Reviewer's report

Title: Quantitative metric profiles capture three-dimensional temporospatial architecture to discriminate cellular functional states.

Version: 2 Date: 22 February 2011

Reviewer: Stephen Lockett

Reviewer's report:

The authors have revised the manuscript closely along the lines suggested by the reviewers, and therefore I do not have major compulsory revisions.

Minor Essential Revisions:

(1) The manuscript describes the application of cell graphs and tensor analysis to identify and quantify changes in the organization patterns of cells in 3D hydrogels and in histological tissue sections. This application of cell graphs and tensor analysis appears novel to me, but I am not an expert. Therefore the authors need to very clearly state what is novel in their application of this analysis method and what is not novel. In a similar vein, the authors need to clearly state which significant results they obtained are novel. For instance the similarity of metrics between histology and 3D hydrogels in figure 5 seems very significant and novel. If this is true, the manuscript needs to clearly state this.

(2) The abstract needs several improvements to increase clarity.

(2.1) As it stands, the opening sentence in the background of the abstract says nothing. A statement about how computational analysis of tissue structure reveals sub-visual differences in diseased or injured tissues would be an appropriate start.

(2.2) The term, "structural organization of the cell nuclei ...." is misleading. The analysis is not measuring internal features of cell nuclei, but is measuring contextual relationships between nuclei in tissue.

(2.3) Biological interactions are between cells, not between nuclei. The edges do not measure biological interactions, they simply indicate approximate adjacencies of cells using nuclei as surrogate markers for the cell positions.

(2.4) I would argue that no cell line is healthy. Terms such as non-transformed, non-cancerous are OK where appropriate. This comment applies to other places in the manuscript.

(2.5) The term, "number of central points" should not be used in the abstract, because it is not defined until later in the manuscript.

(2.6) The conclusion to the abstract states that the 3D hydrogel results can be used to improve diagnostic accuracy. While figure 5 shows some significant correlation between 3D hydrogel results and histology results in terms of the same discriminating metrics, it is not at all clear how one goes from this
agreement to an improvement in diagnosis.

(3) Segmentation of nuclei: How are touching nuclei dealt with?

(4) Discussion. It is an overstatement to say that cell lines that correspond to different degrees of tumorigeneity represent different stages of a cancer.

(5) Table 3, rows 2, 3 and 4: Provide a full definition of these clustering coefficients.

(6) Figures 3 and 4: Each graph could be reduced to single horizontal line where the x-axis is the Hotelling’s T-square value or metric influence. There is nothing added by plotting the same information on a diagonal line. Figure 3 and 4 can be merged into 1 figure.

(7) Figure 7: Use thicker edges so that one can see which edges are red in the clustering row.

Discretionary Revisions

(1) Could propose to do co-culture analysis of epithelial and stromal cells.

Level of interest: An article of importance in its field

Quality of written English: Acceptable

Statistical review: No, the manuscript does not need to be seen by a statistician.

Declaration of competing interests:

I declare that I have no competing interests.