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RESEARCH ARTICLE

Automated recognition of urinary microscopic solid particles

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Abstract

Urine analysis reveals the presence of many problems and diseases in the human body. Manual microscopic urine analysis is time-consuming, subjective to human observation and causes mistakes. Computer aided automatic microscopic analysis can help to overcome these problems. This paper introduces a comprehensive approach for automating procedures for detecting and recognition of microscopic urine particles. Samples of red blood cells (RBC), white blood cells (WBC), calcium oxalate, triple phosphate and other undefined images were used in experiments. Image processing functions and segmentation were applied, shape and textural features were extracted and five classifiers were tested to get the best results. Repeated experiments were done for adjusting factors to produce the best evaluation results. A good performance was achieved compared with many related works.

Keywords

Automatic recognition, classification, computer aided medical analysis, data mining, feature extraction/selection, image pre-processing, image segmentation, microscopic urine analysis

History

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1. Introduction

Microscopic urinalysis detects the presence of elements that often provide early diagnostic information concerning dysfunction, infection or inflammation of the kidneys and urinary tract [1–3]. One of the main parts of that test is handled by human eye observations through a microscope. As human eye observation is subjective, time-consuming and causes mistakes, researchers studied methods of automating microscopic analysis with the aid of computer and software systems. Computer-aided medical analysis is using computer hardware and software tools to apply medical analysis instead of human manual procedures [4]. Automated procedures are those procedures done by computer hardware and software tools to replace the manual actions. Automatic detection of urine particles occurs through specifying those regions that contain target objects and extract them.

To make computer-aided urine analysis systems effective and highly required in medical labs, they should be trusted, and this can be achieved by increasing the rate of successful recognition of specimen objects. In addition, to address variation between using different classifiers, a comparison is needed to locate best suitable classifiers for this field. Also, noisy and undefined particles that appear in lots of urine specimens need a special treatment. The methodology of research is as follows:

- (1) Collecting a big set of images of urine samples captured using a microscope.
- (2) Image enhancement was applied to improve the perception of information inside images.
- (3) Segmentation was applied to simplify the representation of an image into something that is more meaningful and easier to analyse and to locate objects for better object extraction.
- (4) Evaluation on different segmentation adjustments to show the best approach that maximizes well extraction of particles.
- (5) Shape and texture features were extracted to reduce information and speed up the recognition process.
- (6) Correlation-based feature selection was used to remove redundant features and different experiments were applied to adjust the best correlation factor.
- (7) Different classification models with different adjustments were applied on data of extracted features.
- (8) Finally, evaluation techniques were used for assessing results and to choose the best classification that gets out the best performance.

Section 2 is about some of the related works, section 3 shows our experiment methodology and results, section 4 provides a solution for the undefined particles problem and the last section gives our conclusion and suggestions for future work.

This paper introduces a comprehensive approach for automating procedures for detecting and recognition of particles that appear inside microscopic urine samples.

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2. Related works

Zhou *et al.* [5] proposed a comprehensive description of the principles and process to develop automatic recognition and a count system for urine-sediment visual components. With the help of image processing technology, image segmentation, representation and description, geometric features and texture features have been extracted. They used a Gaussian filter, min–max threshold and edge connector in image segmentation. They used a neural network classifier based on a genetic algorithm for micro image automatic recognition and a count system. They stated that theneural network can be effectively improved by combining genetic algorithms with proper feature selection. Accuracy rates ranged from 77% for waxy casts to 96% for leukocytes.

The main drawback of this research is that it did not consider the undefined particles in their classification and did not test how the classification model will treat when input data instances have undefined particles. Also they tested only neural network without trying other classifiers. Their results show that they considered all crystals particles are one type and one accuracy rate was for crystals without considering different types of crystals, this causes unstable recognition accuracy when different crystal forms are tested. They did not consider precision or recall so their evaluation is not guaranteed. They did not show the rate of hold out samples for testing and this affects accuracy too much.

Ranzato *et al.* [6] proposed a general-purpose system to recognize biological particles. Their system was developed to classify 12 categories of particles found in human urine. Promising locations in the image are detected and small regions containing interesting samples are extracted. The detection algorithm extracts a square bounding box around a particle and then an invariant feature vector is computed without segmenting the particle from the background of the patch. A Bayesian classifier was used with an input dataset of 500 images per class. The average error rate was 6.8%. The same experiment was applied also on a set of images of airborne pollen with a resulting error rate of 22% and 35.1% when unknown particles were considered.

The main weaknesses of this research are that their accuracy is not stable and not expressive because they used random selection of the 10% testing samples in the 10 experiments in classification process, and this causes duplication in using the same sample different times, so using cross-validation will be better. Also, they did not consider the unknown particles in classification process of urine samples and did not show how their system may deal with this issue. In addition, they did not try classifiers other than Bayesian to get the best classifier.

Song *et al.* [7] proposed a technique for incorporating contextual information into object classification applied to microscopic urinalysis image recognition. They proposed a framework that incorporates partial context at a linear cost, unlike previous techniques that cost exponential computation. The automated urinalysis system they developed consists of three steps: image processing and feature extraction, learning and pattern recognition and context incorporation. Features are fed into a neural network with 16 inputs. They stated that augmenting these features by incorporating context into the

classification process yielded significant benefits. The algorithm using partial context was tested on a database of 83 urine specimens, containing a total of 20,276 images. Four classes were considered: bacteria, red blood cells, white blood cells and amorphous crystals. The diagnosis for a specimen is either normal or abnormal. A significant improvement of recognition rate achieved by decreasing the error rate from 44.48% before using the partial context to 36.66% after.

The weakness in this research is that success rates are low, where the best success rate was 63.34%. Also they did not work on counting the inner particles, but their system diagnoses the specimen by just telling if it's normal or not.

Cao et al. [8] worked in research on detection of red blood cells in urine image captured under a microscope. They used an improved Sobel operator for image pre-processing to smooth noise by using eight directions instead of four and edge tracking through sensing similarity between edge points' intensities unlike noisy points. Red blood cells were localized using Hough Transform. Features were extracted and selected by dimension reduction using Principal Component Analysis (PCA). Finally, values of dimensionality reduction were classified by LDA (Linear Discriminant Analysis). The experiment adopted 90 images, 720 images as experimental samples. The authors selected 55 original images as a training set (440 samples) and 35 as a test set (280 samples) randomly. The best result with the 440 training was 94% and with the 280 test samples was 91%. Classification rates in the process of training and test changed when PCA energy intensity changes from 0.1 to 0.99.

What we can criticize in this research is that authors did not evaluate their improved Sobel operator and what are improvement success rates. Also, they did not show how Hough Transform localization treats with other particles. There is no consideration for images that contain other particles like WBC which have a similar geometrical circular shape to RBC and test how classifier treats with this issue. In addition, they did not try other classifiers to test which is better to choose. As for accuracy, this research will show an improvement over their accuracy rate of more than 5%.

3. Experiment

Images were captured from two different places, first the University College of Applied Sciences medical lab and second the Alamal medical lab (which is a private lab and located in Gaza).

The images format is PNG, with a resolution of 1280×960 and RGB colour. Table 1 lists the number of images that contain different particles. The nature of solid particles is that they have solid boundaries that make them prominent from the background. This formation can help us in extracting the binary shape of these particles and, hence, shape descriptors can be extracted.

Figure 1 shows the work flow of overall experiment, each step matches more details in the next sub-sections.

3.1. Prepare images

Choose a set of images that contain RBCs, WBCs, Crystals–Calcium Oxalate and Crystals–Triple Phosphate.

Table 1. Number of images in each group.

Urine particle	Number of images
RBC WBC Calcium oxalate	340 296 220
Triple phosphate	152



Figure 1. Experiment work flow.

3.2. Pre-processing

3.2.1. Image pre-processing

Pre-processing was applied on each image by following the next steps:

- (1) Convert image from a 32-bit to a 16-bit grey image as this was required by the third step (curvatures computing).
- (2) Apply a Gaussian blur filter with sigma = 2.5 where this step makes the image smoother and decreases the number of curvatures produced by the third step.
- (3) Apply principal curvatures computing on the image to produce an image of surface curves.
- (4) Run the Default auto-threshold which is a modified version of the isoData algorithm (by decreasing the highest tone of the histogram to be 1.5-times of the tone with second order of arrangement, but this occurs just when the highest tone was more than twice the second tone and the second tone is not zero. This modification was done by ImageJ developers) to segment image data (ImageJ is a public domain Java image processing program, URL: http://rsbweb.nih.gov/ij/docs/intro.html).
- (5) Convert image to binary in order to make it ready for blob (Binary Large Object) extraction.
- (6) Image was re-sized to 640×480 , which is half the original size, to make image data less and to decrease the size of processing.

These steps were applied using the Fiji system using a batch process which automatically processes on all images inside some folder and saves output images to another folder. (Fiji is an image processing package. It can be described as a distribution of ImageJ together with Java, Java 3D and a lot of plugins organized into a coherent menu structure, URL: http://fiji.sc/wiki/index.php/Fiji.)

Figure 2 shows the effect of applying pre-processing steps on an image containing calcium oxalate crystals. Preprocessing steps were applied with changing three values to get the most suitable pre-processing for input images. Process was applied on 16 images captured from a microscope for the four types, five images for calcium oxalate crystals, five images for triple phosphate crystals, two images for RBCs, four images for WBCs. The 16 images entered the pre-processing steps and a blob extraction was applied to extract binary blobs, then extracted binary blobs were counted for each type. Table 2 shows resulted counts for each adjustment.

After blob extraction there is a set of particles that are lost because of breaking their parts or losing borders or touching the background. The best segmentation adjustment should get out the highest numbers of particles. So, we can deduce that the best adjustment can be achieved by using:

- (1) Gaussian Blur sigma = 2.5;
- (2) Compute Curvatures sigma = 1;
- (3) Using the Default auto-threshold algorithm.

Default auto-threshold is a variation of the IsoData algorithm used by ImageJ 1.41 and earlier.

3.2.2. Blob extraction and manual labelling

The blob extraction process is used to extract the 8-connected objects in the binary image and save each blob into a particular image file, and extracts its position and saves it into the database.

This process was applied using a code written in VC++ and IPL98 library, and data was recorded in the MSAcess database file. (The Image Processing Library 98 (IPL98) is a platform independent image manipulating C/C++ library. URL: http://ipl98.sourceforge.net/.)

The code started by segmenting the image pixels into groups up to eight-connectivity method with discarding those connected pixels groups that contain fewer than 100 pixels to decrease the number of unneeded noisy blobs. Figure 3 is an example of 8-connectivity segmentation.

Each pixels group was saved into an image file and it's co-ordinates in the original image were recorded in a database (for future texture features extraction) by getting positioning values which are left, right, width and height.

Manual labelling was applied to extract the candidate blobs images (RBCs, WBCs and Crystals) for feature extraction and classification.

3.2.3. Features extraction

The feature extraction process extracts a set of features from the shape and texture of all blobs images. Shape descriptors were extracted from the binary image. Texture descriptors were extracted from the original image. These features were saved to an excel file.

A matlab code was written to apply this functionality.

- (1) Shape descriptors: Table 3 shows extracted features from the geometrical shape of particles.
- (2) Texture descriptors:
- 16 Histogram features (Mean, variance, skewness and kurtosis. For each of channels: Red, Green, Blue and Grey).
- 22 Co-occurrence matrix parameters (Table 4).

Table 4 lists all extracted co-occurrence matrix parameters. These parameters were calculated for offset 0 3.

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Figure 2. Example of image under pre-processing. (a) Original image. (b) After converting image to 16 bit. (c) Result of Gaussian blur. (d) By applying curvature computing. (e) After auto-threshold. (f) Converted to binary.



Table 2. Pre-processing experiments with different parameters.

Gaussian blur sigma	Compute curvatures sigma	Auto-threshold algorithm	# of oxalates	# of RBCs	# of WBCs	# of phosphates
1	1	Default	21	62	16	15
2.5	1	Default	37	90	26	18
3.5	1	Default	31	88	28	15
2.5	2	Default	29	69	28	15
2.5	3	Default	44	60	30	16
2.5	1	Huang dark	42	56	25	15
2.5	1	Intermodes	28	75	13	16
2.5	1	Li	29	72	16	18
2.5	1	MaxEntropy	8	11	1	1
2.5	1	Mean	27	80	20	16
2.5	1	IsoData	34	83	19	22

To extract texture features, each blob positioning values was extracted from the MSAcess database file and used to crop the particle image from its original image.

3.3. Data mining classification process

The classification process was built on rapidminer software to apply classification operations and get the best model. Process steps are shown in Figure 4.

The input dataset consists of 807 instances, 268 for white blood cells and labelled 'Class = WBC', 188 for red blood cells and labelled 'Class = RBC', 201 for calcium oxalate

crystals and labelled 'Class = Crystals–Calcium Oxalate' and 150 for triple phosphate crystals and labelled 'Class = Crystals–Triple Phosphate'.

The experiment was enumerated on different changing parameters to adjust parameter values and get the best adjustment.

Changing parameters were:

 Remove correlated attributes operator: changing the value of correlation changes input attributes to the x-validation which changes the learning model and performance.



Figure 3. Blob extraction and saving into image file.

Table 3. Shape descriptors.

Area	U.,1
Alea	nui
Orientation	Hu2
Perimeter	Hu3
Convex area	Hu4
Filled area	Hu5
Solidity	Hu6
Eccentricity	Hu7
Equivalent diameter	Roundness

Table 4. Co-occurrence matrix parameters.

GLCM parameters	GLCM parameters				
Autocorrelation	Maximum probability				
Contrast	Sum of squares				
Correlation (matlab)	Sum average				
Correlation (paper)	Sum variance				
Cluster prominence	Sum entropy				
Cluster shade	Difference variance				
Dissimilarity	Difference entropy				
Energy	Information measure of correlation 1				
Entropy	Information measure of correlation 2				
Homogeneity (matlab)	Inverse difference normalized (INN)				
Homogeneity (paper)	Inverse difference moment normalized				

(2) Classification model: five models were used to test the resulting performance vector which are K Nearest Neighbors (K-NN), Neural network, Naïve Bays, Decision tree and rule induction, but a special changing parameter were tested for k-NN which is K [the used number of nearest neighbours].

4. Results and evaluation

After trying the five classifiers K-NN, Decision Tree, Naïve Byes, Rule Induction and Neural Network. Each experiment of the five was applied with



Figure 4. Solid particles classification process in rapidminer.

- Remove correlated attributes operator: with changing correlations 0.8, 0.85, 0.9, 0.95 and 1 (five enumerations).
- (2) Cross-validation with 10 validations and stratified sampling.

In addition, K-NN enumerated the experiment 11-times by changing k between 5–15.

Table 5 lists the best performance vector acheived from each classification model. It shows also the correlation value that produced the best performance.

It is noticed that accuracy, recall and precision values produced by the neural network are better than those produced by others, so it can be deduced that the Neural Network classifier is the best between all tested classification models to make the best recognition of urinary solid particles.

Table 6 shows the best performance vector when using a Neural Network classifier. It's accuracy was $96.41\% \pm 1.51\%$.

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As for attribute selection, the 'Remove correlated attributes' operator resulted in removing just one attribute, which is Correlation (paper) extracted from gray level co-occurrence matrix (GLCM) features of offset 0 3.

5. Undefined particles

There were some other particles that were not listed above; these particles can be called 'undefined' or unknown. These are noisy particles that should be trained in the classifier to be filtered out of results.

To consider filtering undefined particles out of results, another process was built to testify how the classification models will treat with that situation. Two cascade classifiers were used, the first was used to remove undefined particles from the input dataset as much as possible, the second classifier was used to classify the remaining instances of input dataset to one of the classes: WBC, RBC, Crystals-Calcium Oxalate, Crystals-Triple Phosphate and Undefined.

The first classifier was the AODE (aeraged one-dependence estimators) classifier which classifies instances to 'undefined' or not. This classifier was trained on 1435 samples of only undefined particles. Then a classifier model was applied on input testing particles (input dataset) to remove the undefined particles and keep other particles on output to enter the second classifier. The second classifier was trained on all dataset instances of WBC, RBC, Crystals-Calcium Oxalate, Crystals-Triple Phosphate and Undefined and was applied on those particles which got out from the first classifier. K-NN, Decision tree, Neural Network, Rule

Table 5. Best performance vector from each classification model.

Classification model	Correlation	Accuracy	Oxalate recall	Oxalate precision	RBC recall	RBC precision	WBC recall	WBC precision	Phosphate recall	Phosphate precision
K-NN (with $K = 11$)	1	93.06	87.06	93.09	93.09	93.58	98.88	90.44	90.67	97.84
Decision Tree	0.95	90.33	81.09	86.24	93.09	88.38	93.66	91.94	93.33	95.24
Naïve Byes	1	85.38	85.07	76.68	80.85	87.86	91.04	89.05	81.33	89.05
Rule Induction	0.95	90.99	83.56	86.32	91.63	92.08	95.49	91.06	91.46	94.94
Neural Network	1	96.41	94.53	93.14	97.34	96.32	96.64	97.74	97.33	98.65

Table 6. Performance vector of best adjustment for classification of solid particles.

	True Crystals-Calcium Oxalate	True RBC	True WBC	True Crystals-Triple Phosphate	Class precision
Predicted Crystals-Calcium Oxalate	190	4	6	4	93.14
Predicted RBC	5	183	2	0	96.32
Predicted WBC	5	1	259	0	97.74
Predicted Crystals-Triple Phosphate	1	0	1	146	98.65
Class recall	94.53	97.34	96.64	97.33	

Table 7. Counted particles by KNN classifier, k = 5, remove correlated = 0.85.

	Pred. Undefined	Pred. Crystals –Calcium Oxalate	Pred. Crystals –Triple Phosphate	Pred. RBC	Pred. WBC	Success rate	Error rate
Input Images							
Calcium Oxalate (11 images)	196	43	10	12	13	89.58	12.77
Triple Phosphate (10 images)	124	1	45	1	1	75.00	1.74
RBC (5 images)	181	20	25	79	41	52.67	24.86
WBC (6 images)	164	4	2	7	34	80.95	6.16

Induction and Naïve Byes were exchanged to act as a second classifier in order to test which of them produce the best success rates. Using the cascade classifiers helped in minimizing error rate.

To act as the real system, a set of testing images were selected to enter the process of classification, these images will be processed to extract particles from them, these particles will be of the five types: WBC, RBC, Crystals-Calcium Oxalate, Crystals-Triple Phosphate and Undefined. The classification process should count how many particles there are in each image for each of the five types. Thirty-two images were captured for testing (11 contain Calcium Oxalate, 10 Triple Phosphate, five RBC, six WBC).

Table 7 shows the counted particles inside input images after applying the experiment with the K-NN classifier. In Table 7, for calcium oxalate, there were 11 input testing images, these images contains calcium oxalate particles and some other undefined particles inside them, these images were captured from the same specimen slide for one patient, each image passed through pre-processing, blob extraction and feature extraction steps and, finally, all features' instances of all extracted particles from the 11 images were entered to rapidminer software to apply classification and count how many particles there are for each type. Results for calcium oxalate are shown in the shaded row, it is noticed that 196 objects were not classified to any of the four class labels, 43 objects were classified as calcium oxalate, but the real number is 48 (counted manually), so the success rate can be calculated by

Number of classified objects as "calcium oxalate" Number of real existing "calcium oxalate objects"

As for RBC, WBC and triple phosphate, these counts are considered error rates because they are not found in the 11 images of calcium oxalate samples. Error rate can be calculated by

> Number of error objects Number of all objects

In the same way, other input images of RBCs, WBCs and triple phosphate were calculated. Like the K-NN results in Table 7, other classification models resulted in different success and error rates. The Neural Network model produced the highest success rates and low error rates for all types except for WBC. It was noticed that WBC was classified successfully with a low error rate by k-NN.

6. Conclusion

Urine analysis is an important test in medical labs and deserves interest as it reveals the presence of many problems and diseases in the human body. One of the main parts of that test is handled by human eye observations through a microscope. As human eye observation is subjective, timeconsuming and causes mistakes, researchers studied methods of automating microscopic analysis with the aid of computer and software systems.

This research introduced a comprehensive approach for automating procedures for detecting and mining microscopic urine particles. Microscopic images were captured and filtered, 340 of them contain RBC particles, 296 contain WBC, 220 calcium oxalate, 152 triple phosphate and others. This experiment considered solid particles that have strong boundaries. Gaussian blur and curvature computing was applied for enhancing images with several experiments for adjusting factors. A modified version of IsoData threshold algorithm was proved to be the best choice, after running a set of histogram-driven threshold algorithms on images. 8-connected blob extraction with manual labelling emerged 268 WBC blobs, 188 RBC blobs, 201 calcium oxalate blobs and 150 triple phosphate blobs. Sixteen shape descriptors and 38 textural features were extracted. By trying the five classifiers K-NN, Neural Network, Naïve Bays, Decision Tree and Rule Induction with 10-fold cross-validation, evaluation resulted performance of 96.41% of accuracy with minimum f-measure of 93.83% by using the neural network.

In addition, undefined particles were entered into the experiment using cascade classification, by running an AODE classifier before the main classifier for filtering undefined particles, but this caused degradation in performance of WBCs recognition.

In future works, the following issues will be considered:

- More particles will be collected from those which were not studied by this research.
- Also, add the functionality of detecting moving particles such as bacteria which appears like small slow moving sticks inside urine specimen.
- In addition, overlapped and touching particles produce undefined forms of particles after segmentation and applying threshold. This problem increased error rate and finding a technique to split touching particles or extract some particle from overlapping will increase performance of segmentation.

Declaration of interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of this article.

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