

Computer Based Grid Detection on Makler Images

Hamza O. Ilhan and Fethullah Karabiber

Computer Engineering Department, Yildiz Technical University, Istanbul, Turkey

Email: hoilhan@yildiz.edu.tr, fkara@ce.yildiz.edu.tr

Abstract—Makler counting chamber is commonly used microscope kit for the evaluation of semen cells in terms of number and motility analyzes. Mainly, a physically located micron-size grid structure provides the generalization of observations over entire samples owing to the globally accepted criteria by academicians and experts. Evaluation process is mostly realized manually, which can cause mistakes. At first glance, mistakes can be ignored, but when it is generalized by the Makler criteria, results can mislead the diagnoses. In this sense, an autonomous analysis of Makler images is necessity instead of manual observation. This study specifically focuses on grid detection of Makler images, which is planned as the first phase of mentioned system. In this manner, a new approach, combination Hough Transform and K-Means clustering, is presented and tested over synthetically formed Makler images with different angles. Furthermore, pattern-matching idea is performed on the angle detection and evaluation steps.

Index Terms—Makler images, microscopic images, Hough transform, K-means clustering, pattern matching, grid detection, semen analyses.

I. INTRODUCTION

Today, regular diagnoses about male infertility mostly carry out with manual observation in the laboratories. Although there is advantages in manual observation, disadvantages are more and important such as being a time consuming process and requiring more experienced observer [1]. Additionally, diagnoses can differ according to experts in laboratories. With the advances in computer science, several automatic diagnosis tools based on computers are developed and located in medical market such as CASA systems [2].

CASA systems mainly work on the images obtained by embedded camera. Information such as the number and several basic movement analyses of semen derived within video and image processing concepts in computer science terminology. However, results are still not an acceptable rate in terms of reliability and accuracy according to doctors. Hence, many laboratories are still applying manual observation methods. In this sense, academicians have researched several studies [3], [4] in order to facilitate the process and standardize the result in common metric. Makler is one of them invented by Amnon Makler and firstly published in 1980 [5].

Makler is a sperm counting device using in embryology laboratories or IVF units in hospitals. It is a unique innovation in medical science, because it provides generalization of hypothesis over entire samples. Otherwise, measuring all samples individually and manually is time consuming process and hard to generalize the diagnoses. In this manner, Makler counting chamber is a standardization way to express the results within a short period. Not only about counting, but also motility analysis can be assessed as well.

Makler chamber is built on physical 10 squares of grid with light-absorbing material. Grid remains black color under microscope light because of the material and is used as reference points. In image processing concept, the material is also provides easy to eliminate the grid on the images. Furthermore, Makler has 10 microns depth. In regular microscopic experiments, depth between lamellae and cover glass is an important factor, because in some cases, it causes blurring. However, Makler restrains the amount of sample, thus results with more clear images. It also provides more fluid semen movements.

In medical manner, Makler gives more pure results because samples are not diluted. Evaluation made upon any strips of 10 squares across all Makler chamber. The number of spermatozoa within selected strip indicates their concentration in millions/mL. In this sense, decision step of counting the semen are made on grid structure.

This study includes an automatic grid detection application for Makler images. Well-known line detection algorithm; Hough transform, is utilized with several pre-processing techniques, and K-Means clustering method to detect only grids as post-processing. Additionally, Makler can be placed with different angle under microscopes. Identification of grid also provides the angle information of the Makler. Designed software automatically coordinates the images to 0 degree according to detected grid angle by presented algorithm. Counting of semen by computer can be maintained with detected grid information in order to generalize result as in manual observation. Moreover, further studies such as motility analysis of the semen can be investigated based on detected lines in computer side. It will be more reliable because the process similar to manual observation.

This paper consists of five sections. Several computers based medical image analysis studies similar to our approach are mentioned in Section 2. Image acquisition with specifications, Signal preprocessing steps and usage

of Hough transform with classical K-Means clustering algorithm are described in Section 3. Results and discussions are placed in Section 4 and the paper end up with conclusion and future works in Section 5.

II. RELATED WORKS

Grid structure contains mainly two straight lines having a fixed 90° angle difference between. In theory, detection of lines gives the detection of grid. Therefore, several line detection algorithms are explored.

During the early period of studies of line detections, edge points in the images are mainly used reference points in algorithms. Djemel proposed a new algorithm using canny points with infinite impulse response filter (IIR) [6]. It is highlighted that proposed algorithm has strengths on computational time and memory storage. Afterwards, another line detection algorithm based on edge detection is made by Guru *et al.* They combined several methods to form more robust line detection algorithm [7]. Firstly they detected edge points of the objects then applied eigenvalue analysis to connect the edge points. But as it is also mentioned in the study, algorithm is practically useful for only straight line detections.

Marlet and Zerubia proposed a new mathematical formalization of the F^* algorithm within the concept of dynamic programming [8]. They used line detection for valley and roads on satellite images. Another line detection algorithm; recursive line fitting method, is used in [9] for robot applications. Paper emphasizes that algorithm is slow because it is iteratively process and not useful for robot applications. Radon transform is studied in [10]. Author mentioned that it is robust in noisy images, but fails on detection of centerline because of the peak selection problem. They introduce a mean filter to overcome that problem. But still the necessity of improvements is reported. Principle Component Analysis (PCA) is utilized for straight line detection in [11]. They extracted and classified edges as row and column then they detect the straight one between the edge points according to eigenvalues. Straight lines should have higher eigenvalue. They also presented a comparative table in the paper with Hough Transform.

In this study, we modified Hough Transform with K-Means clustering method to detect grid structures which is similar to straight line detection studies. In this sense differently Hough Transform algorithms are reviewed such as regularized HT [12], probabilistic HT [13], and progressive probabilistic HT [14]. After detection of the lines, elimination for non-grid lines will be provided by clustering because HT is one of the most sensitive algorithms to noise and non-momentary lines. A basic clustering algorithm, K-Means, is utilized. K-Means clustering is generally applied on trajectory clustering. Similar to our study, Lee *et al.* proposed a framework for grouping common parts of multi-trajectories according to their angles [15].

Evaluation of this study is performed by pattern matching idea. Many different studies on pattern

matching such as text classification [16], biomedical [17], [18] and biometric applications [19] are located in literature. This study also includes a basic pixel to pixel matching idea between a generated pattern and grid detected images.

III. METHODOLOGY

Our study especially focuses on automatic rotation software based on grid detection. After preprocessing step, a well-known algorithm; Hough Transform, is used for line detection. Then, several mathematically calculations are performed to identify angle of lines. K-Means clustering is utilized over angles to line elimination process for outlier values. Finally, the image is rotated by the weighted angle value. In order to present numerical success results of the utilized method, a grid pattern is formed, and used for matching. See Fig. 1 for flow diagram of the presented study.

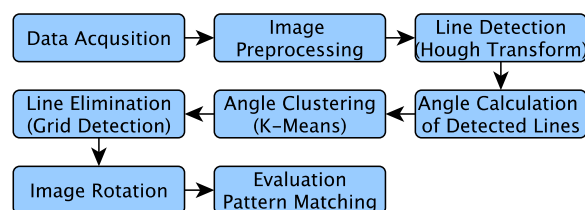


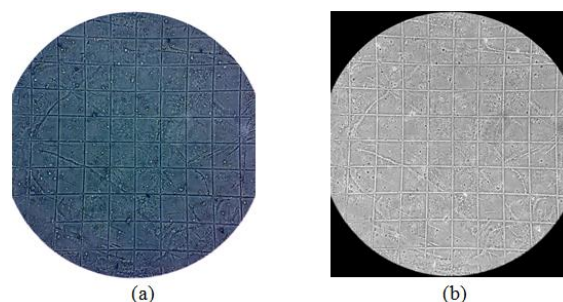
Figure 1. Flow diagram of study.

A. Data Acquisition

Semen images are obtained by mobile phone with 1920×1080 resolutions from ocular part of microscopy in Medical Department of Turgut Ozal University. Images include Makler grid at behind, noises and debris with all black colors under the light effects of microscopy. On the other hand, semen cells are glared due to reflection of mitochondria. See Fig. 2(a) for an image as sample.

B. Image Pre-Processing

Pre-processing is unavoidable and principle set of processes in information retrieval applications. Debris and misleading short lines should be eliminated. Otherwise, debris affects the analyses of semen and lines other than grid structure give false calculation of angle. Mainly; inverse process, several morphological operations, average median filter with a fixed window size (128×128), high pass filter for sharpening the lines and image adjustment based on histogram are applied. See Fig. 2 in the following page for the entire steps in pre-processing over a sample frame.



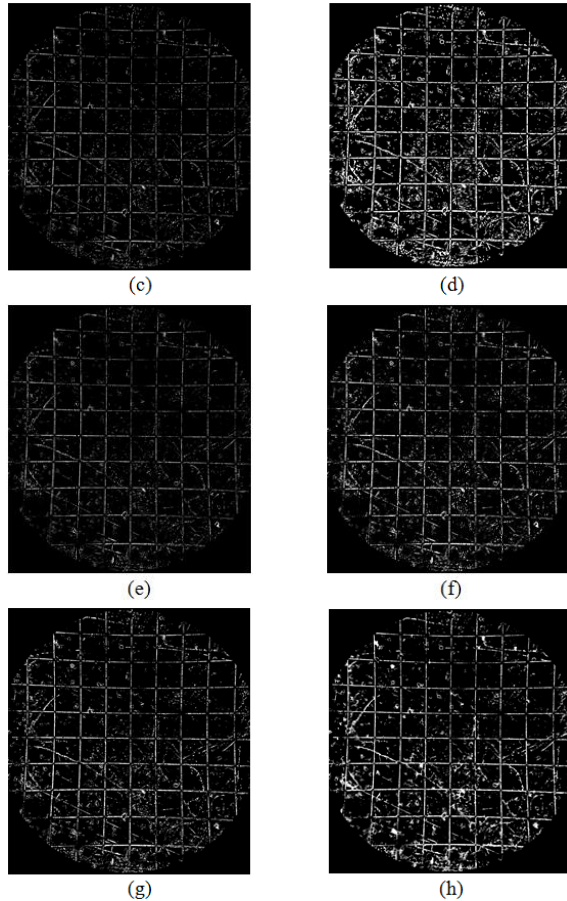


Figure 2. Image preprocessing steps: a) raw image, b) inverse form, c) morphological operations, d) average median filter (128x128), e) morphological operations, f) high pass filter, g) histogram adjustment, h) sharpening.

C. Hough Transform

Hough transform is a special case of the Radon Transform [20]. It extracts several key features that are mainly used in line detection and performs a voting procedure for a certain class of shapes.

The heavy burden of computational complexity and massive storage requirement is the drawback of the standard Hough Transform (HT). Besides, the Hough transform is also quite a “blind” algorithm, which is much inferior to the intellectual mechanism of human’s visual recognition.

In order to overcome weaknesses of Hough transform, many modified approaches have been presented such as regularized and progressive HT [12], [13]. However, it is still slow, not fully adaptive and easily effected by noises. We tried to eliminate noise effect by K-Means combination over probabilistic HT algorithm. On the other hand, K-Means integration makes more computational complexity, which results with more calculation time.

D. Angle Calculation and Image Rotation

Hough Transform gives starting and ending positions of detected lines in 2D (x, y) coordinate system. According to basic geometry theorem, slopes of the lines are calculated by using (1). Rotation of the images is

provided with angle detection. Derived slope information is used in (2) to find the angle. Final image is obtained by rotation of original image with calculated angle.

$$y = mx + n \tag{1}$$

$$\angle = \tan^{-1} m \times \frac{180}{\pi} \tag{2}$$

E. K-Means Clustering

Clustering algorithms split data into predefined class numbers based on distances between each point. In that meaning, clustering idea used after Hough transform to eliminate false detected lines. Not only lines in grid structure, but also other lines caused by external factors with different angles are detected by Hough transform because of the illumination changes. In that case, K-Means clustering is performed with two-class structure to eliminate false detected lines.

Slope information of detected lines is derived and used as feature set that will be classified by K-Means. Majority slopes belong to grid structure, hence two classes are defined because of the horizontal and vertical alignment of the lines in grid, and other lines with different angles are eliminated. As a result, lines having the most iterative slopes remain in the frame which indicates the grid. See Fig. 3 for the initial founded slopes and iteratively classified result for 12 °rotated points.

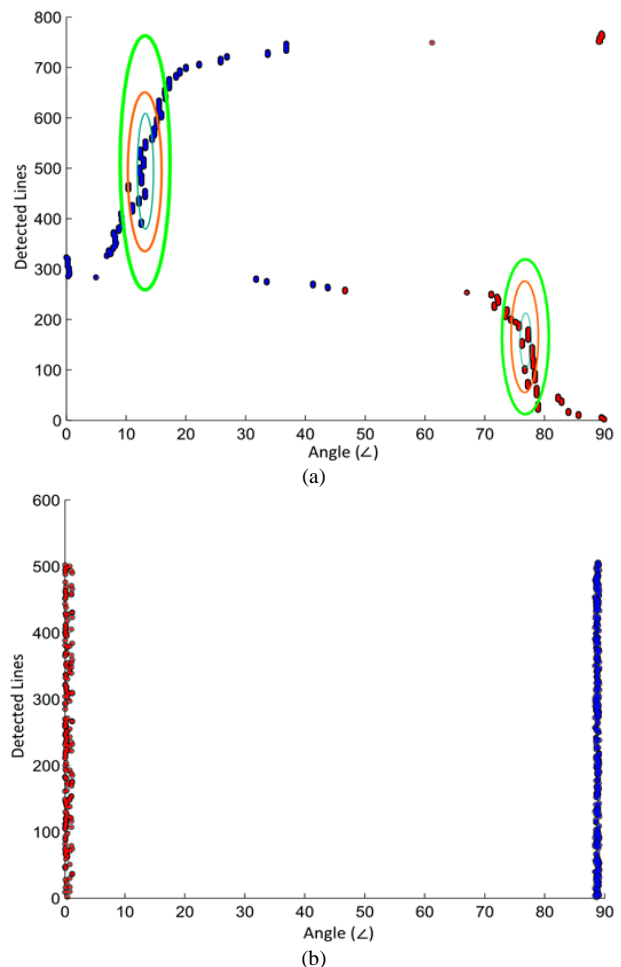


Figure 3. a) K-Means clustering of each detected lines angle, b) angles of the detected lines for rotated and classified image.

F. Pattern Matching

Success of the Hough + K-Means is evaluated by pattern matching. In this sense, a grid pattern is synthetically generated. After rotation of the original raw image with the angle information, occlusion points between detected lines as grid and pattern are measured as accuracy rate. See Fig. 4 for the generated pattern for reference points to use in matching.

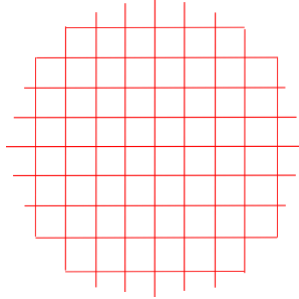


Figure 4. Generated pattern for matching with detected lines as grid.

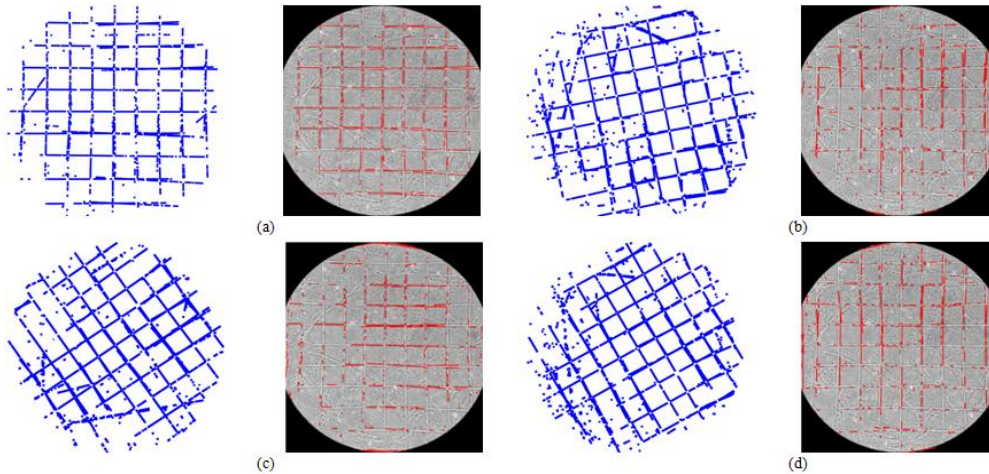


Figure 5. Grid detection at various angles; a) 0 °, b) 12 °, c) 125 °, d) 30 °. (Hough T. results (left), K-Means clustering and rotated results (right)).

Confusion matrix is formed as in Table I. TP represents the correctly matched pixels with pattern and final image with detected grid lines. FP indicates the wrongly detected lines in outcome when compare to pattern image pixels. At the state where grid lines are present in pattern, but our algorithm erase by mistake and assigned that pixels as not-line signified by FN number. TN is the number for matching of non-grid line pixels.

TABLE I. CONFUSION MATRIX FOR EVALUATION OF STUDY

# of Matched Pixels	Pixels in Pattern	
	Pixels in outcome image (Hough + K-Means)	True Positive (TP)
	False Negative (FN)	True Negative (TN)

Accuracy rates only focus on truly matching between detected grid lines and pattern grid lines, but FP and FN is also important. According to abovementioned definition of confusion matrix, if detected grid lines are different than actual generated pattern lines, it causes FP. Contrary, if combination of Hough and K-Means gives

IV. TEST RESULTS

Hough + K-Means combination is tested on synthetically generated 500 differently rotated images. Fig. 5 demonstrates several images' initial line detection of Hough after preprocessing (left) and detection of grid structure after K-Means clustering of angles (right).

Pattern matching idea is used between founded grid lines images and generated pattern. Accuracy rates and F-Measure scores according to confusion matrix are calculated as performance metrics. Equations of the metrics are presented in (3) and (4) as Accuracy and F-measure, respectively.

$$Accuracy = \frac{TP+TN}{TP+FP+FN+TN} \tag{3}$$

$$Precision = \frac{TP}{TP+FP} \tag{4}$$

$$Recall = \frac{TP}{TP+FN} \tag{4}$$

$$F - Measure = 2x \frac{Precision \times Recall}{Precision+Recall}$$

the pixels as not grid line, but they are stated as grid line in pattern, then it gives FN. Those errors mentioned in literature as Type I and II error or a general terminology as Accuracy Paradox [21]. Overcome this problem carry out with F-Measure scores which involves FP and FN into the equation. F-Measure scores generally ends up with low scores compare to Accuracy rates, because it includes accuracy and some other criteria together.

Accuracy rates and F-Measure scores are noted in Table II with 30 ° intervals. Each interval has an average score belongs to 30 synthetically generated images with corresponding rotated angles. F-Measure scores are less successful than accuracy score according to presence of FP and FN values. The main reason is about the detection of Hough Transform. HT found several lines, not only straight one, in frame. We tried to eliminate them by preprocessing and K-Means clustering, but still some other lines with same horizontal or vertical angles similar to grid lines couldn't be erased. That caused FP issues. On the other hand, FN is occurred because of using preprocessing and K-Means clustering. Line elimination process also affected the main grid lines. Hence, erased parts of lines resulted as FN owing to pattern matching.

TABLE II. PERFORMANCE METRIC RESULTS OF PROPOSED APPROACH

%	Angle Intervals					
	0-30	30-60	60-90	90-120	120-150	150-180
Accuracy	94	87	91	88	91	93
F-Measure	76	79	75	78	83	77

V. CONCLUSION

This paper presents a modified line detection algorithm to use in grid detection of Makler images. Makler images are mainly used for Spermogram tests which deal with number of semen and motility analyses. It provides simplicity and standardization for deciding step. Otherwise, it is hard to evaluate within all samples. In terms of organizing an automated system in this issue will provide more accurate diagnoses. Grid detection is the first phase in desired system. However, detecting of lines is not easy challenging owing to noises, undesirable particles, microscopic characteristics etc. In this study, we aim to overcome problems with a new approach as combination of Hough Transform and K-Means clustering. Results are promising, but still needs to improved. Extra pre-processing and different clustering algorithms will be utilized in further studies in order to decrease FN and FP errors, and increase F-Measure scores. Additionally, this study will be continued with testing for computer based analyzing methods on images in further studies. Detected grids will be used as reference points for computerized methods.

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Hamza O. İlhan received the B.Sc. in electronics and computer science from Marmara University, Istanbul, Turkey in 2010. M.Sc. degree is received in computer engineering from Yalova University in 2012. He is currently Ph.D. student in Yıldız Technical University (YTU), Istanbul, Turkey. He is appointed to Yıldız Technical University as research assistant in 2011. His research interests are in the areas of autonomous robots, image and signal processing, machine learning and pattern recognition with applications to biomedical engineering.



Fethullah Karabiber received the B.Sc. in Electronic Engineering from Istanbul Teknik University (ITU). M.Sc. and Ph.D. degrees are received in Computer Engineering from the Istanbul University, Turkey, in 2005 and 2009 respectively. Dr. Karabiber is currently an Associate Professor at the Computer Engineering Department, Yıldız Technical University. His interests include machine learning, bioinformatics, signal and image processing. He has published several scientific papers.