TEXTURAL ANALYSIS IN MEIBOMIAN GLAND IMAGE

AINUN KHALILAH MOHAMAD, BOptom. DEPARTMENT OF OPTOMETRY AND VISUAL SCIENCE, KULLIYYAH OF ALLIED HEALTH SCIENCES, INTERNATIONAL ISLAMIC UNIVERSITY MALAYSIA, JALAN SULTAN AHMAD SHAH, BANDAR INDERA MAHKOTA, 25200 KUANTAN, PAHANG.

ainun.khalilah@gmail.com

MOHD ZULFAEZAL BIN CHE AZEMIN, PhD (CORRESPONDING AUTHOR) DEPARTMENT OF OPTOMETRY AND VISUAL SCIENCE, KULLIYYAH OF ALLIED HEALTH SCIENCES, INTERNATIONAL ISLAMIC UNIVERSITY MALAYSIA, JALAN SULTAN AHMAD SHAH, BANDAR INDERA MAHKOTA, 25200 KUANTAN, PAHANG.

zulfaezal@iium.edu.my

ABSTRACT

Introduction: Meibomian gland is one of the anatomical eye structures to provide oily lipid layer to the anterior part of the eye. Textural analysis by using Mean Histogram Method was proposed as a method of estimating area of Meibomian Gland Loss (MGL). Objective: The main objective of this study was to objectively measure the meibomian gland image by using textural analysis. Methodology: This was a retrospective study consisted of 48 participants with readily available data of meibomian gland images captured by OCULUS Keratograph 5M. The data consisted of eye images from contact lens wearer (n=22) and non-contact lens wearer (n=26). The region of interests (ROIs) of total meibomian gland image were traced by one examiner by using ImageJ software. The other method adapted from previous project (Abdul Rahman N. F., 2015) named as Percentage Method. The method needed tracing to be done twice; total area of Meibomian gland and total area of MGL. The percent of MGL was calculated. Results: The repeatability of intra-grader was assessed by using intraclass correlation coefficients (ICC) that showed very high correlation based on Mean Histogram; right upper (ICC = 0.984, p < 0.001) and right lower eyelid (ICC = 0.983, p < 0.001). Spearman's rho was used to test the correlation between Percentage Method and Mean Histogram Method. On right upper eyelid Spearman's rho showed significant value (rho = -0.311, p = 0.031). Right lower eyelid showed very poor correlation (rho = -0.083, p = 0.573). The comparison of MGL between contact lens wearer (CW) and noncontact lens wearer (NCW) by using textural analysis method showed there was no statistical difference (P = 0.214 on upper eyelid, p = 0.634 on lower eyelid). Conclusion: Textural analysis based on Mean Histogram Method was shown repeatable. However, the measurement was not sensitive enough to discriminate between CW and NCW group.

KEYWORDS: Textural analysis, Meibomian gland image, Contact lens

INTRODUCTION

Nailon (2004) refers textural analysis as an area of imaging science that focuses on characteristics of image properties. Textural analysis is an actively area of study about thirty years ago (Zhang and Tan, 2002). Since then, they are several methods in textural analysis such as structural, statistical, model based and transformation method that have been developed to analyse image properties (Materka and Strzelecki, 1998). Some of the actively imaging area in medical that used textural analysis are microscopic image of biological tissues and image analysis techniques for detection changes in bone mineral density (BMD) (Materka and Strzelecki, 1998)

In the field of optometry, a research conducted by Calvo *et al.* (2010) analyses and classifies different tear lipid layer by using textural analysis based on different colours formed. Alonso-caneiro *et al.* (2013) had done a study on dry eye based on textural analysis. There is significant result in analysing dry eye based on videokerastocopy images and placido disc pattern with the help of textural analysis technique.

Lack of study found regarding the textural analysis and meibomian gland images. Meibomian gland is a sebaceous gland that works to provide oily lipid layer into anterior part of the eye that function to protect the aqueous layer of the tear film to evaporate. Meanwhile, meibography is an imaging area for the purpose of observing the morphology of the meibomian gland image (Wise *et al.*, 2012). Some disease that commonly found in meibomian gland area is meibomian gland dysfunction (MGD) and blepharitis. The dysfunctional of meibomian gland is due to terminal duct obstruction or changes in glandular secretion leading to dry eye disease (Pult and Riddle, 2013).

Currently, most clinicians only use slit lamp biomicroscopy to observe the area of defects in meibomian gland subjectively. Thus, this study proposed semi-automatic method in analysing the meibomian gland area with the help of OCULUS Keratograph 5M and also imaging software name as ImageJ

As stated by Tuceryan (1998), everyone can see texture but it is difficult to manually define and analyse the texture. Textural analysis using image processing software (ImageJ) is one of the method that can be used to objectively quantify the meibomian gland image. However, there is a lack of published literature to explore this strategy.

Meanwhile, Pult and Nichols (2012) reported that, there is no objective method in analysing the meibomian gland condition. Thus textural analysis is one of proposed methods that use semi-automatic procedure in determining area of meibomian gland simultaneously reducing human error.

The objective of this study is to measure the meibomian gland image using textural analysis. To assess the relationship between the method using textural analysis with the existing meibomian gland loss measurement, to evaluate the intra-grader repeatability of the textural analysis method in analysing meibomian gland image and to compare between meibomian gland images from contact lens and non-contact lens wearers by using textural analysis.

Textural analysis helps observer to reduce the steps involved in analysing the digital image. Comparing the previous study (Abdul Rahman N. F., 2015), the study used percentage between area of meibomian gland loss (MGL) over total area of meibomian gland. Previous study needs to trace the image area two times; area of MGL and total area of meibomian gland as opposed to the method that employed in this study. The method only use single image tracing that includes all the area of meibomian gland image.

The total area of meibomian gland image processed automatically by image processing software (ImageJ), able to minimize the time during analysis the image. The significant obtained will help the practitioner to practice using this method in the clinical as

one of objective method in analysing meibomian gland dysfunction (MGD). It is hypothesized that, there is no significant relationship between textural analysis and Meibomian gland loss measurement.

METHODOLOGY

This is a retrospective study. Readily available data from previous study done by Abdul Rahman N. F. (2015) were taken and analysed. The previous data constituted of both eyes from 60 participants of their upper and lower meibomian gland images. Informed consent was obtained from the subjects and approval from IIUM Research Ethics Committee (IREC) was granted in accordance with the principles laid down by the Declaration of Helsinki.

In this study, there were several loss of data from the previous study done. The missing data were searched again through the OCULUS Keratograph 5M which was a tool that had been used from the previous study to capture the meibomian gland area. The missing data were not able to be retrieved and the remaining data left came from 48 participant. The data consisted of eye images from contact lens wearer (N=22) and non-wearer (N=26). In total, 192 eye imaged were finally analysed.

The inclusion criteria's of participants in those data were:

- I. Age between 15-30 years old
- II. Wearing contact lens at least one year

ImageJ software as shown in Figure 1 is an image processing software which is publicly available.



Figure 1. ImageJ software

In order to measure correlation between two measurements used in this study. The study procedure consisted of two methods:

Mean Histogram Method

Mean Histogram Method is a methodology in the textural analysis that uses statistical based method. To process and recognize an image feature specifically in the meibomian gland image, this study used statistical technique to extract the information from the digital image. The statistical textural analysis applied in this study was done by calculating the number of light and dark pixels of an image.

It analysed spatial distribution of gray values in an image (Figure 2). The dark area represented area of meibomian gland loss and the light area represented area of meibomian gland present.



Figure 2. Mean histogram method

A statistical value was revealed and the result was taken from the histogram (Figure 3).



Figure 3. Statistical information extracted from meibomian gland image

The mean of cropped region was taken as the result of MGL. The mean calculated based on the formula:

Mean =
$$\frac{Sum \, of \, gray \, values \, of \, all \, pixels \, in \, ROI}{Total \, number \, all \, pixels \, in \, ROI}$$

Percentage method

The second method was based on procedures followed by previous study done by Abdul Rahman N.F., (2015). The region of interests (ROIs) in Meibomian gland image were traced twice (Figure 4).



Figure 4. Steps based on percentage calculation

First tracing constituted of total area of Meibomian gland image. Second tracing involved dark area Meibomian gland that represented the area of MGL. Both upper and lower eyelids were analysed and calculated. The percentage from two ROIs was calculated. The formula given to calculate the area of MGL is as follow:

Percentage of MGL =
$$\frac{Total area of MGL}{Total area of meibomian gland} X 100\%$$

Data were analysed using SPSS (Statistical Package for Social Science Software) version 12.0.1 for windows. The normality test used in this study was Shapiro-Wilk normality test. In general, our data were analysed using Intraclass Correlation (ICC), Spearman's rho correlation and Mann Whitney test.

RESULTS

The agreement for two measurements of one examiner, one week apart for Mean Histogram Method was analysed as depicted in Figure 5. The 95% limit of agreement for two measurements in same technique are +14 and -16 intensity level. There was no linear trend detected on the data (P > 0.05, $R^2 = 0.0106$), which indicated random variability. The figure shows good intra-grader agreement for Mean Histogram Method with only two points out of 48 (4.2%) outside the 95% confidence interval.



Figure 5. Bland Altman plot used to find repeatability of Mean Histogram Method (MHM) by one examiner on right lower eyelid, one week apart.

For right upper eyelid, there was a significant correlation between these Mean Histogram Method and Percentage Method (r = -0.311, P = 0.031). However, there is very weak correlation between these two methods (r = 0.035, p = 0.813) on right lower eyelid. The comparison between two groups of subjects was not significant (Z = -1.241, p = 0.214) on right upper eyelid and right lower eyelid (Z = -0.476, p = 0.634).

DISCUSSION

This study describes semi-automatic method in analysing the meibomian gland image. Textural analysis is one of the computerized techniques to extract information from medical imaging data. It transforms the information of an image into numerical value. To date, the previous research conducted on meibomian gland image analysis was by Arita *et al.*, (2013). Their study's methodology was based on statistical technique in which the software automatically detect and analyse the intensity levels in the area of meibomian gland. To further explore and to improve the processing speed of the image, this study proposed to perform a new method to analyse the image with freely available image processing software (ImageJ).

Previous project done by Abdul Rahman N. F. (2015) calculated the area of meibomian gland loss (MGL) by manually traced two Region of Interest (ROIs) that consist of total area of meibomian gland and total area of MGL. Then, the examiner needed to manually calculate the percentage of MGL on both upper and lower eyelid. This causes time consuming to get the result. The present study, the examiner only needed to locate and trace total area of meibomian gland. The high repeatability in intra-grader for this method shows the repeatability of measurements was independent of its time of measurement (Zhao *et al.*, 2015). The intra-grader repeatability test Mean Histogram Method showed very high correlation on both upper and lower eyelid.

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There is correlation between Percentage Method and Mean Histogram Method for right upper eyelid. Meanwhile, there is poor correlation on lower eyelids. The differences of significant value between upper and lower eyelid probably, the area of upper eyelid provide better contrast image to trace. Some of the meibomian gland image data from lower eyelids have low contrast and a lot of noise. Upper eyelid have better image because the amount and area of meibomian gland is greater than lower eyelid thus providing minimal error from inversion of eyelid during capturing the image (Driver and Lemp, 1996). Meanwhile, for lower eyelid the examiner gave some errors during capturing the image due to small area of lower tarsal plate. This is supported from Arita *et al.* (2013) in which the meibomian gland in lower eyelid is thicker than upper eyelid indicate the narrow space between them thus giving different calculation from automatic and manual method.

The poor correlation on lower eyelid might be due to the overestimation area of MGL from the low contrast image. The Mean Histogram Method automatically detects all dark pixels that consider as area of MGL which include the space area between meibomian gland in which that is not considered as area of MGL. In Percentage Method, the examiner includes the total area of MGL based on manual tracing whereby the image is traced without including the dark but non-MGL areas which may explain its ability to discriminate between CW and NCW in the previous study.

The comparison between area of MGL in contact lens wearer (CW) and non-contact lens wearer (NCW) on both upper and lower eyelids shows there was no significant difference between CW and NCW on right upper eyelid (p = 0.214) and right lower eyelid (p = 0.634). The result found on the present study was contradicted Abdul Rahman (2015) reported. The high differences of significance value between these two studies might indicate that the Mean Histogram Method has low sensitivity to analyse the area of MGL. Meanwhile, Clausi (2002) mentioned that, by analysing the image at gray level, the statistical analysis demonstrates a decrease in ability to classify the image.

Michael *et al.* (2016) also reported on low sensitivity in automatic grayscale value of histogram from ImageJ comparing to manual tracing utilizing Photoshop. Their study of repeatability and comparison of Photoshop and ImageJ for the grayscale analysis of muscle echogenicity found that there was error of echogenicity estimation by using grayscale value or histogram when comparing using the Photoshop method.

CONCLUSION

In conclusion, this study found that textural analysis based on Mean Histogram Method has relationship with Percentage Method on upper eyelid. However, there is no relationship between these two methods for lower eyelid. The Mean Histogram able to analyses the meibomian gland on upper eyelid only, but it is opposed to lower eyelid. The Mean Histogram for intra-grader repeatability is very high showing this method has high repeatability. There was no effect of area selecting during analysis of meibomian image between one examiner in one week apart for both upper and lower eyelid. However, this Mean Histogram Method is not sensitive enough to compare meibomian gland loss between contact lens wearer and non-contact lens wearer group.

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