Spatial Quantification of Morphological Changes in Retinal Pigment Epithelium

Haitao Huang[1], Micah Chrenek[2], John Nickerson[2], Hans Grossniklauss[2], Yi Jiang[1]

***Short Abstract* — Morphological changes in retinal pigment epithelium (RPE) is often associated with the disease progression of age-related macula degeneration (AMD). We applied different statistical methods to quantify the morphology of RPE in both mouse and human eyes as well as in simulated AMD eyes. Distinct pattern of second-order spatial properties reveal the RPE pattern signatures of AMD eyes. Classification of genotypes and ages by RPE cell morphometric measures suggests there is little difference in prediction rates in angular locations, but significant differences in radial locations.**

***Keywords* — retinal pigment epithelium, age-related macula degeneration, quantification, morphological changes.**

I. Introduction

RETINAL pigment epithelium (RPE) is a monolayer of cells key to the wellbeing of photoreceptor cells in eyes. Previous study has quantified the RPE morphological changes in AMD [1, 2]. In this study we further quantify such changes in spatial locations, which will help to understand whether AMD progresses as well as how RPE morphologies differ in individuals with different genotypes and ages during and after AMD. Developing a set of quantitative tools for this purpose will also have practical applications in the early diagnosis of AMD.

II. Methods

RPE images were obtained from mouse and human (donor) eye flatmounts, with RPE cell borders stained by anti-ZO-1. We developed a reproducible segmentation procedure to identify cells with ImageJ and represented 2D cell distribution by a spatial point process of cell centroids. We then studied the second-order properties to investigate the changing patterns of clustering in both experimental and simulated RPE [2]. Cell morphometric features (24 in total) including cell shape, area, etc. were extracted using CellProfiler [4]. K-nearest neighbor (KNN) algorithm, combined with leave-one-out cross validation, were applied to classify the genotype and age in C57BL/6J (wild type) and RD10 mice and to calculate the prediction accuracy in various spatial regions.

[1]Department of Mathematics and Statistics, Georgia State University.
[2]Department of Ophthomology, Emory University.

III. Results

Spatial analysis of simulated human RPE images showed that the oscillations in the variance stabilized K-function and pair correlation function (PCF) for normal RPEs gradually disappear as hexagonal cells stretch and distort. The AMD-like RPE patterns show an increased clustering between distance 100 and 200 (in pixels), indicative of the disordered RPE pattern in AMD. The same analysis on experimental human RPE images shows the same change from normal eyes to diseased eyes.

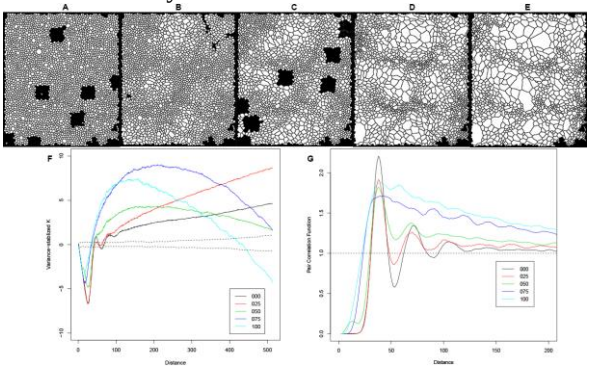

Figure 1. Simulated RPE tissue undergoing repeated clustered damage (black area: regions of cell apoptosis) and recovery from a normal RPE pattern.(A-E). F: Variance stabilized K function; G: Pair correction function.

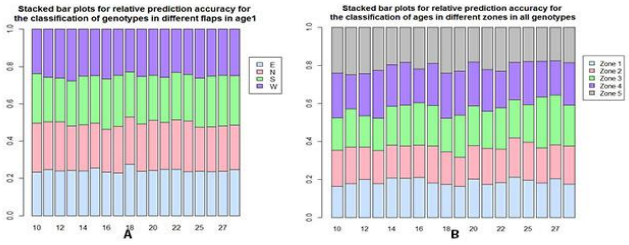

Figure 2. Stacked bar plots of relative prediction rates

Previous research hypothesized a certain difference exists in different spatial regions of RPE sheets. The KNN classification showed little difference in RPE pattern in our defined 4 flaps, yet an increasing accuracy rate with zone numbers [1]. Morphometric variables such as eccentricity consistently perform well in a classification (prediction rates over 90%). We also find the linear weighted combination of the morphometric variables as morphometric signatures that best distinguish the disease progression of AMD.

References

[1] Jiang Y,,et al. (2013). *IOVS* , 7274-7283.
[2] Jiang Y, et al. (2014). *Adv Exp Med Biol* 801, 601-7.
[3] Waller LA, et al. (2012). *Statist. Med.* 30, 2827–2841.
[4] http://www.cellprofiler.org