fixed and defined gingiva tissue region. However, it is difficult to guarantee that the same tissue volume is examined with the OCT for every evaluation, simply because of the possible movement of the subject and the possible misalignment of the OCT probe. Every OCT image thus must be co-registered in order to achieve accurate quantification of the changes in the gingiva tissue volume over time [27,28]. It is clear that the registration between 3D images requires a reference image. For the purpose of the experiments described in this manuscript, we made the assumption that the morphological structure of the tooth was constant over time. This is a reasonable assumption since the tooth is a hard tissue and is unlikely to change its shape (or deform) over time in a short period during our investigation. Therefore, the solid tooth surface can be used as a reference for the purpose of co-registration. Another consideration for volumetric image registration is that the OCT images should be acquired in a relatively stable environment to minimize the tissue motion between intra- and inter- imaging sessions. Typically, this requirement can be fulfilled by the hardware setup and subject preparation before the imaging takes place. Therefore, the registration in our investigation consists of two main steps: pre-imaging measures (i.e. hardware and subject preparation) and post-imaging registration (i.e. software manipulations).
Fig. 1. (a) Modified slit-lamp frame for OCT imaging of tooth and gingiva, where the OCT imaging probe was mounted on the U-arm. A custom-fitted bite-piece was designed and placed just in the front of chin-rest. On the bite piece, there is situated impression material that was made to the contour and shape of tooth of each individual subject. (b) schematic showing how the scanning beam is delivered to the sample through the scan lens. (c) oral OCT image sampling of subject with bite-piece and lip retractor.

2.1. Pre-imaging hardware consideration

Volumetric OCT scans usually take a few seconds of time to acquire. During this scanning time frame, the patient needs to keep still to prevent distortions and artifacts from affecting the acquired OCT image. Patient motion and misalignment, relative to the OCT probe, will reduce the number of qualified and matched feature points (for registration), leading to a failure or large error in the post-image registration. Hence, measures have to be taken to stabilize the patient and keep their motion at a minimum during imaging.

To meet this requirement, we designed a mechanical frame and bite-piece as shown in Fig. 1(a). By moving the guide-rail and rotating the U-arm, the probe could be translated in three linear directions and rotated around the vertical axis. The bite-piece used impression material to help constrain the position of the tooth. Moreover, the bite-piece size was custom-fitted for each patient and fabricated by a 3D printer. A lip retractor was also used for patients to ensure a wide-open imaging window, facilitating the OCT imaging. Using separate bite-piece and lip-retractor for each subject can effectively avoid cross infection. The oral OCT image sampling of a subject under the above constraint is shown in Fig. 1(c).

For OCT imaging, the scanning beam was delivered to the sample through a scan lens, as shown in Fig. 1(b). However, there was distortion of the optical images caused by the lens, particularly for scanning rays at the extreme edges of the lens, which could influence the image registration. In order to avoid this edge distortion, only the middle FOV of the scan lens was selected for OCT imaging. A CMOS camera was situated within the probe to capture a photograph of the tooth with a slightly larger FOV compared to the OCT imaging FOV to help determine the imaging region, and keep the imaging region constant during each visit, as shown in Fig. 2(a).

Another challenge for OCT imaging is to maintain the same focal position of the 3D image in the depth(Z) direction at different imaging sessions. An inability to maintain the focal position would affect the accuracy of image quantification in subsequent processing. To address this problem, three real-time B-frames of OCT images were displayed on the fly to the operator, which
were equally spaced in the lateral (Y) direction, as shown in Fig. 2(b). The surface topology of the tooth and gingiva at positions left (L), medial (M) and right (R) in Fig. 2(b)) were extracted (shown as red lines) at the initial baseline scan, and then used as the reference for the operator to position the imaging probe at the follow-up visits. This method also helped to reduce rotation differences of the tooth/gingiva during the different visits.

2.2. Software registration algorithm

Registration was grossly adjusted using the above described hardware adjustments. For fine adjustments, the point cloud method [29,30] was employed. This method is based on the iterative closest point (ICP) algorithm [31,32], which can iteratively minimize the difference between two clouds of feature points. The individual points are not aligned, but rather the surfaces defined by the populations of points are aligned. The points are generated randomly based on the surface of the tooth in the 3D-OCT volume. The iterative transformation includes translation and rotation, and the point-cloud registration minimizes the rotation and translation of any 3D point cloud generated from the OCT. Moreover, due to the difference in refractive index between air, tooth and gingiva, there were strong reflections from the tissue surfaces. Therefore, the maximum gradient value in a typical A-line (Z direction) was located at the surface of the gingiva/tooth, as shown in Fig. 3. The spatial positions of maximum gradient values within the 3D OCT scans were extracted as the feature points of the tooth surface, which were used to conduct the point cloud registration for consecutive 3D OCT volumetric images of the gingiva/tooth structure.

The point cloud registration is a kind of rigid transformation, which only includes translation and rotation around X/Y/Z axis of the volumetric OCT image. The X/Y/Z direction is defined in Fig. 3(a). In OCT images, the X, Y-dimension pixel sizes are specified by the scanning, while the Z-dimension pixel size is determined by the OCT A-line acquisition. Therefore, the pixel size in the lateral direction (X/Y) is different from the pixel size in the axial direction (Z) for raw 3D OCT images as determined in this study. The pixel size was calibrated by OCT imaging using a Vernier caliper. For the images captured at different visits, the orientation of the tooth presented in the volumetric images could differ due to differences in the probing angle or patient positioning, which would cause a difference in the voxel size of the 3D tooth images. Therefore, the 3D OCT images for registration needed to be re-sampled to provide equal pixel sizes in all three-dimensions before conducting the point cloud registration. Ultimately, the purpose of the registration is to find an affined transformation matrix $T_{form}$ for aligning the moving point cloud with the reference point cloud.
To perform the point cloud registration, one should start with two 3D OCT images: one ($I_{\text{ref}}$) is a reference image, and the other ($I_{\text{new}}$) is the image to be co-registered with $I_{\text{ref}}$. The OCT images have different voxel sizes in the z (depth) dimension (5.2 µm) versus the x and y dimensions (32 µm). Before co-registering the images, they are first resampled so that the z size of all the voxels is also 32 µm. The resampled reference image is called $I_{\text{ref,rs}}$ and the resampled new image is called $I_{\text{new,rs}}$. The process of co-registering a new image $I_{\text{new}}$ with a reference image $I_{\text{ref}}$ involves four steps:

1. Extract feature points (x, y, z positions) on the surface of the tooth and gingiva using the resampled images:
   
   $P_{\text{ref}} = \text{plotPointCloud}(I_{\text{ref,rs}})$
   
   $P_{\text{new}} = \text{plotPointCloud}(I_{\text{new,rs}})$
   
   This step finds the 3D tooth and gingiva surfaces in an OCT image, yielding 500 positions on the x-axis, and 400 positions along the y axis, for a total of 200,000 points on the entire tissue surface. Then a subset of points ($P_{\text{ref}}$ or $P_{\text{new}}$) on the tooth only are selected (~10,000 points). The program plotPointCloud.m was written by the authors, which mainly included the function of finding gradient magnitude of 3-D image and writing 3-D point cloud.

2. Generate a transformation matrix, $T_{\text{form}}$, that will translate and rotate the set of points, $P_{\text{new}}$, to generate the set $P_{\text{aligned}}$, which is aligned with the set of points $P_{\text{ref}}$:
   
   $[T_{\text{form}}, P_{\text{aligned}}, \text{RMSE}] = \text{pcregistericp}(P_{\text{new}}, P_{\text{ref}})$
   
   where RMSE is the root mean squared error. The program pcregistericp.m is a function in the MATLAB Computer Vision System Toolbox. This program co-registers the surfaces specified by the points $P_{\text{ref}}$ and $P_{\text{new}}$, not the individual points which are different in the two images.

3. Apply $T_{\text{form}}$ to translate and rotate the image $I_{\text{new,rs}}$ to yield $I_{\text{aligned,rs}}$:
   
   $I_{\text{aligned,rs}} = \text{imwarp}(I_{\text{new,rs}}, T_{\text{form}})$
   
   which is now aligned with $I_{\text{ref,rs}}$. The program imwarp.m is a standard MATLAB routine.

4. After completing the registration, the image $I_{\text{aligned,rs}}$ is resampled so that the size of the voxels in the z direction are returned to being 5.2 µm, to yield $I_{\text{aligned}}$, which is now aligned with $I_{\text{ref}}$. 
3. Measurement of gingival tissue volume

In this study, the inter-dental area of the gingiva was selected as the region of interest (ROI, light blue area in Fig. 4) to quantify the change of tissue volume, because the inter-dental area of the gingiva is more sensitive to inflammation than other areas. The blue boundary line marking the ROI was extracted by manual segmentation, as shown in the right image of Fig. 4. Meanwhile, the upper boundary line was determined by the top of the red-dot frame, and this red-dot frame area was used to create a binary mask to classify the region of interest and was constant for each different visit. The red-dot box was located in the first and fourth area of two adjacent teeth. The volume change of the gingival tissue was calculated based on the difference between the height of two co-registered OCT volumetric gingival images within the ROI. However, because the volume of gingival tissue varies for different teeth at the baseline, it is not appropriate to compare the volume change of gingival tissue among different teeth. To solve this problem, the average thickness change ($\Delta z$) was employed to reflect the gingival volume change of different teeth, and was calculated according to the following:

$$\Delta z = \frac{\sum \text{pixel}_{\text{volume}} \cdot \text{voxel}_z}{\sum \text{pixel}_{\text{area}}}$$

where $\sum \text{pixel}_{\text{volume}}$ denotes the number of pixels between two gingival surfaces within the co-registered ROI, $\sum \text{pixel}_{\text{area}}$ denotes the number of pixels in the 2D ROI, and voxel$_z$ denotes the pixel size in the z direction.

![Fig. 4. Illustration for ROI area selection (light blue area) and gingival margin segmentation of the inter-dental gingiva (blue boundary line), one tooth is evenly divided into 4 areas, the first and fourth area of any two adjacent teeth are defined as the inter-dental region.](image)

4. Experimental results and discussion

We first conducted an experiment to test the ability of registration algorithm using the tooth surface as the reference. In the experiment, we used a tooth model onto which a piece of scotch tape (approximately 5mm×3mm) with known thickness was firmly attached. We first captured one 3D OCT image of the tooth model without the tape as the reference image. And then 6 consecutive OCT images of the tooth model with tape were captured, wherein between imaging sessions, the tooth model was repositioned to simulate the in vivo situation. These 3D images were then subject to the registration as described in Section 2. After registration, the average thickness of the scotch tape on top of the tooth model for each image was evaluated (Fig. 5(a)). The results showed that the average thickness of the scotch tape was 19.99 ± 1.29µm (3σ), agreed well with the physical thickness of the scotch tape used. To observe the registration result directly,
the images with surface height difference information after registration were employed, and the surface height difference was calculated and mapped to an en face plane, as shown in Fig. 5(b). This experiment confirms that the proposed registration approach works to achieve sufficient accuracy that should be suitable for the quantification of the changes in the volume measurement in vivo (see below).

![Fig. 5.](image)

In the in vivo study below, the root mean square error (RMSE) was employed to assess the error of the point cloud registration for the oral OCT images, which refers to the RMSE of Euclidean distance between the fixed point cloud (illustrated with purple in Fig. 6(a)) and the affined point cloud (illustrated with green in Fig. 6(b)). The RMSE of registration was calculated according to Eq. (2):

$$RMSE = \sqrt{\frac{1}{n} \sum_{i=1}^{n} (x_{fi} - x_{ai})^2}$$

where $x_{fi}$ denotes the coordinate value of the fixed point cloud, $x_{ai}$ denotes the coordinate value of the affined point cloud, and $n$ is the number of points used for registration. Therefore, the higher RMSE value corresponds to a larger error of registration.

To verify the repeatability of this registration method in vivo, we had 17 in-vivo volumetric OCT imaging sessions from the same tooth and gingiva within 15 min period, which were taken after repositioning the tooth for each image. Then the point cloud registration was conducted between 40 possible pairs of volumetric OCT images randomly chosen from all the possible comparisons. The histogram of RMSE for the 40 comparisons is shown in Fig. 6(b), which indicates that the mean value of RMSE is 1.93 pixels, and the standard deviation (SD) of RMSE is 0.138 pixel. This means that the average error of the point cloud registration method when applied to oral OCT images is $1.93 \pm 1.04$ pixel ($63.08 \pm 4.52 \mu m$) under 1σ standard deviation, and $1.93 \pm 0.41$ pixel ($63.08 \pm 13.56 \mu m$) under 3σ standard deviation.

The RMSE value of registration method is influenced by many factors. For volumetric OCT images, the motion during scanning has significant influence on the RMSE value. In order to validate the influence of motion on the RMSE value of registration in oral OCT imaging, two registered images with different RMSE values were selected (Fig. 7) to observe the difference due to motion. The RMSE values of registration were 1.74 and 2.28, respectively. Visually, the images with surface height difference information after registration are shown in Figs. 7(a) and 7(d). It is worth noting that the height difference should be close to zero on the tooth area for the
image without motion, since the registration procedure should be able to register two volumes using tooth surface as reference. Any residual height difference on the tooth was considered as motion and registration error (except for the cases where there is plaque at the junction of teeth and gingiva).

Compared with Fig. 7(a), the motion is more obvious in the A1 and A2 areas of Fig. 7(d), and the surface height difference of tooth tends to zero in other areas of the registered tooth. This result also can be observed in cross-section images (Fig. 7(b) and 7(e) for the lateral cross-section image and Fig. 7(c) and 7(f) for the vertical cross-section image). When the surface of merged image presents green, purple, or white color, it indicates that the surface height of the affined tooth is higher, lower, or the same respectively than the reference tooth. For the A1 area of Fig. 7(d), the surface height of the affined tooth is higher than the height of the reference tooth, but for the A2 area of Fig. 7(d), it is lower, and the motion in both A1 and A2 areas of Fig. 7(d) is higher than the same areas of Fig. 7(a). Overall, the magnitude of the RMSE value is positively related to the magnitude of the motion during scanning, and the motion has significant influence on the RMSE value. Therefore, it is essential to adopt the measures in section 2.1. The average thickness change of the right inter-dental area of Fig. 7(a) and 7(d) was 16.12 \( \mu m \) and 32.90 \( \mu m \), respectively.

To calculate the volume measurement error of gingival tissue based on the registration and segmentation method proposed in this study, the average thickness change of the inter-dental gingiva region was measured on the aforementioned co-registered OCT images after registration. The gingiva average thickness changes are shown in Fig. 8(a). The error of the repeatability test of volume measurement is shown in Fig. 8(b), the result shows that the value of apparent gingiva average thickness change, i.e., for the variability of the measurement, the mean value was -3.40 \( \mu m \), the interquartile range(IQR) was 27.07 \( \mu m \), and the standard deviation was 21.85 \( \mu m \). When there is no error in the registration, the volume change should be zero for the repeatability test, this means that the measurement error of gingival tissue average thickness for oral OCT imaging is \(-3.40 \pm 21.85 \mu m\) (1σ).

To reduce the error in gingival volume measurement during imaging, one way is to capture a number of repeated 3D OCT scans at each tooth per visit. These 3D OCT images are then registered with the specific 3D OCT reference image from the first visit for the same tooth separately, and the change of gingival tissue volume averaged from these repeated measurements.

In this study, the affine transformation was employed in the registration method, which does not consider the localized distortion of the 3D image. The B-spline (elastic) transformation may
Fig. 7. Two co-registered images for the same tooth with different RMSE values. (a) merged image after registration (RMSE = 1.74) with color-coded surface height difference, (b) lateral cross-section image of Fig. 7(a), and green/purple color respects affined/ reference image respectively, (c) vertical cross-section image of Fig. 7(a), (d) merged image after registration (RMSE = 2.28) with color-coded surface height difference, (e) lateral cross-section image of Fig. 7(d), (f) vertical cross-section image of Fig. 7(d).

Fig. 8. The distribution and error of same gingiva thickness measurement at same visit. (a) Histogram of gingiva average thickness, (b) error bar of gingiva average thickness.
be used to further minimize the influence of patient motion, but with a cost of computational power. Moreover, in order to facilitate point cloud registration, it is recommended to collect a relatively large FOV to include at least one edge of the tooth, otherwise, the point cloud data would be too smooth to provide enough features for a reliable registration. In addition, in our study, only the tooth surface topology was utilized in the point cloud registration, which has a high failure rate when there are not enough features in the point cloud data. OCT can provide 3-dimensional intensity data for co-registration, but the OCT signal from the tooth surface will decrease if the surface is not perpendicular to the incident angle for the OCT beam. The tooth surface contour should always be considered.

5. Conclusion

A semi-automated registration and segmentation method for gingival tissue volume measurement has been proposed based on three-dimension OCT images, which could be used to monitor changes from health to disease in gingiva tissue. In order to measure the volumetric change of gingival tissue in different visits, the gingiva area was registered based on: 1) the pre-registration method using manual alignment of the imaging probe on the mechanical frame with bite-piece, 2) the point cloud registration method based on the ICP algorithm on the surface of the corresponding tooth. Moreover, the measurement method of gingival average thickness change was proposed to reflect the volume change of gingiva tissue. The results of repeatability test indicated that the volume change of gingival tissue can be measured sensitively based on the method proposed in this paper, the error of point cloud registration method for oral OCT imaging is 1.93 ± 0.14 pixel (63.08 ± 4.52 µm) under 1σ level, and the measurement error of gingival tissue average thickness is -3.40 ± 21.85 µm (1σ) for oral OCT imaging. From this results, we concluded that if we make one measurement, there is a 68.2% chance that the measured value will be within ±21.85 µm from the real change in gingival thickness (or 99.7% chance within ±65.55 µm). In future clinical study that addresses the progression of gingival inflammation, we suggest a repeated measurement of not less than five (N≥5) in order to improve the accuracy of the measured thickness change.

Ethics

The imaging of subjects reported in this study using laboratory-built investigational device was conducted in accordance with a protocol approved by the Institutional Review Board of the University of Washington and informed consents were obtained from all subjects. The study followed the tenets of the Declaration of Helsinki and was conducted in compliance with the Health Insurance Portability and Accountability Act.

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Disclosures

The authors declare no conflicts of interest.
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