Accurate Segmentation of Single Isolated Human Insulin Crystals for In-Situ Microscopy

G. Martinez[1], P. Lindner[2], A. Bluma[2], and T. Scheper[2]

[1]Image Processing and Computer Vision
Research Laboratory (IPCV-LAB), Universidad de Costa Rica

[2]Institute of Technical Chemie (TCI), Leibniz Universität Hannover,
Hannover, Germany

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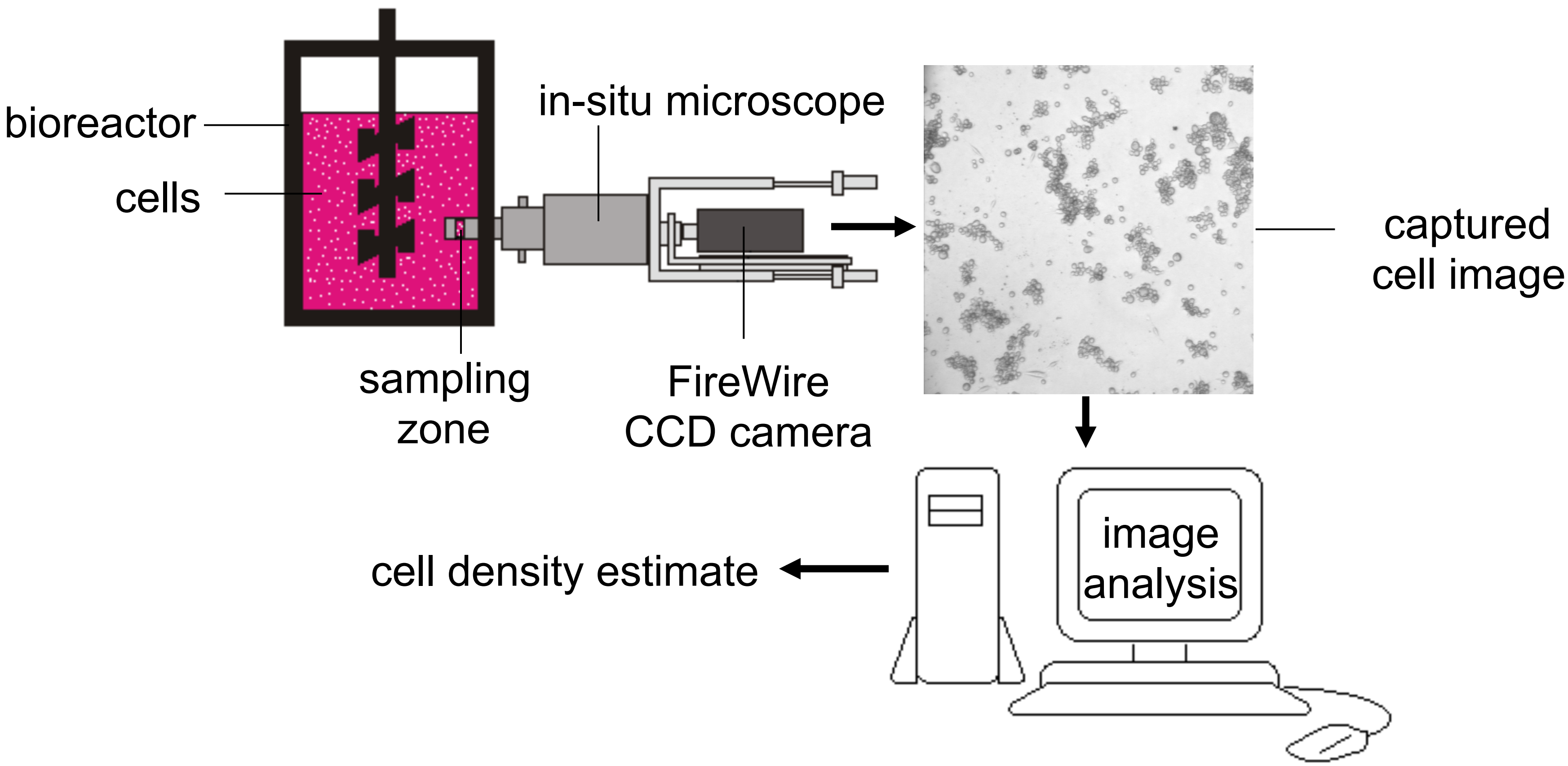
Overview

- Introduction
- Problem
- Approach
- Algorithm
- Experimental results
- Summary

Introduction

On-line automatic cell density estimation with no risk of culture contamination

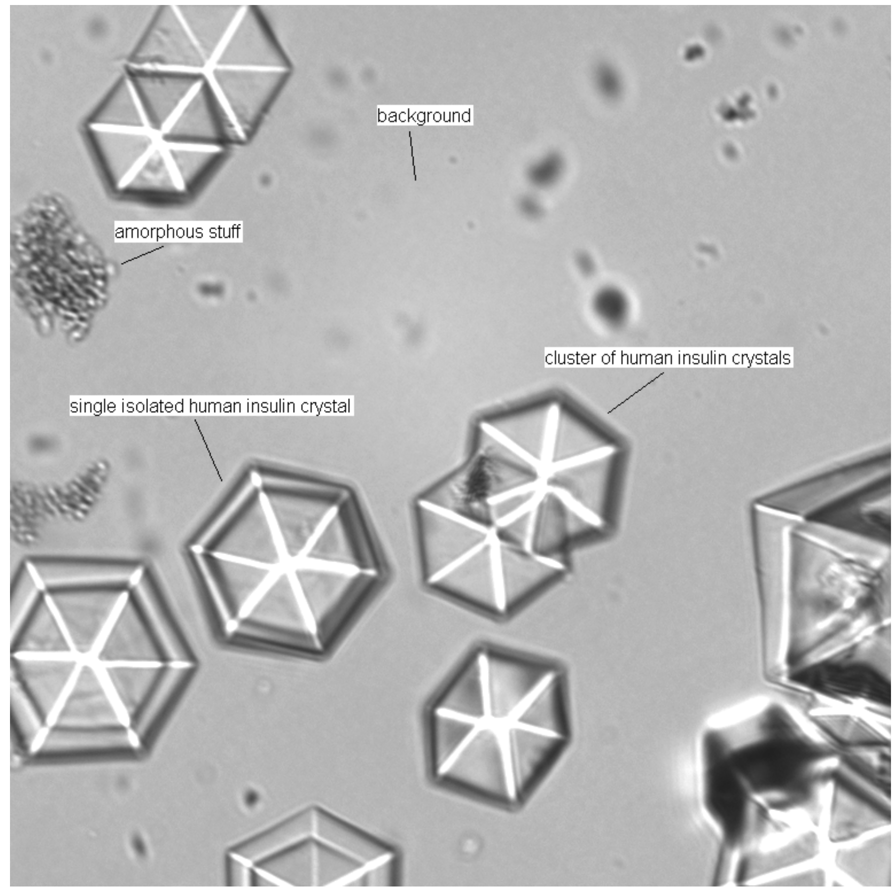


Introduction

As an alternative application, we are using the in-situ microscope for on-line analysis of human insulin crystallization processes

Images of first experiments show:

- Homogeneous background
- 4 different classes of foreground regions C_n, n=0,…3: single crystals (C_0), crystal clusters (C_1), amorphous stuff (C_2) and mixed regions (C_3)



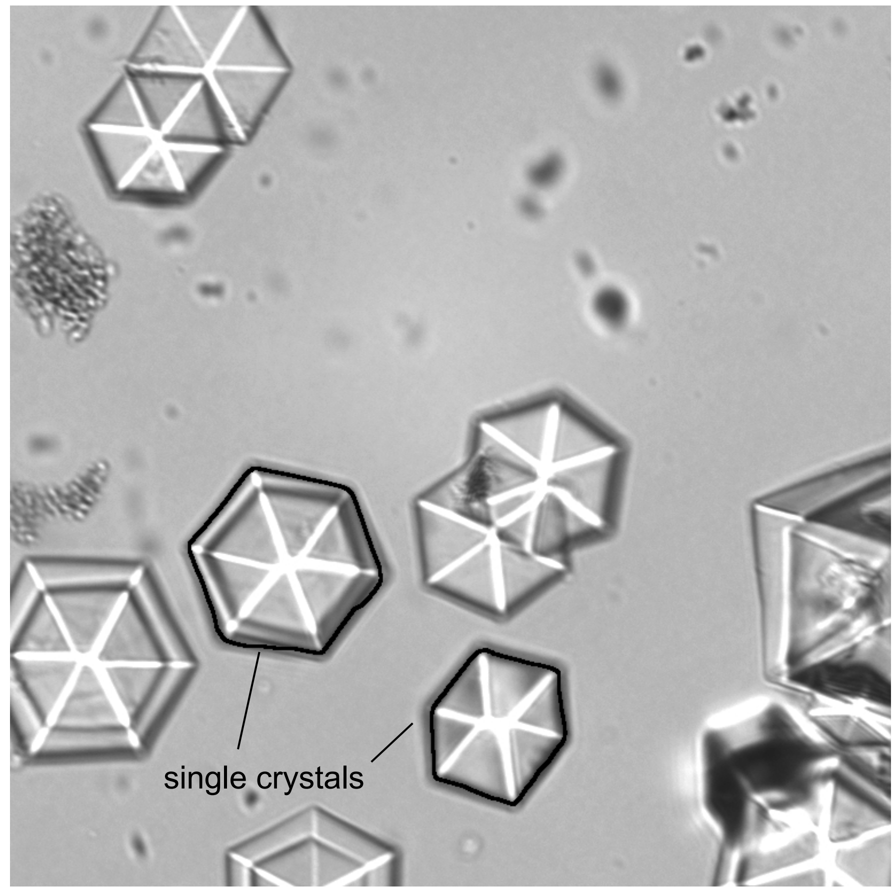
Human insulin crystal image captured by an in-situ microscope

Problem

Currently, we are very interested on the development of algorithms for accurately finding the image regions of the single crystals

Problem:

How do you accurately find them?

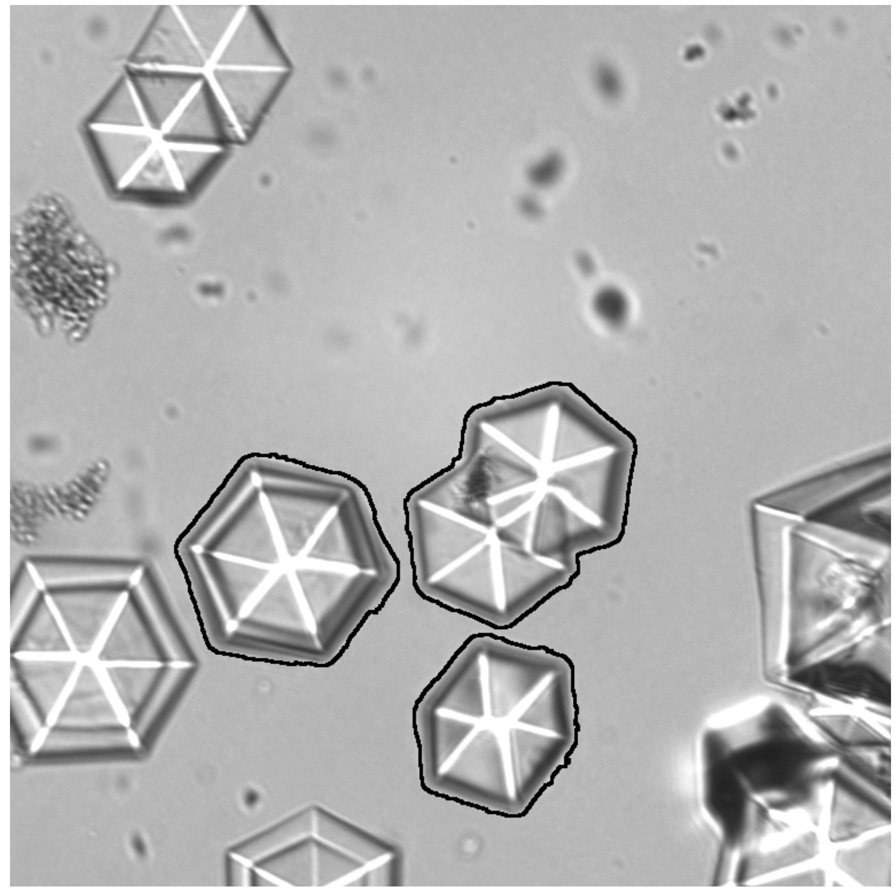


Three-Stage Approach

- First, all the foreground regions are segmented

  For foreground segmentation, the local intensity variance of the pixels is evaluated by using a threshold based rule

    The pixels belonging to the foreground regions will be those, whose local intensity variance is greater than a threshold th



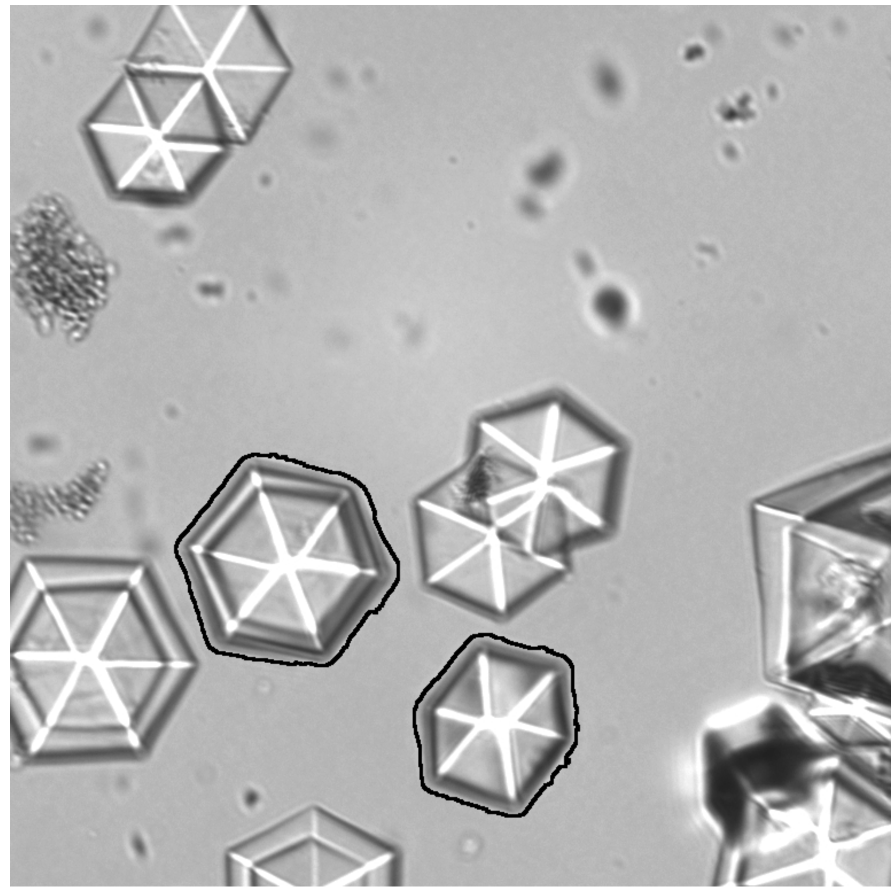
Segmented foreground regions

Three-Stage Approach

- Second, the single crystal regions are detected among the previously segmented foreground regions

  For detection, the shape of the segmented foreground regions is evaluated by using a single nearest prototype rule

    An arbitrary foreground region will be detected as a single crystal region, if its shape is much closer to the shape prototype of the single crystals than to the shape prototypes of the other classes of foreground regions

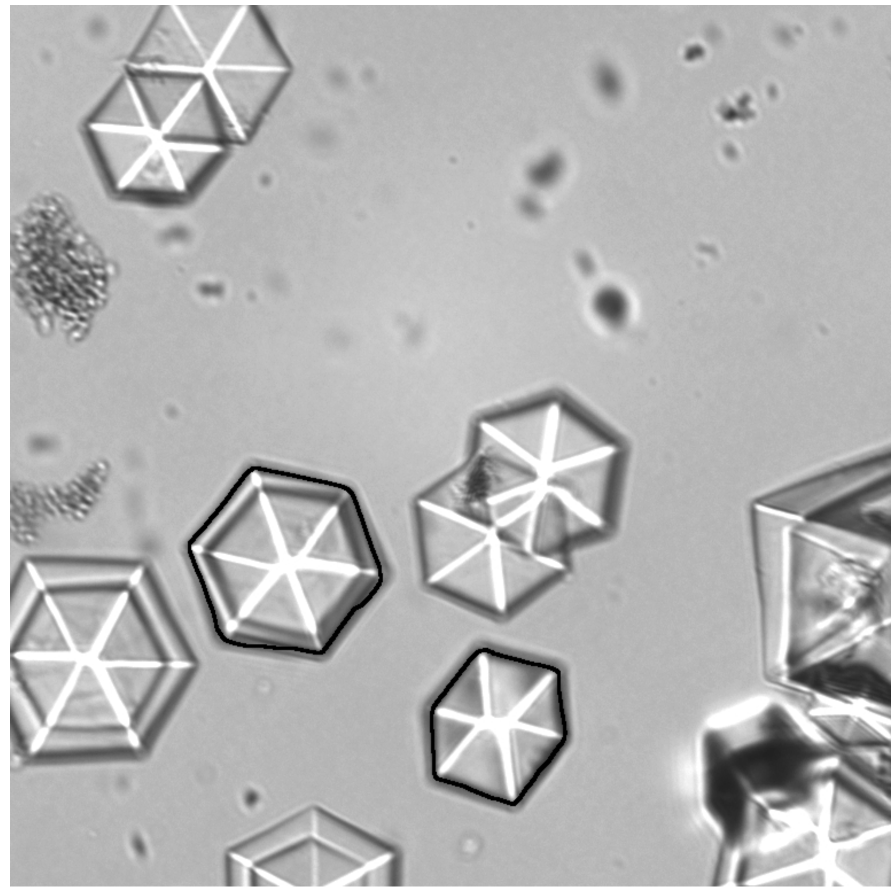


Detected single crystal regions

Three-Stage Approach

- Third, the contour accuracy of the detected single crystal regions are improved

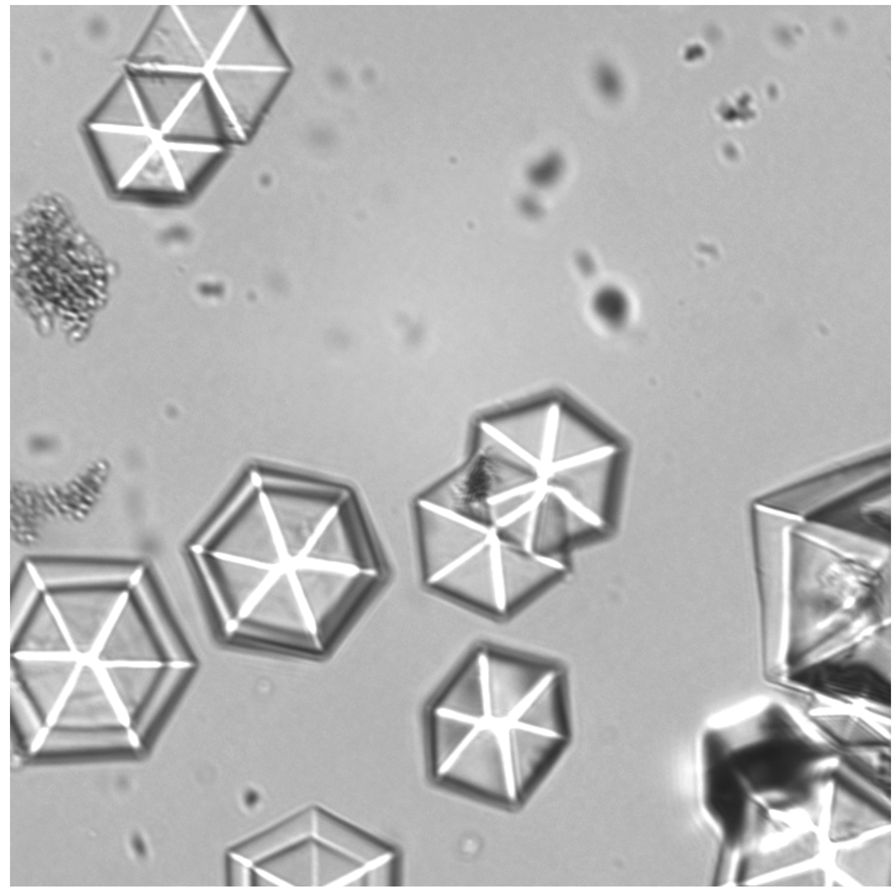
  Each region contour point is moved toward the region center of gravity, until a minimum on intensity and contour derivatives is achieved



Detected single crystal regions with improved contour accuracy

Algorithm

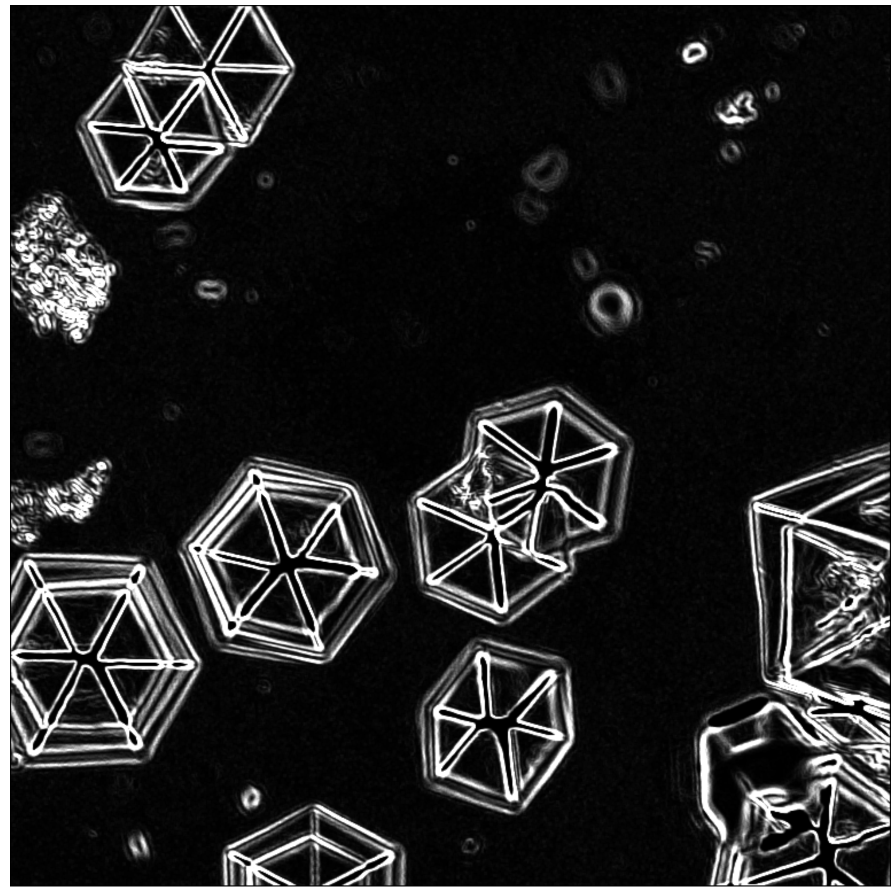
1) Capture an intensity image I



Intensity image I

Algorithm

2) Estimate the local intensity variance at each pixel position



Variance image V

Algorithm

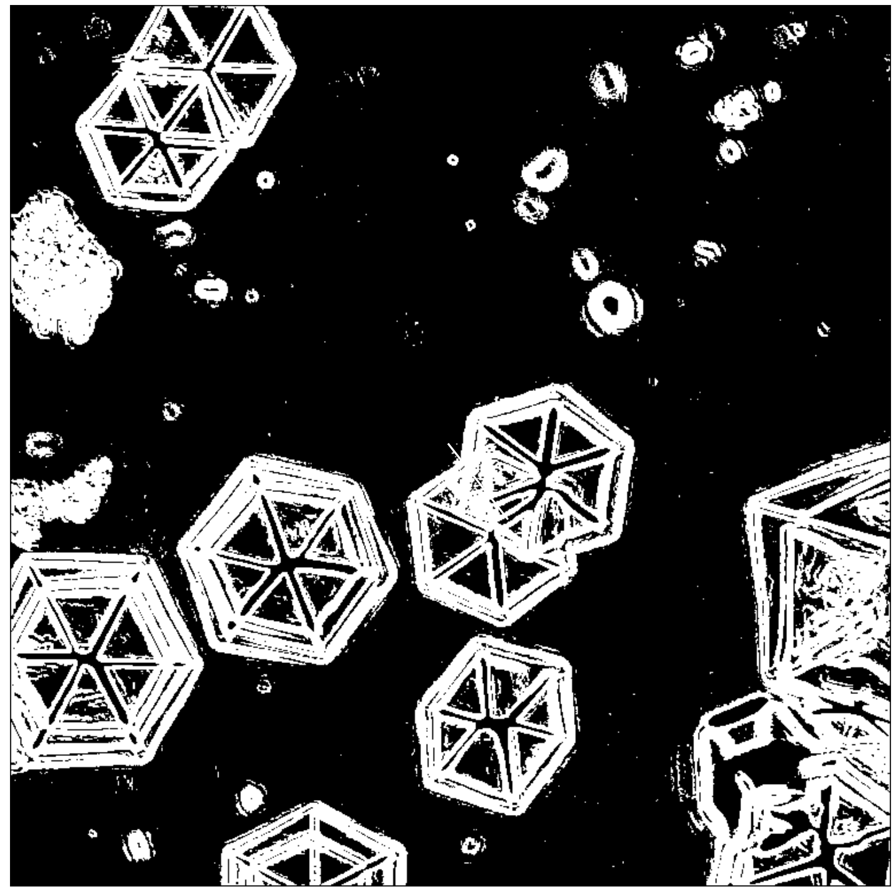
3) Threshold the variance image V

   $V_i > th$: foreground (white)
   $V_i \leq th$: background (black)

where $th = m_1 + 4\sigma_1$

σ_1: standard deviation of the variance values at the background

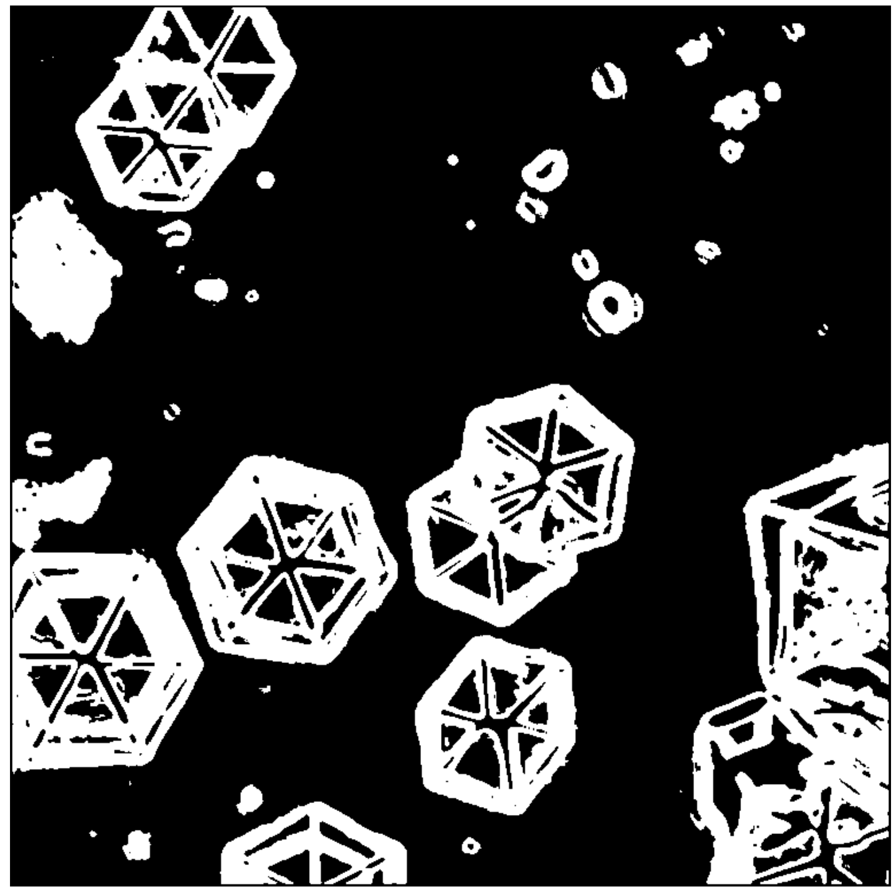
m_1: mean of the variance values at the background



Thresholded image V_{th}

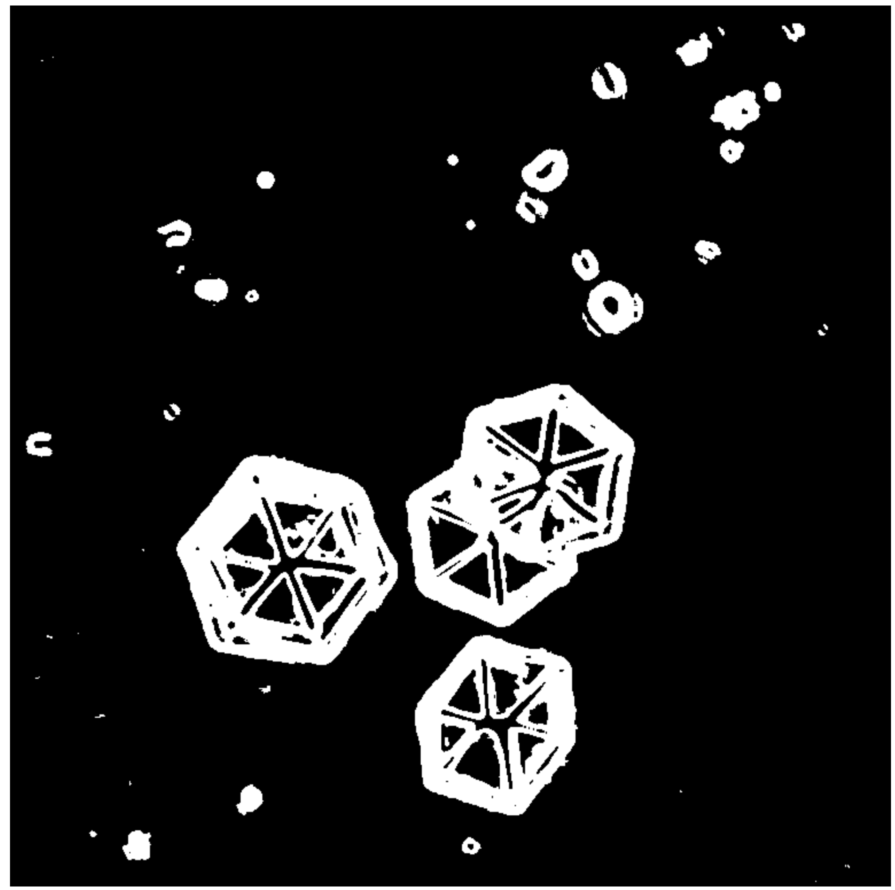
Algorithm

4) Eliminate isolated white pixels by applying a 5x5 median filter



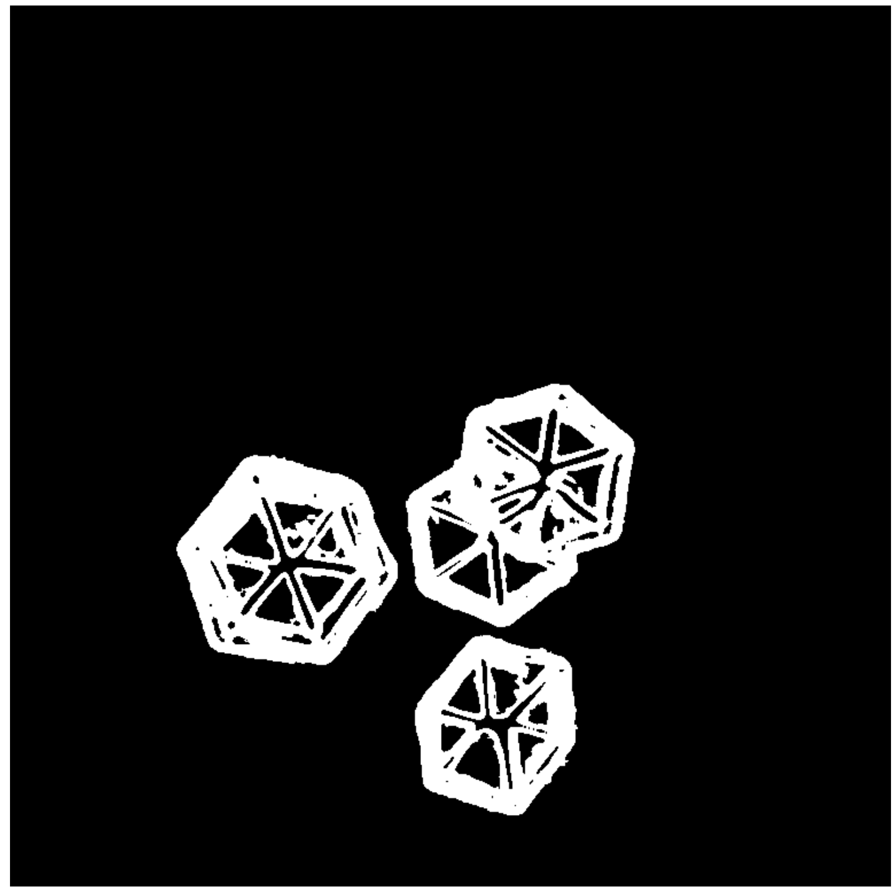
Algorithm

5) Eliminate any white region touching any image border



Algorithm

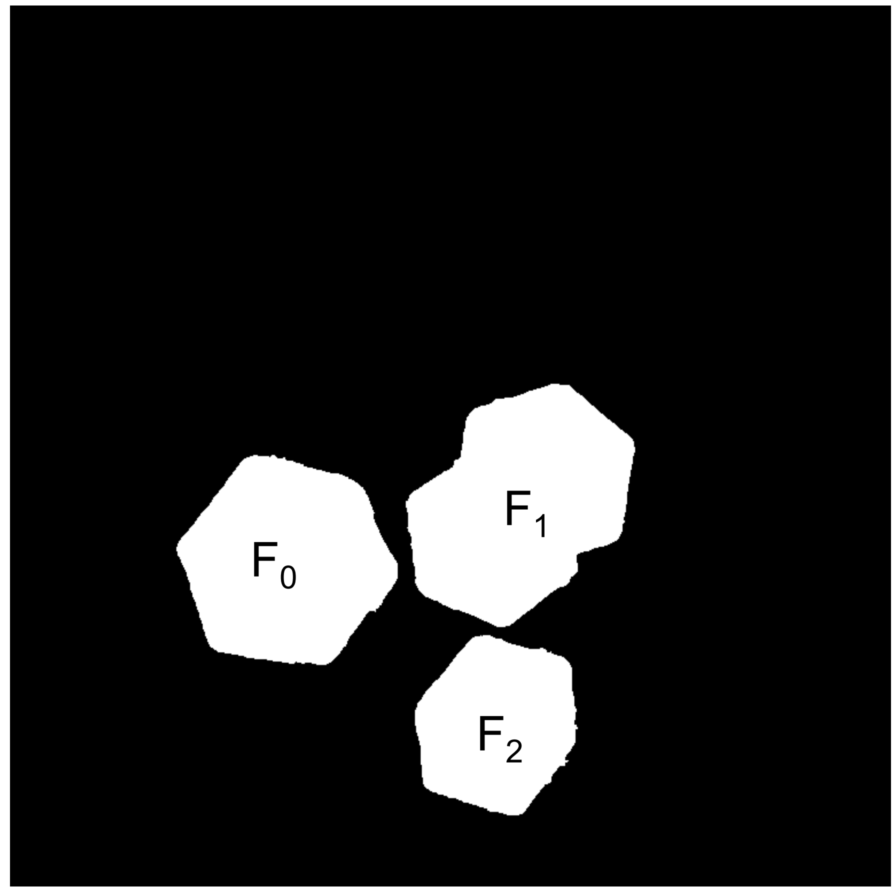
6) Eliminate white regions whose image area is less than 0.09% of the total image area



Algorithm

7) Eliminate black holes inside white regions

   The remaining white regions represent the segmented foreground regions F_s, s=0, …,S-1

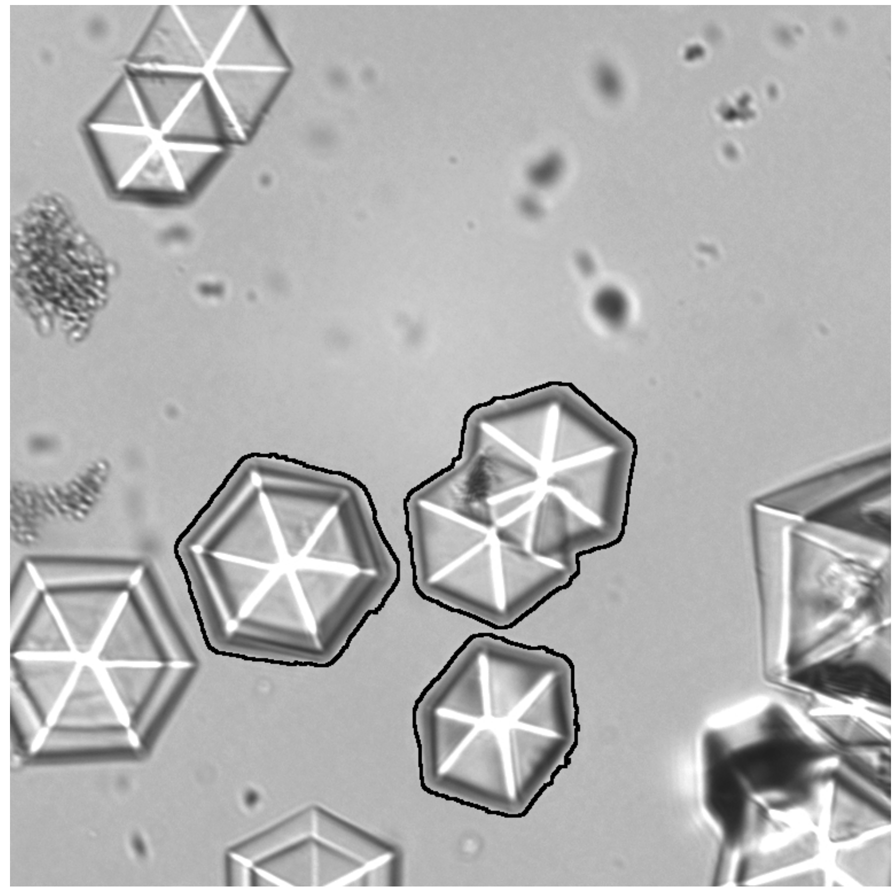


Segmented foreground regions

F_s, s=0, 1, 2

Algorithm

8) Compute for each segmented foreground region F_s a 7-dimensional vector of rotation, translation and scale invariant shape characteristics $C_s(k)$, k=0, …, 6, according to M.-K. Hu



Algorithm

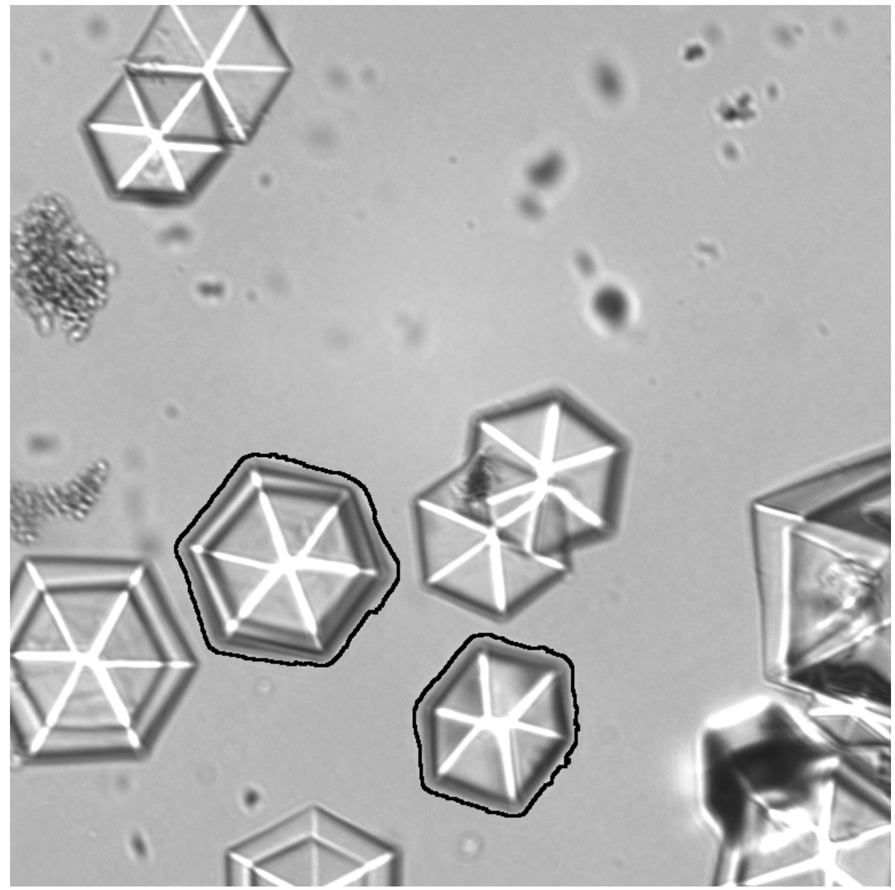
9) Detect an arbitrary segmented foreground region F_s as the region of a single crystal if its shape vector C_s is much closer to the shape vector prototype P_0 of the single crystals than to the shape vector prototypes P_1, P_2 and P_3 of the other classes of foreground regions

$$\left\| C_s - P_0 \right\| \leq \left\| C_s - P_n \right\|, \forall \; n = 0,..,3$$

where

$$\left\| C_s - P_n \right\| = \sqrt{\sum_{k=0}^{6} \left(P_n(k) - C_s(k) \right)^2}$$

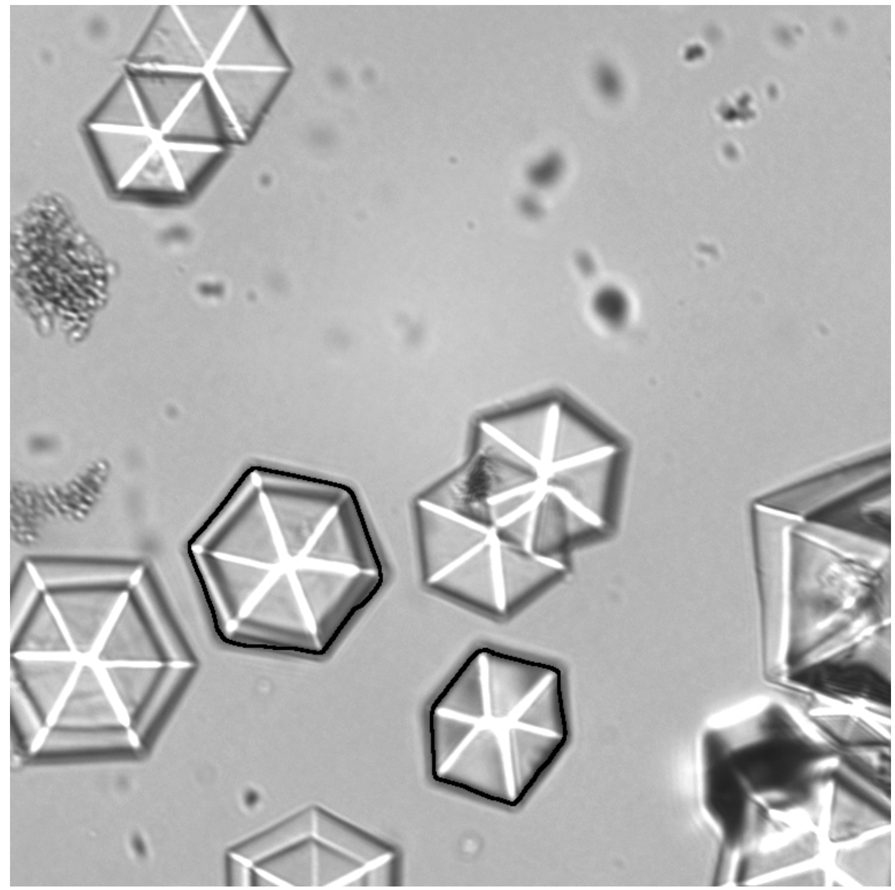
The shape vector prototypes are computed a priori from a training set of images



Detected single crystals

Algorithm

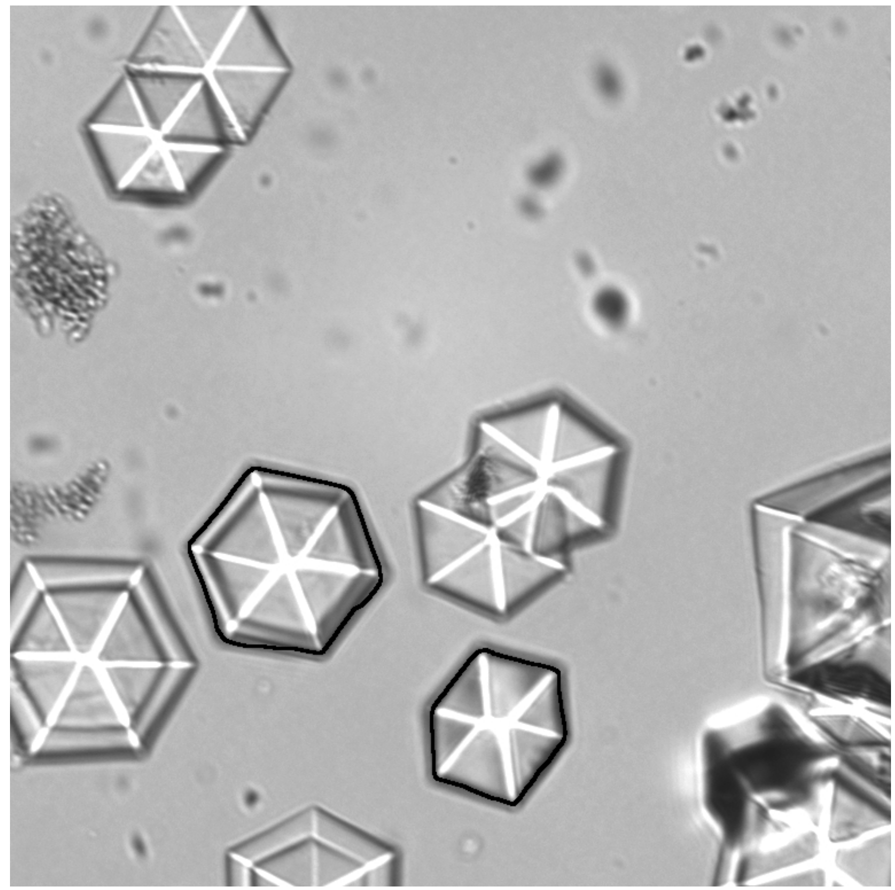
10) Move each region contour point along the line segment, which goes from the region contour point to the region center of gravity, to that image position α, where the weighted sum of the intensity value and the values of the first and the second region contour derivatives becomes minimal



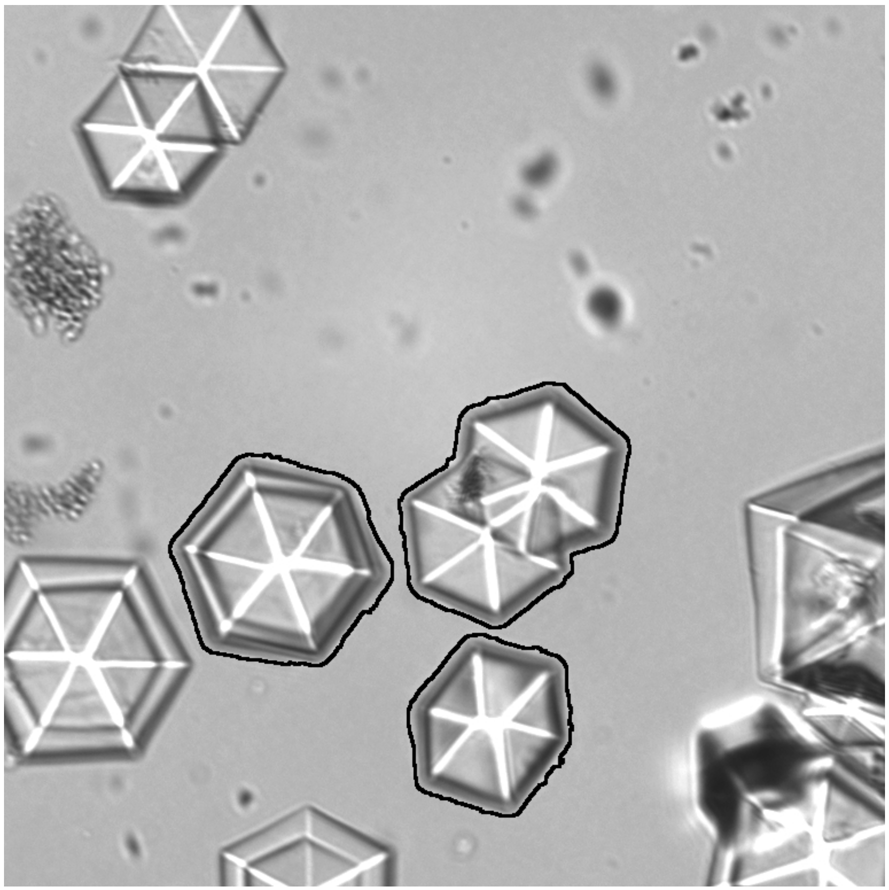
Detected single crystals
with improved contour accuracy

Experimental Results

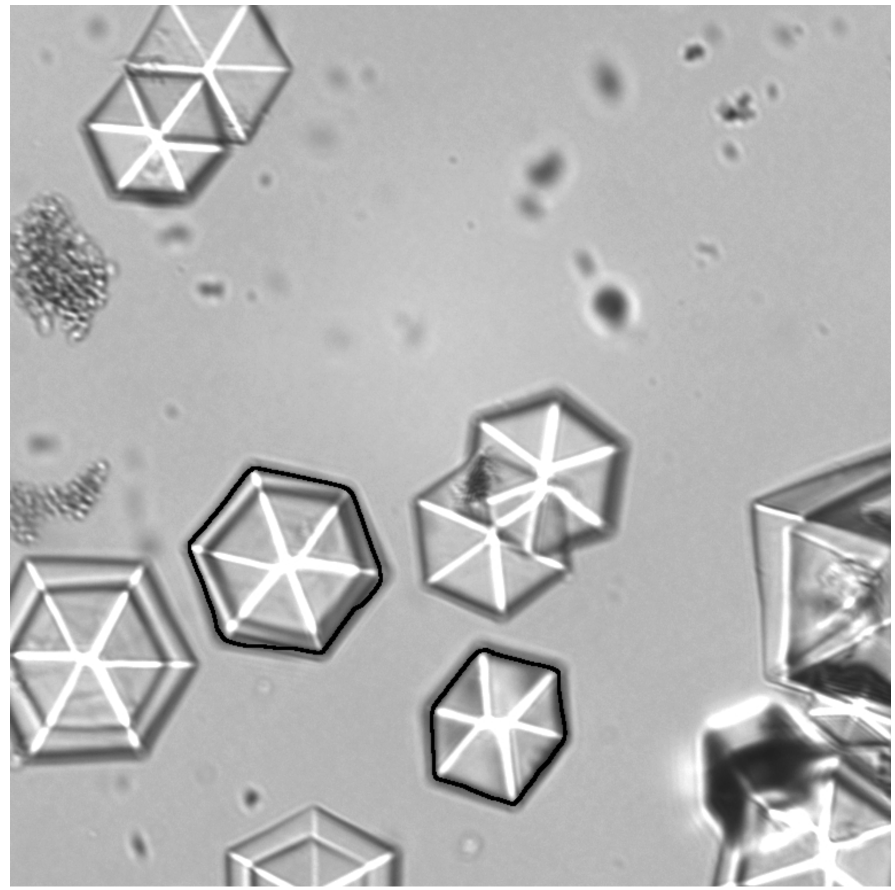
- Implemented in C under Windows XP
- Tested with 60 real images
- Intel Core Duo CPU at 2.2 GHz and 2 GB RAM
- Average processing time of 0.4 sec/image
- Contour accuracy improvement of 80%



Experimental Results

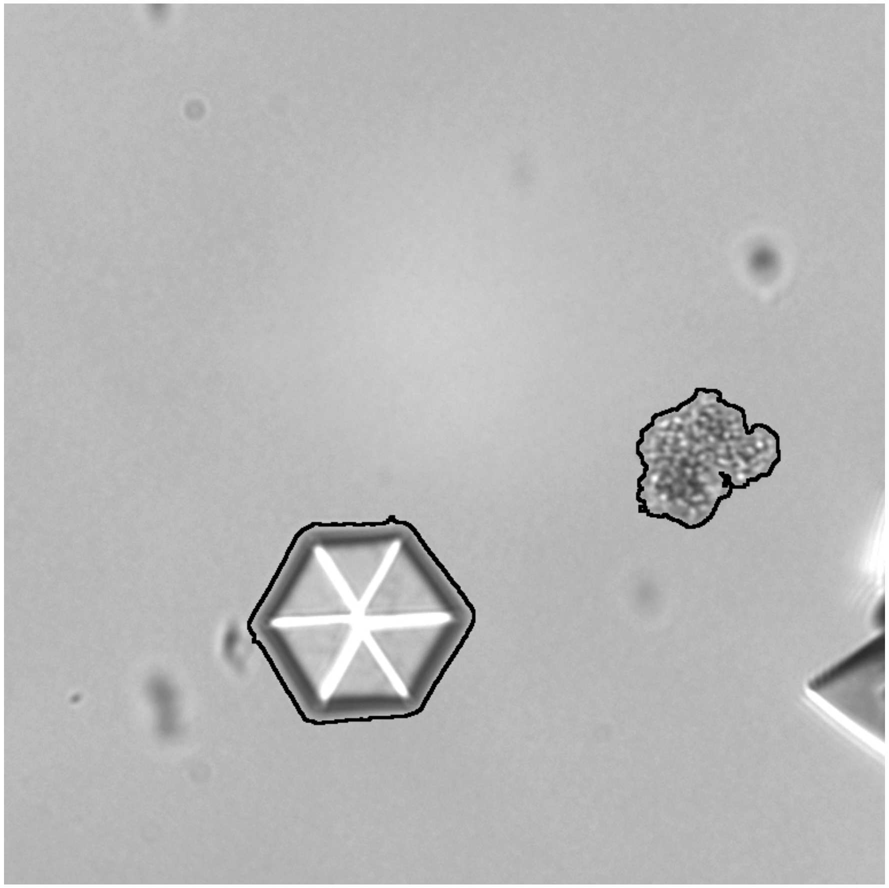


Segmented foreground regions

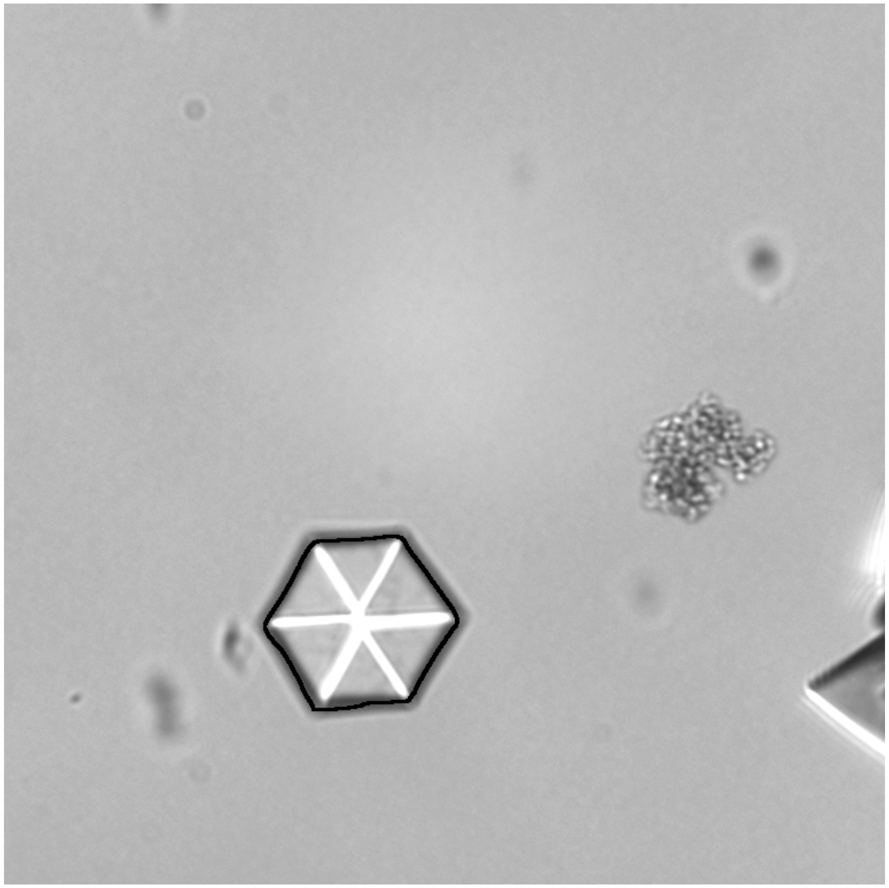


Detected single crystal regions with improved contour accuracy

Experimental Results

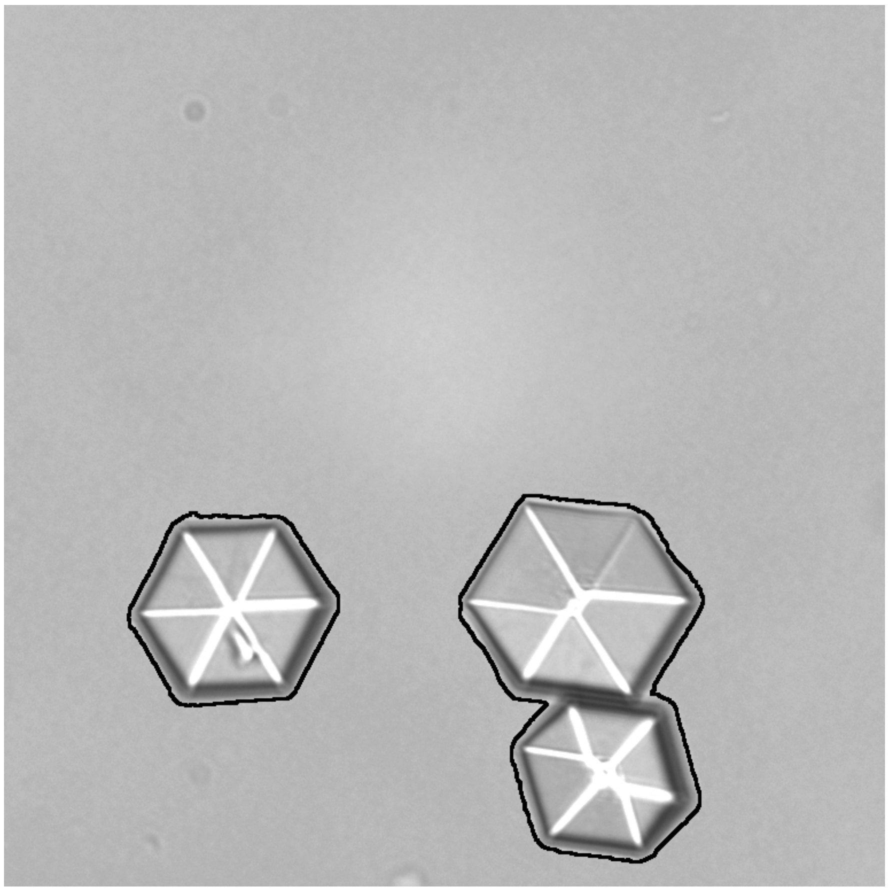


Segmented foreground regions

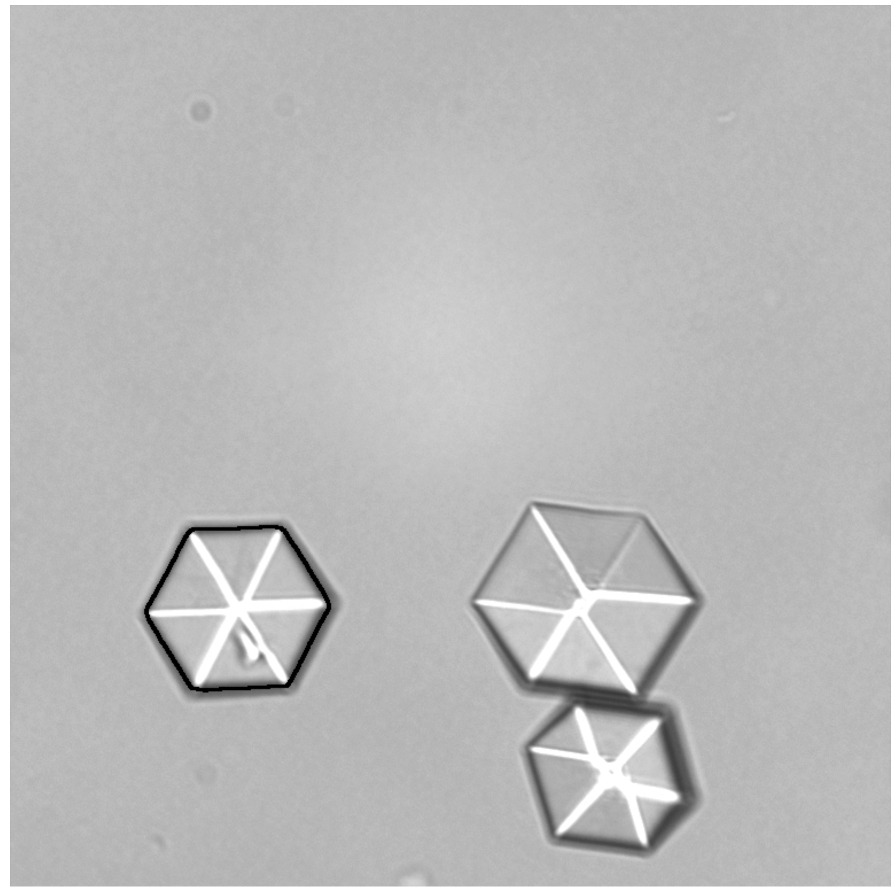


Detected single crystal regions with improved contour accuracy

Experimental Results



Segmented foreground regions



Detected single crystal regions with improved contour accuracy

Summary

- First, the foreground regions are segmented by using a global threshold technique. Then, the single crystal regions are detected by using a single nearest prototype detection technique. Finally, the contours of the detected single crystal regions are improved by using an active contour technique.
- Tested with 60 real images captured by the in-situ microscope
- Average processing time of 0.4 sec/image
- Contour accuracy improvement of 80%

Thank you!

Contact Information

Prof. Geovanni Martínez, PhD.
Image Processing and Computer Vision Research Lab. (IPCV-LAB)
Escuela de Ingeniería Eléctrica
Código postal 2060
Universidad de Costa Rica
San José, Costa Rica

Tel: +506 2511 3864
Fax: +506 2511 3920
E-mail: gmartin@eie.ucr.ac.cr
www: http://ipcv-lab.eie.ucr.ac.cr