

Decomposition of Overlapping and Touchingm-Fish Chromosomes Using Image Process Techniques

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ABSTRACT

Automatically segmenting partially obscured and touching objects is an extremely challenging task. Chromosome imaging may be impacted by partial blockage and chromosomal contact. This is one of the primary obstacles to automating the analysis. Many segmentation (decomposition) approaches have been developed for typical banded chromosomal images. With differing degrees of success, some of these strategies only address touching situations, while others address both. Most techniques employ simply the skeleton, convex hulls, and curvature of chromosomal clusters as geometry information. The geometry-based methods only look at the form of the boundary of a chromosomal cluster. Even while the border shape provides a wealth of information on the cluster formation, it is often insufficient in certain cases, such as when two chromosomes touch by their short or long sides to form a long or thick chromosome. These touching cases are easily recognised when the pixelmemberships are displayed by two distinct hues, like in M-FISH.

1. INTRODUCTION

The pixel classification accuracy is high, the color information itself may be sufficient for the chromosome segmentation for most cases. Schwartzkopf et al. proposed a maximum likelihood decomposition method using the pixel classification results and chromosome size for M-FISH images. Authors compared their results to that of commercially available software (Cytovision), and reported that much better results were achieved for touching cases and less reliable results for overlapping cases. When only the colors are used, touching or overlaps of the same type of chromosomes cannot be segmented, and the segmentation accuracy heavily relies on the initial pixel classification accuracy.

Thus the both geometry and pixel classification results have to be merged in order to achieve better segmentation results. In this chapter, we present a novel decomposition method for overlapping and touching chromosomes that utilizes the geometry of a cluster, pixel classification results, and chromosome sizes. We also introduce basic elements of overlap and touching cases. These basic elements yield hypotheses of possible overlapping and/or touching cases. Given a cluster, multiple hypotheses are evaluated, and the most likely hypothesis is chosen as the correct decomposition.

1.1 G-banded Chromosome Decomposition

Ji et al. had developed a simple but effective method to segment touching chromosomes based on two hypotheses: (a) at points where chromosomes touch, the optical density is relatively low; (b) where chromosomes touch, the cluster boundary tends to form an acute angle. Based on these ideas, touching chromosomes were effectively segmented. This algorithm is implemented in one of the current commercially available karyotyping systems. Some of the results are shown in Figure 1.5. As shown in the figure, overlapping chromosomes are not segmented.



Result 1



Result 2



Final Result

Figure 1.1: Separation results of Ji's method

By examining the concave locations on the boundary, Agam and Dinstein created a technique that can deal with both touching and overlapping chromosomes. Pairs of parallel lines are kept as legitimate cut lines until all concave points have been connected, as seen in Figure 1.2. Chromosomes were fitted with a polygon that is contracted and bent at one point. Out of all the conceivable combinations, the one that met the best requirements was selected as the appropriate separation. Figure 1.3 displays three alternative configurations for a cluster of three interacting chromosomes. When a chromosome fails to meet a specific requirement for fitting the polygon, a rectangle is depicted. Twenty-five carefully chosen photos that were appropriate for the analysis were used to test the created procedure. For two, three, and more than four chromosomal clusters, the corresponding accuracy rates were 88%, 68%, and 63%.

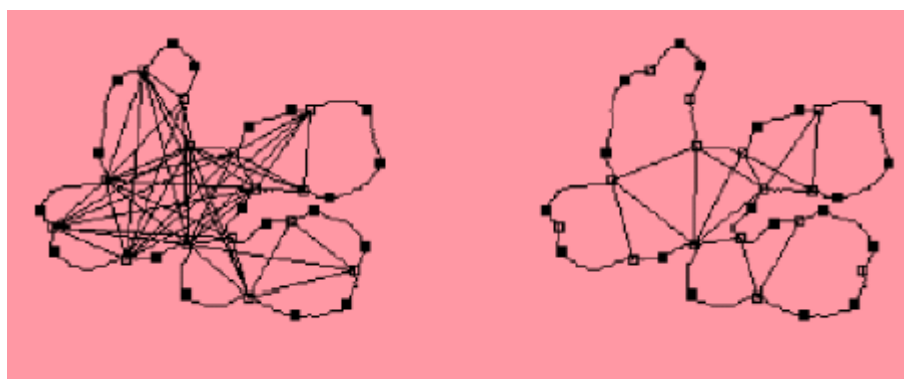


Figure 1.2: Possible separation lines of Agam and Dinstein's method

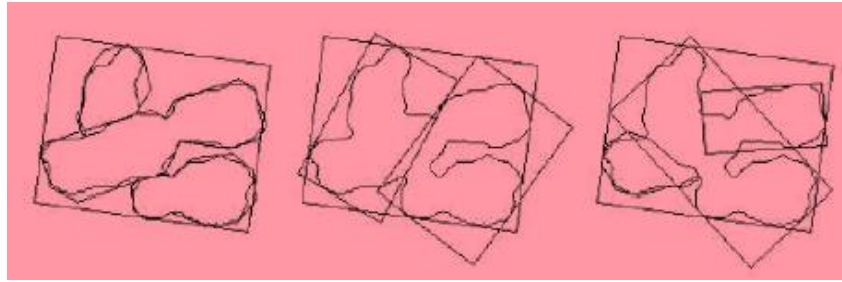


Figure 1.3: Several hypotheses for a cluster of three chromosomes

1.2 M-FISH Chromosome Decomposition

Schwartzkopf et al. used the colour information in a maximum likelihood framework to create a combined pixel classification and segmentation approach that can handle overlapping and contacting chromosomes because the pixel membership information is accessible for M-FISH pictures. A 17x17 majority filter was used to rectify minor misclassifications following the initial pixel classification using a 6-feature, 24-class maximum-likelihood classifier. To maximise the overall likelihood in terms of chromosomal size and pixel membership, touching and overlapping chromosomes were grouped together. In Figure 1.4, a sample result is displayed. A touching example that appeared to be a lengthy chromosome and was impossible to segment using the commercial Cytovision program was successfully segmented using Schwartzkopf's approach, as seen in Figure 1.5 (a).

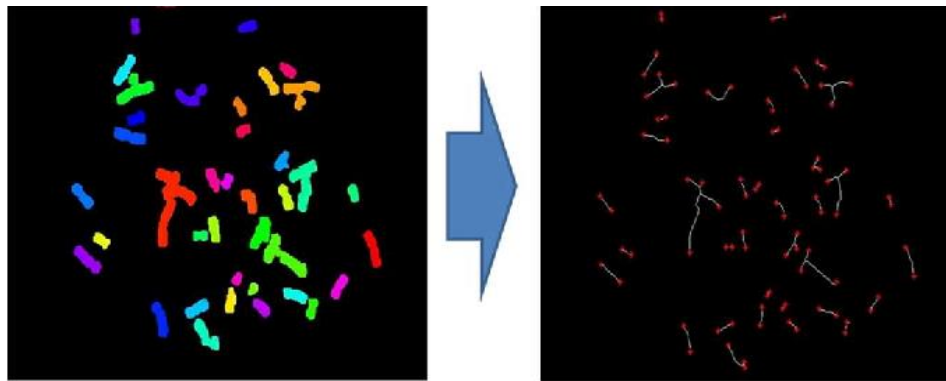


Figure 1.4: Segmentation results of an M-FISH image by Schwartzkopf's method

As can be seen in Figure 1.5(b), Schwartzkopf's method was unsuccessful because two overlapping chromosomes were members of the same class. The segregated chromosomes result in a greater pixel classification accuracy since the approach corrects misclassifications when mixing colour blobs. However, the merging procedure is greedy rather than optimal: given a set of blobs, the method combines the pair with the highest likelihood in relation to all other pairings, which may not produce the correct segmentation.

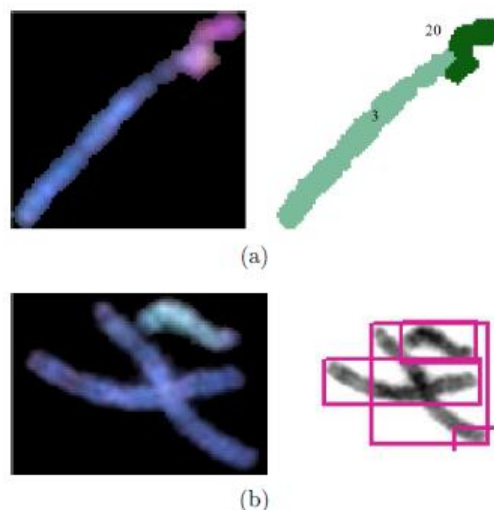


Figure 1.5: (a) Schwartzkopf's method successfully decomposed touching chromosomes, whereas grayscale based method (using Cytovision software) could not since two chromosomes appear as a long chromosome. (b) Grayscale based method could decompose, whereas Schwartzkopf's method could not since two overlapping chromosomes belong to the same class

1.3 Methods

The drawbacks of both color-based and geometry-based methods can be overcome by merging data from both. The new method uses cluster geometry, pixel categorisation results, and chromosomal sizes. This section discusses the details of the implementation. After the chromosomes have been isolated from the background, just the chromosomal pixels are normalised using EM normalisation. Then, classification is done using an unsupervised nonparametric method called the minimum-distance classifier.

1.3.1 Components of the cluster

A group of pixels joined by 4-connectivity is called a cluster, or Si. Clusters are found when the chromosomes are separated from the background using the segmentation method that was provided and the segmentation result is shredded using a 3 x 3 structuring element. Erosive processes are used to avoid evaluating simple touching scenarios when chromosomes are connected by a single pixel. Each cluster is dilated back before being evaluated for touching and overlap. A cluster can be formed by one or more chromosomes. Regardless of whether a cluster consists of one or many chromosomes, it is all assessed.

1. Cross shape cluster
2. T shape cluster
3. I shape cluster

1.3.2 Assessment of the hypothesis

One or more chromosomes make up each of the several cluster-based ideas. When a cluster is split into the appropriate number of chromosomes of the appropriate sizes while also optimising the homogeneity of the pixel memberships within each chromosome, the ideal cut is obtained.

1.3.3 Steps in Decomposition

The landmarks are located on the boundary as indicated, given a set of pixels and their class memberships. The cluster is classified as cross-, T-, I-, or multiple-shaped based on the landmarks. The evaluation of the corresponding subcases determines whether the cluster is classified as a T or Cross form. Given the form limitation, the case with the highest probability among the subcases is selected as the optimal separation for the cluster. The best subcase's individual chromosomes are next assessed for touchings (Ishape evaluation). A group of two chromosomes from the same class crossing each other serves as an example of the breakdown process. The method demonstrated successfully breaks down the chromosomal overlaps of the same class. It displays the T-shape cluster's decomposition steps.

1.5 Experimental Setup

Existing 1	Existing 2	Proposed
25	45	67
43	66	123
39	46	154
65	87	165
68	98	200

Table 1.5: ENTROPHY

Table 1.5 represents ENTROPHY values. These ENTROPHY is compare with existing values and proposed also. But their proposed values of ENTROPHY are higher than compare with existing 1 and 2.

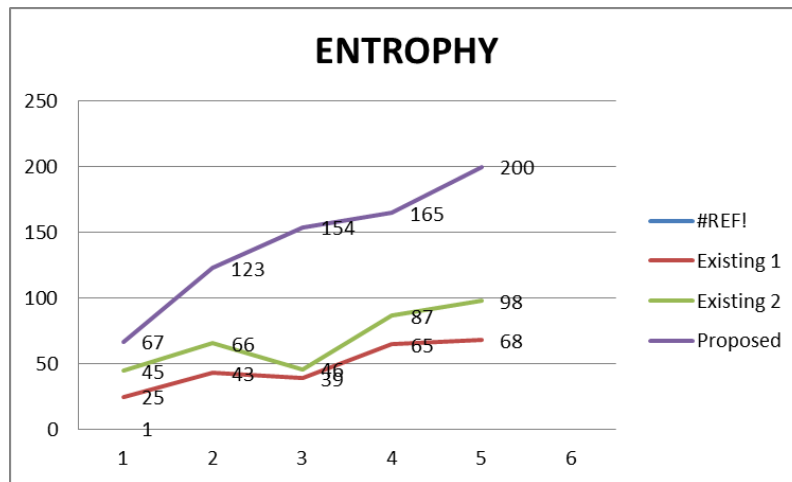


Figure 1.6: ENTROPHY

Figure 5.6 represents ENTROPHY values of existing and proposed. Two variations of existing values are compare with proposed value. Their proposed values are higher than compare with existing values.

Existing 1	Existing 2	Proposed
25	32	45
35	39	65
47	49	76
88	98	154
122	145	200

Table 1.2: RANGE

Table 1.2 represents RANGE values. These RANGE is compare with existing values and proposed also. But their proposed values of RANGE are higher than compare with existing 1 and 2.

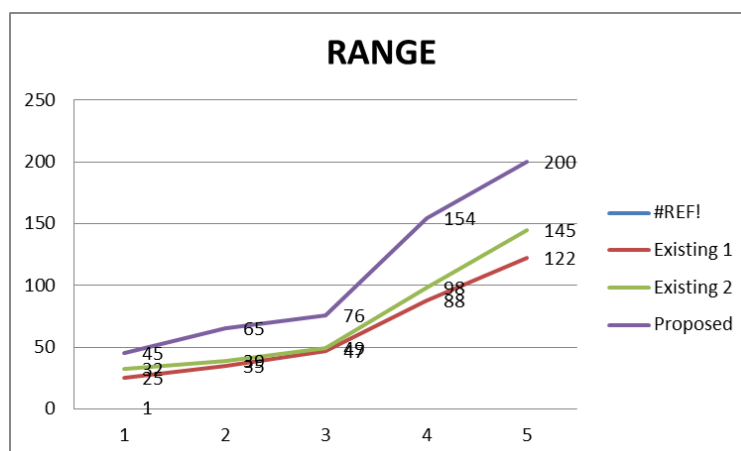


Figure 1.7: RANGE

Figure 1.7 displays the current and suggested range values. The suggested value is contrasted with two versions of the current values. The current values are lower than their suggested values.

Existing 1	Existing 2	Proposed
23	36	45
34	45	56
47	59	66
54	67	88
58	176	198

Table 5.3: STANDARD DEVIATION

The STANDARD DEVIATION values are shown in Table 1.3. For these standard deviations, comparisons to the present values are provided as well as recommendations. However, their suggested standard deviation values are more than the current 1 and 2.

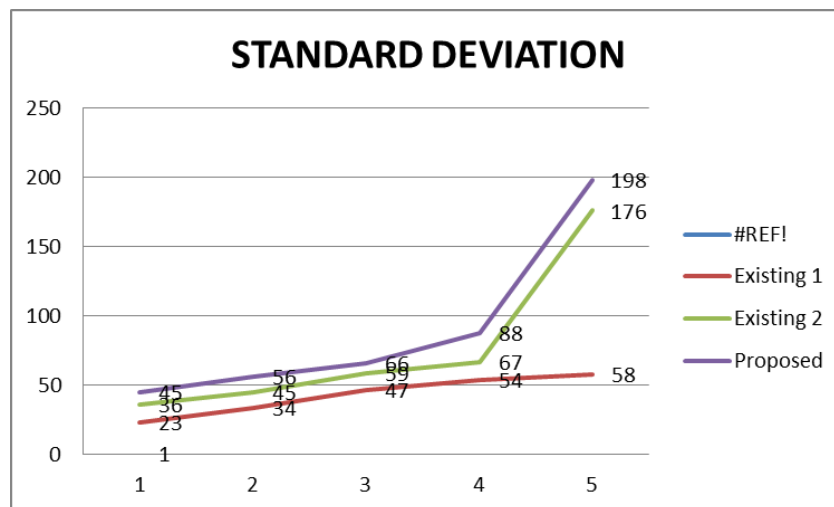


Figure 1.8: STANDARD DEVIATION

Figure 1.8 displays the current and suggested STANDARD DEVIATION values. Two variations of the present values are contrasted with the recommended value. Their recommended values are higher than the present values.

2. CONCLUSION

For M-FISH images, two new unsupervised nonparametric classification methods were presented: a fuzzylogic classifier and a template matching algorithm. Their accuracy was comparable to that of a maximum likelihood classifier, and both methods provide a significant computational time advantage over supervised methods. Overlapping and contacting chromosomes were successfully broken down by the developed decomposition approach. Numerous hypotheses were generated based on cluster organisation, chromosomal sizes, and pixel categorisation data. The best decomposition was determined to be the hypothesis that maximised the probability function. After chromosomes were identified independently, misclassified pixels were effectively corrected while retaining the translocated pixels using the previously adjusted reclassification method.

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