Rotation and focal depth fusion of DIC microscopic images

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This work proposes a new DIC image fusion method that is well suited for merging angle information and extends the field of focus of DIC microscopic images. The proposed method only fuses information specifically linked to the epidermal cell layer of Arabidopsis leafs, thus avoiding clutter related to underlying cell layers.

I. INTRODUCTION

Differential interference contrast (DIC) microscopy is a type of optical microscopy that enhances the contrast in the image based on interferometry. This high contrast allows for the visualization of transparent cell walls, without requiring any staining. The major drawback of DIC microscopy is that it shows high contrast in one direction, the shear direction, but practically no contrast to the perpendicular direction [1]. While this is typically not an issue for a human operator, it seriously hampers the use of computer vision methods. Furthermore, in the leaf samples not all cells are located within the same focal plane. To overcome both the directional contrast and out-of-focus cells, we propose a novel fusion method that merges images with different shear directions and different focal depths.

II. DIC FUSION

The proposed method works in two steps: first, images with different shear directions are merged, in a second step, the method fuses information coming from different focal depths. Just as in [2] we perform the fusion not in the image domain, but in a specific feature space. In contrast to common fusion methods, which use generic features, such as wavelets or shearlets, we propose to use specific edge features based on the average profile of a DIC edge. The added value of such dedicated edge features is thoroughly discussed in [1,3]. Rotation fusion becomes then simple in this feature space, e.g. by applying a maximum operator [2].

For the fusion of different focal depths, the same edge features are used. By fusing feature information from different focal depths, there is a risk of including information from non-epidermal cells, i.e. cells that are not part of the cell layer of interest, but which lie just beneath the epidermis layer. In order to avoid clutter from other cell layers, we propose a novel depth fusion approach. This approach first estimates a depth map for the cell layer of interest. Based on this depth

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map, only feature responses at the right depth are merged into a single image. Feature responses coming from cell layers of no interest, typically occur at a different depth than the ones from the estimated depth map. As such, features of underlying cell layers are not merged within the final image.

The depth map is estimated based on the detection of small regions that have a high feature response in one of the different focal images. The actual depth of each of these regions corresponds to the index of the image that has the highest feature response for the region. Finally, these region depths are used to calculate a depth map for the full image. This map is calculated based on polyharmonic spline interpolation. This depth map allows us to select the exact layer of interest, and only feature responses from this depth layer are selected for the final image.

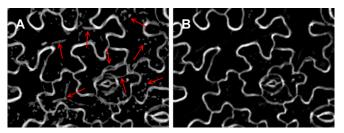


Figure 1 Results of DIC edge fusion: A) state-of-the-art DIC rotation and depth fusion [1], B) the proposed rotation and depth fusion

III. EXPERIMENTAL RESULTS

Fig. 1 shows an example of two methods for the fusion of the edge responses. Classical extended field of depth results in a fused image cluttered with blobs and false edges coming from underlying cell layers. Fig. 1.A shows such an example, where clutter from other cell layers are indicated by red arrows. Fig. 1.B shows the fusion result of our proposed method. This method shows edges that are all in focus, without adding false edges or clutter originating from underlying cell layers. Avoiding edges from irrelevant cells in the fused image will facilitate further post-processing and analysis of the epidermis cells.

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