Fractal dimension of U373 astrocytoma cells in DMEM or RPMI cultures

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Abstract

In order to characterize a possible difference in the organization of U373 astrocytoma cells under different culture medium (DMEM or RPMI), we have obtained the fractal dimension (FD) of cell cultures using HarFA image analysis in the whole range of thresholding conditions (http://www.fch.vutbr.cz/lectures/imagesci/). The obtained results showed a significant increase in the astrocytoma FD depending on the time culture but not on the growth medium. We may conclude that the increased cellular organization and complexity reached in astrocytoma cultures with time, deduced from FD, is not related to the culture medium.

1. Introduction

Fractal geometry is based on the observation that structures growing apparently according to stochastic processes are not really as disordered as they appear; thus, these structures may be characterized using fractal dimension (FD) as a quantitative parameter (Fernández and Jelinek, 2001). One of the advantages of fractal analysis is the ability to quantify the irregularity and complexity of objects. In this sense, a tissue has been described as a self-organizing cellular system with fractal dynamics, where an increase in the FD has been related to aggregation and cell expansion, and a decrease in the FD with cellular differentiation (Waliszewski and Konarski, 2001).

DMEM and RPMI are the habitual media used for U373 astrocytoma cell cultures. The aim of our work is to analyse the possible influence of the culture medium in the complex organization of astrocytoma cell cultures using the FD as a discriminative parameter.

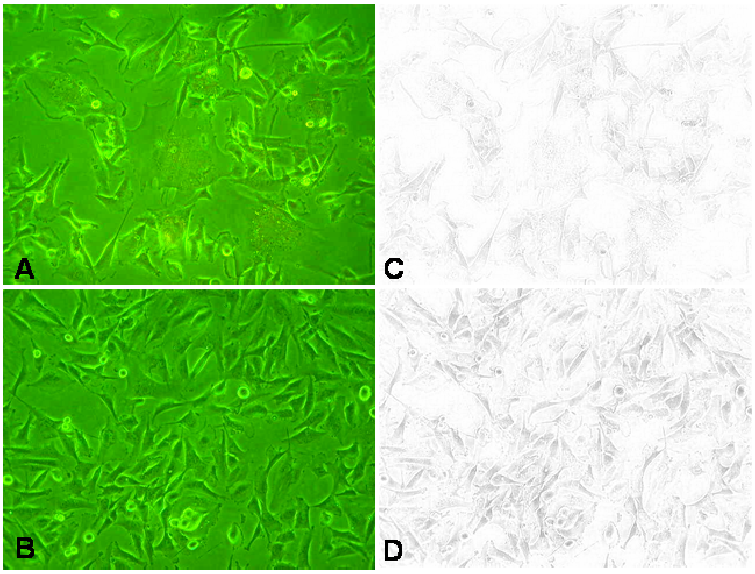

Figure 1 Astrocytoma cultures in RPMI medium at time 0h and 24 h, before (A, B) and after processing (C, D) with ImageJ, respectively.

Obtaining the fractal dimension from image analysis is not a trivial procedure because it depends on the particular image thresholding. To avoid a biased decision, we have selected the FD corresponding

to the slope change detected in the whole range of thresholding conditions, a very useful processing tool implemented in HarFA fractal analysis software.

2. Methods

U373 human astrocytoma cells were propagated in RPMI (with Glutamax) or DMEM (Gibco) media, both supplemented with 10 % (v/v) heat-inactivated fetal calf serum (Linus) and Penicillin-Streptomycin (Sigma). RGB images were taken 24 hours after seedling (time 0h) and twenty-four hours later (time 24h) for each experimental group ($n = 5$). Images were processed (RGB to 8-bit conversion and background subtraction) using ImageJ software (http://rsb.info.nih.gov/ij/) (*Figure 1*). After this, a FD analysis was carried out from the whole range of thresholding conditions using HarFA v4.9 software (http://www.fch.vutbr.cz/ lectures/imagesci/); we selected the FD corresponding to the slope change as indicated in *Figure 2*. Statgraphics Plus 5.1 was used as the statistical software.

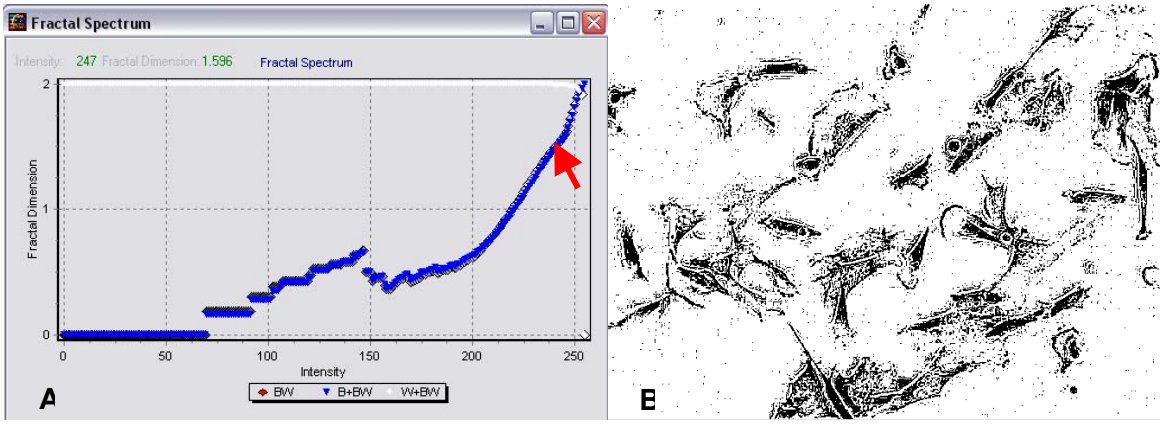

Figure 2 A: HarFA fractal spectrum; the red arrow indicates the change in the slope where the FD is selected. B: thresholding related to FD in A.

3. Results and Conclusion

Figure 3 shows the obtained individual values of FD corresponding to each experimental group. A significant increase was only detected when comparing the mean (±σ) FD related to time of culture (DMEM, 0h vs 24h: 1.54±0.11 vs 1.76±0.10, $p < 0.05$; RPMI 0h vs 24h: 1.60±0.07 vs 1.78±0.02, $p < 0.05$).

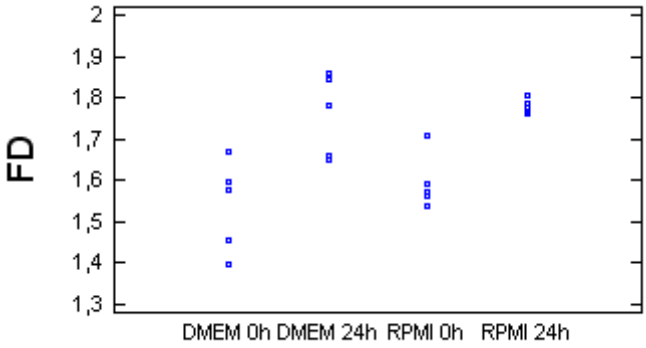

Figure 3 Fractal dimension (FD) for each image and group

From the obtained results, we conclude that the increased cellular organization and complexity reached in astrocytoma cultures with time, deduced from FD image analysis, is not related to the culture medium.

Acknowledgements

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References

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[2] Walisewski P, Konarski J. (2001) Tissue as a self-organizing system with fractal dynamics. Adv. Space Res., 28: 545-548.