Lane Detection on Gel Electrophoresis Images using Active Shape Models

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Abstract—Biocontrols are one of the factors to be considered in the search for sustainable agricultural activities. The amount of samples to be analyzed while searching for such agents draws mandatory the use of technological tools that accelerate the processes involved and improve the accuracy of the experimental results. The molecular characterization of organisms is one of the steps involved while searching for such biocontrols. Several techniques used for the molecular characterization of organisms make use of gel electrophoresis images, which permit to work at coarser but faster level of information analysis than genetic sequencing.

During the creation of gel electrophoresis images, two types of distortions occur. The first type originates on the gel creation process due to variations of the electric field and temperature gradients. The second type is induced in the image capture process and involves optical distortions, such as the barrel distortion.

This project is focused on lane detection over images of gel electrophoresis processes, which are used for molecular characterization of organisms, even if distortions are present during the acquisition process.

Index Terms—electrophoresis gel, image analysis, lane detection, distortion

I. INTRODUCTION

The Brundtland Report [1] defines human development as sustainable if a balance among social, economical and environmental factors is attained in a way that the needs of both, present and future generations are ensured. Novel technological solutions are necessary to deal with the problems encountered while pursuing that balance. To strive for cleaner and more effective production methods, and to find ways to measure and compensate the effects of such methods on the environment and on the human population are the challenges of modern engineering disciplines. In this regard, molecular biology offers tools to precisely characterize organisms, what in turn supports the optimization of quality, quantity and impact of the products of agricultural, forestry, and farming activities, and also makes it possible via biomonitors to keep track of the environmental health.

Even though the biomolecular tools and methods provide means to find novel solutions to present problems, the complexity of the required methodologies and equipment and the associated costs limit their spread use and slow down possible advances toward a sustainable development. Hence, additional tools have to be conceived to support and reduce the time required by experts on the analysis of experimental results.

The present paper collaborates with the development of a tool to support the analysis of gel electrophoresis images, which are used to analyze the presence of particular molecular chains in proteins or nucleic acids.

The analysis of gel electrophoresis images is found as a previous phase of DNA sequencing, as it permits to select particular DNA segments that are of interest to particular applications. Additionally, these images are produced as a validation tool, for instance, for DNA extraction. They can also be used to compare DNA samples at a “coarser” level than sequencing, since their precision is high enough to distinguish samples in a wide range of applications.

Gel electrophoresis is based on the dependency of the mobility of molecules in terms of their size and electric charge while moving through the gel. As a first step, the substances to be characterized, such as DNA or RNA, are combined with a mixture of restriction enzymes in order to perform a segmentation of the molecules. Subsequently, the sample is injected into an agarose or polyacrylamide gel matrix that is under the influence of an electric field. The images obtained once the electrophoresis process has end encode the information on the molecular composition of the sample through the final locations of “bands” or molecule fragments within “lanes” formed by the propagation of the samples through the gel. The band locations are used to perform comparisons between two or more lanes.

Figure 1 shows a block diagram of the electrophoresis gel image analysis tool being developed. The present work concentrates on the lane detection block.

![Figure 1. Block diagram of analysis tool for electrophoresis gel images.](image)

During the creation of gel electrophoresis images, two types of distortions occur. The first type is intrinsic to the
electrophoresis process, where variations in the electric field and temperature induce irregularities in the lane paths and band positions. The second type of distortions are related to the capture process, and involve for instance optical distortions, such as the barrel distortion. 

This work proposes a novel method to detect the lanes in a gel electrophoresis image, even if distortions are present during the acquisition process. The method is based on Active Shape Models (ASMs) [2] and an additional strategy to consistently couple the models to the borders of the lanes.

This paper is structured as follows: next section presents a brief review of related works. Section III summarizes the proposed method, followed by a description and evaluation of the results achieved in section IV. Section V concludes this work.

II. RELATED WORKS

Molecular biologists use different sizes of gels, depending on the needs of each application. Small and medium sized gels up to A3 format (297 mm × 420 mm) are captured using flat scanners when no UV lighting is necessary. Larger gels require more sophisticated scene configurations. When using small gels or flat scanners the optical distortions are negligible and simple lane detection methods as the ones proposed in [3], [4], [5] may be used.

Other systems proposed (e.g. [6]) required the lane detection to be manually performed by a human user.

No other work known to the authors describes methods to perform the lane detection in the presence of optical distortions, and therefore, the available methods are not applicable to the gel electrophoresis images available to this work, which are larger to the A3 format and must be captured with a lens/camera setup that introduces barrel distortion.

III. LANE DETECTION USING ASM

The shape of an object can be represented by $n$ points placed on characteristic features of an object’s border and on particular locations of interest. These point-based shapes can additionally be interpreted as points in a $2n$-dimensional linear space [2]. A group of shapes on this space can be approximated by projections on a subspace spanned by the axes holding most of the shape variations on the $2n$-dimensional space. These axes correspond to the eigenvectors of the covariance matrix of all points (i.e. shapes in the $2n$-dimensional space), and the variance on each axis is given by the corresponding eigenvalues. The projective model is then called a Point Distribution Model (PDM).

An Active Shape Model is a template capable of optimally adjust itself to an image shape through a similarity transformation (rotation, scaling, translation) and by restraining its shape variation modes to the ones described through a PDM.

For the particular lane detection task an ASM is created using for its training theoretical shape models for barrel distortion described by:

$$r_d(r_u) = r_u + ar_u^3 + br_u^5 + \ldots$$

where $a$ and $b$ are scale factors, $r_d$ is the distorted radius and $r_u$ is the undistorted radius.

There are four assumptions about the gel electrophoresis images on which this work relies to detect the lanes:

1. Lane orientation is fixed and known a-priori in the images (horizontal or vertical). 
2. Lanes in a gel electrophoresis image have the same widths and are separated by the same distances (which can be zero). 
3. The maxima of the magnitude of the image gradient correspond to the lane borders. 
4. On the image gradient, there should be nothing between the lanes.

On a further step, an initial guess of the lane width is needed. This is estimated using an algorithm based on the autocorrelation of the signals obtained perpendicular to the lane orientation axis (procedure based on fact 2) such that when distortions are present, an approximation of this width can be obtained by finding the period of the autocorrelation, which is the same as the one of the signal. This period is calculated on several sites of the image and a width histogram is created as a probabilistic non-parametric width distribution model.

With the image gradient on lines perpendicular to the lane orientation axis and the previous width distribution model estimation, several shapes are placed along the image using a selective maximum filter. This filter looks for potential lane borders by finding the largest connected line on the image gradient, which is the strongest lane. After that, it looks for maxima every lane width, and gives them weights using the histogram from the previous step, this discards false positives for lane borders.

Finally, an iterative process is started to align the models with each other and with the lane borders. In this step, the ASM is used to keep the shapes in the Allowable Shape Domain i.e. to remain as closest as possible to the most common shapes found in the training process. Based on those shape models it is possible to compute the inverse mapping that compensates the distortion.

The iterative algorithm is analyzed in terms of the lane deviation from the human detection criterion and an “ideal lane” (a straight line). This analysis is based on the comparison of deviations at two different stages of the process. The first
measurement instant is the initial detection (before iterating) where the mean lane deviation against a reference given by a human is measured. The second measurement occurs after the iterative and rectification processes. Figure 2 shows a block diagram of the solution exposed above.

IV. RESULTS

The results presented have been obtained for gel electrophoresis images where a small separation gap exist between lanes. The method is applicable though to images without a gap. Figure 3 shows an example of a detection using the method proposed where blue lines represent the lane borders.

![Figure 3. Result of lane detection using the method proposed](image)

As mentioned before, two measurement stages were used to assess the lane detection. In the first one, the lanes were both, manually identified and automatically detected with the proposed algorithm on the image used for figure 3. The deviation between both methods are then measured. Knowing that \( M \) is the total number of shapes describing the lane borders, \( N \) the number of points for each shape, \( p_{iy} \) the \( y \) coordinate of the manually detected lane and \( j_{iy} \) the one detected by the automatic algorithm, the deviation between both criteria is computed as

\[
D = \frac{1}{MN} \sum_{i=0}^{MN-1} |p_{iy} - j_{iy}|
\]  

(2)

Table I shows the deviation results as a function of the subspace dimensionality used. The first column show two values. The first one corresponds to the percentage of the total variation captured by the subspace, where the total variation corresponds to the sum of all eigenvalues of the covariance matrix used while training the ASM. The second value indicates how many dimensions the subspace required to reach the indicated percentage of the total variation. The maximal dimensionality for the subspace of two-dimensional shapes described by \( n \) points is \( 2n \) (as each point requires two values \((x, y)\)). For the experimental setup used, \( n \) is 110 which implies a maximal subspace dimensionality of 220. The results show a deviation barely affected by the number of dimensions used. However, further results will show that this does not mean that similar distortion corrections are obtained for the given conditions.

For the second measuring stage, three lanes (one near the center and the other two on the upper and lower part of image 3), are manually labeled. For a set of points \((x_i, y_i)\) that describe a shape the standard deviation for one of the coordinates is computed. Since for the images used in the experiments the lane orientation is horizontal, the standard deviation of the \( y \) coordinate is estimated before and after the iterations. Table II shows the results before the iterative process. In order to show the effects of the iterative process and the number of iterations required to convergence, the algorithm is evaluated taking the number of iterations as a parameter. This is summarized on table III and illustrated on figure 4.

<table>
<thead>
<tr>
<th>Lane</th>
<th>Std. Dev. prev. rectification (pixels)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>8.279</td>
</tr>
<tr>
<td>2</td>
<td>1.8139</td>
</tr>
<tr>
<td>3</td>
<td>8.4060</td>
</tr>
</tbody>
</table>

Table II: LANE STANDARD DEVIATION \( \sigma \) BEFORE THE RECTIFICATION PROCESS

<table>
<thead>
<tr>
<th>Number of iterations</th>
<th>Std. Dev. post-correction (pixels)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>Lane 1  Lane 2  Lane 3</td>
</tr>
<tr>
<td>1</td>
<td>1.1504  0.9210  2.1183</td>
</tr>
<tr>
<td>2</td>
<td>1.5556  0.8967  1.3792</td>
</tr>
<tr>
<td>3</td>
<td>1.5671  0.7896  0.9821</td>
</tr>
<tr>
<td>4</td>
<td>1.5763  0.7775  0.9776</td>
</tr>
<tr>
<td>5</td>
<td>1.5872  0.7885  0.9760</td>
</tr>
</tbody>
</table>

Table III: STANDARD DEVIATION \( \sigma \) POST-CORRECTION AS A FUNCTION OF THE NUMBER OF ITERATIONS

Even though Lane 1 presents an increase on the standard deviation \( \sigma \), the predominant tendency for \( \sigma \) is to decrease. For the given image, convergence was reached after the second iteration. The magnitude of the deviation indicates that the deviation of the detected lanes is mostly on the one-pixel range, which exceeds the accuracy requirements for the lane segmentation stages.

The effect of modifying the subspace dimensionality is explored in table IV. The results confirm that one dimension is not enough to represent a lane and that too many degrees
Table IV

| Percentages of Freedom | Std. Dev. Post-Correction
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Ln 1</td>
<td>Ln 2</td>
</tr>
<tr>
<td>50/1</td>
<td>9.2502</td>
</tr>
<tr>
<td>80/2</td>
<td>0.8046</td>
</tr>
<tr>
<td>96/3</td>
<td>0.8023</td>
</tr>
<tr>
<td>100/220</td>
<td>4.2715</td>
</tr>
</tbody>
</table>

of freedom on the shape representation allow the shapes to be deformed in ways not valid according to the training set. An inverse mapping is computed that transforms the detected lane boundaries into equidistant straight lines. The application of that mapping warps the image into the final distortion-free result. Figure 5 shows an example of a rectified image.

V. Conclusions

A method to model distortions in gel electrophoresis images using Active Shape Models (ASM) is proposed. These shape models describe the distortion in a way that inverse mappings can be computed to warp the image into a distortion-free version.

Since ASM models are based on the projection of shapes into a low-dimensional subspace, it was evaluated which effect the subspace dimensionality has on the final lane deviation. Three dimensions already capture 96% of the shape variations in the training set. It was observed, that two dimensions (80% variation coverage) are sufficient for the models to represent the distortions present on the example images. However, the number of dimensions used must be limited, since otherwise unnatural shapes not present in the training set would be produced.

The number of points used in a shape model is proportional to the capacity of that model to exactly adapt to image data. However, the higher that number, the larger the computational load to directly process those models. The ASM proved their capability to reduce the amount of dimensions needed for processing. In this case just 0.91% of the total available dimensions were needed.

An additional advantage of the subspace projected shapes (not detailed on this work due to the space limitations), is the processing capabilities in the lower-dimensional representation. A smooth transition between the shapes of adjacent lanes is ensured by means of low-pass filtering on a the lane shape sequence, which implies an elastic model coupling.

In future works a robust parameter estimation for the optical distortion based on the ASM models will be presented.

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References