Gradient field microscopy of unstained specimens: reply to comment

Taewoo Kim,[1] Shamira Sridharan,[2] and Gabriel Popescu[1],[*]

[1]*Quantitative Light Imaging Laboratory, Department of Electrical and Computer Engineering, Beckman Institute for Advanced Science & Technology, University of Illinois at Urbana-Champaign, Urbana, Illinois 61801, USA*
[2]*Quantitative Light Imaging Laboratory, Department of Bioengineering, Beckman Institute for Advanced Science & Technology, University of Illinois at Urbana-Champaign, Urbana, Illinois 61801, USA*
**gpopescu@illinois.edu*

Abstract: We reply to the comment written by Ferrari and Ayubi on our recent paper, Kim et al. [Opt. Express, **20**, 6737 (2012)]. We maintain that our use of Fourier filtering methods lead to valuable intrinsic contrast live cell imaging. Judging by their comments regarding the limit *sin(x)~x*, it seems that the authors overlooked Fig. 2 in our paper, where we show results with sinusoidal masks of different periods.

OCIS codes: (170.0180) Microscopy; (170.1530) Cell analysis; (170.1650) Coherence imaging.

References

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In the comment to our paper by Kim et al. [1], Ferrari and Ayubi claim that the presented gradient field microscopy (GFM) method is not substantially different from other Fourier-methods presented in references [2, 3]. We disagree and believe that the comment missed the point of our paper. We do not claim a new Fourier relationship, which, of course is a classic result. The point of our paper is that, using filtering methods based on known properties of the Fourier transform, leads to new intrinsic contrast approaches for cell and tissue imaging. In particular, we report a DIC-type microscope that does not require specialized polarized illumination or moving parts. To our knowledge, this type of microscope has not been reported before.

Of course, a derivative is obtained in the limit when the transverse shift vanishes. However, in practice, we always deal with differentiations over finite distances (for one thing the camera pixel itself is finite in size). For example, traditional DIC generates the gradient of the phase, but the transverse shift used is half the diffraction spot (definitely non-zero). We demonstrate the capability of using sinusoidal filters of various periods to gain control over this shift. Figure 2 in our paper [1], which apparently the authors overlooked, illustrates these results. GFM introduces various amounts of shift at the image plane without moving physical parts, which adds important versatility when imaging live cells and tissues. Importantly, we have recently found that GFM can be used to study unstained biopsies [4].

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