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A New Method for Sperm Detection in Infertility Cure: Hypothesis Testing Based on Fuzzy Entropy Decision

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ABSTRACT

In this paper, a new method is introduced for sperm detection in microscopic images for infertility treatment. In this method, firstly a hypothesis testing function is defined to separate sperms from plasma, non-sperm semen particles and noise. Then, some primary candidates are selected for sperms by watershed-based segmentation algorithm. Finally, candidates are either confirmed or rejected using fuzzy entropy decision algorithm. Performance of the proposed method is evaluated on real captured images containing sperms and other specimens of semen in two different scenarios. In the first scenario, semen has low density of sperms however the second scenario belongs to semen with high density of sperms. The obtained results show the greater ability of the proposed method in sperm detection compared to present approaches in both of scenarios. Furthermore, it is shown that 8% and 15% improvements in sperm detection in the first and second scenarios can be achieved by the proposed algorithm. As the final results, the proposed algorithm not only doesn't lead to extract more false objects but also decrease the rate of false detections are decreased compared to existing algorithms.

1. Introduction

A great number of problems with infertility arise from the male therefore analyzing of the male semen has received special attention in infertility cure [1]-[2]. The main particles of male semen are sperms which their parameters can be used as indicator of fertility [3]. In recent years, microscopic imaging has provided ability to investigate the sperms behavior in semen [4]-[5]. In these methods images which have been captured from semen specimens are analyzed manually by an expert person.

Unfortunately, the progress of this method has been hampered due to its time consuming nature and human errors [6]. So, automated methods have been substituted for semen analyses which are based on computerized detection and classification of sperms in recorded microscopic images [7]. The main challenge of automatic methods is distinguishing the sperm from other semen particles. The main factors which limit the sperm detection in microscopic images are the low contrast of microscopic images under various conditions and the possibility of neighboring sperms to touch each other [8].

Several methods have been proposed for automatic sperm detection. Some methods utilize region growing based techniques for separating sperms from other semen particles. In many microscopic images there is no evident contrast between sperms and these particles therefore the segmentation using these techniques does not lead to satisfactory results [9]. There is a class of methods that distinguishes sperms based on information provided by contour of sperm head. However, this approach is not capable to extract sperm tail completely [10]. Some methods make use of background detection analysis for detecting sperms. The main limitation of such methods is their high sensitivity to SNR of images [11].

Although thresholding methods have been widely used in image segmentation [12] but unfortunately performance of these methods is highly dependent on the applied threshold. Therefore, applying such methods in sperm detection often leads to extract many false objects [13]. Some researches utilize watershed method for separating sperms from other specimens of semen, but this method often leads to fragmented results. Although a number of modifications have been applied on this method to solve above limitations but they lead to merging the neighbor sperms [14]. More sophisticated methods include various types of matching. In these methods, constant or flexible masks have been used to separate sperms from other semen particles. These approaches face some challenges such as high sensitivity to shape, size and rotation of sperms [15].

In this paper, a new method for sperm detection is introduced which is based on a watershed-based segmentation modified by fuzzy entropy concept. In the proposed method, firstly probable sperms are extracted using watershed-based segmentation as primary "candidates". In the next step, an algorithm which is based on fuzzy entropy decision is applied to confirm correct sperms and reject false candidates. Unlike the existing methods the proposed algorithm is capable to detect sperms in high density semen without either fragmentation or merging sperms and doesn't need to primary knowledge about sperms.

The paper is organized as follows. In section II, the proposed algorithm is introduced including watershed-based segmentation for candidate selection followed by fuzzy entropy decision to confirm sperms. In section III, the performance of the proposed method is evaluated in two different scenarios based on real microscopic images of semen specimens. In section IV, the obtained results from experiments are compared to the results of existing methods using their effective parameters. Conclusion is presented in the last section of the paper.

2. Material and Methods

Suppose I as a microscopic image captured from a semen specimen who contains sperms, plasma and debris which two latter parameters are called artifact in this article. Each pixel of I may be written as:

$$I_{lj} = I(l,j) \qquad 1 \le l \le L, 1 \le j \le J \tag{1}$$

In the above equation, I_{lj} is the value of a pixel in I which is located in l^{th} row and j^{th} column of a $L\times J$ image. In sperm detection problem, the question of interest may be explained as two competing claims (i.e., hypotheses) which between them we have a choice. First hypothesis (H_0) is depend on I_{lj} to artifact and noise and alternative hypotheses (H_1) which means dependence of I_{lj} to a sperm. These situations are explained mathematically via a hypothesis testing equation as:

$$\begin{cases} H_0: I_{lj} = |c_{lj} + n_{lj}| \\ H_1: I_{lj} = |r_{lj} + c_{lj} + n_{lj}| \end{cases} \tag{2}$$

In the above equation, r_{lj}, c_{lj}, and n_{lj} show the sperm, artifact and noise components in I_{lj}, respectively. To accept or reject each of above hypotheses, the chance of each pixel for belonging to a sperm must be determined. For this purpose, firstly imagine I as a topographic surface which is immersed in water. Each local minimum of the topographic surface may be considered as a hole where construct a catchment basin with its surrounding low gray level neighbors. When the water starts filling all catchment basins, if two catchment basins merge as a result of further immersion, a dam that surrounds the connected immersed area of each merged catchment basin is built which represents the watershed line. Actually, such watersheds may be considered as boundaries between several objects in I.

To implement this idea, an efficient algorithm is presented below. Firstly the image pixels are sorted in increasing order of their gray values. Then, M' local minimums of I are extracted as some first members of this sorted list in such manner that their greatest gray level is α. Equation (3) shows that these minimums may construct X_α as a set of catchment basins ($O'_{m'}$). Each of these objects may be either an isolated minimum of image or a set of neighboring pixels which all of them are minimums of sorted list [16].

$$X_\alpha = \{O'_1, \dots, O'_{m'}, \dots, O'_{M'}\} \tag{3}$$

Based on above procedure, it may be said that all pixels of image having gray-level less than or equal to α has already been assigned a unique catchment basin (i.e. one of X_α members).

In the next step, pixels having gray-level the α+1 must be processed. These pixels may fall in one of following cases. In the first situation, the pixel is not assigned to any existing basin. In this case, it may be considered as a member of $\beta_{\alpha+1}$(i.e., union of new local minimums). In the second situation, the pixel may be an extension of an existing basin if and only if at least one of its eight connected neighbors already is a member of $O'_{m'}$. These pixels construct $Z(x_\alpha)$as a union with same size with X_α which its m'th member shows the set of pixels which must be assigned to member m' of X_α (i.e. $O'_{m'}$). Therefore, by combination of both mentioned situations, X_α may expand to $X_{(\alpha+1)}$ as [16-17]:

$$x_{(\alpha+1)} = x_\alpha \cup Z(x_\alpha) \cup \beta_{(\alpha+1)} \tag{4}$$

By repeating such strategy recursively to maximum value of sorted list, finally X_l is obtained as the set of M objects (i.e. O_m) as:

$$X_1 = \{O_1, \dots, O_m, \dots, O_M\} \tag{5}$$

Now, feature vector f_m containing texture features [18-19], mean, variance and location is extracted from members of X_l. Considering X_l as a group of fuzzy sets, the fuzzy entropy function E may be written for X_l as [20]:

$$E = -\sum_{q=1}^{Q}\sum_{m=1}^{M} p_{qm}.\log p_{qm} \tag{6}$$

In which Q shows the number of fuzzy clusters. Furthermore, the probability of belong to each feature vector f_m to each cluster is shown as P_{qm}. The latter parameter is a member of the probability matrix P, which is defined as:

$$P = \begin{bmatrix} p_{11} & p_{12} \cdots & p_{1m} \cdots & p_{1M} \\ p_{21} & p_{22} \cdots & p_{2m} \cdots & p_{2M} \\ \vdots & & & \\ p_{q1} & p_{q2} \cdots & p_{qm} \cdots & p_{qM} \\ \vdots & & & \\ p_{Q1} & p_{Q2} \cdots & p_{Qm} \cdots & p_{QM} \end{bmatrix} \tag{7}$$

where

$1 \le q \le Q, and\ 1 \le m \le M$

Now the fuzzy clustering function is defined by combining difference measure and entropy criterion as:

$$G(P,\mu,F) = \sum_{q=1}^{Q}\sum_{m=1}^{M} p_{qm}.\ \|f_m - \mu_q\|^2 + M\sum_{q=1}^{Q}\sum_{m=1}^{M} p_{qm}.log\, p_{qm} \tag{8}$$

In which μ_q is the center of cluster q and μ is a vector containing centers of clusters. The first part of this equation is the average of fuzzy distances and the second part is (M) times multiplication of fuzzy entropy function E. By optimizing the above function, we will have [21]:

$$p_{qm} = \left(\sum_{q'=1}^{Q}\left[\frac{e^{\|f_m-\mu_q\|}}{e^{\|f_m-\mu_{q'}\|}}\right]^{1/Q}\right)^{-1} \tag{9}$$

Now, the average scattering of clusters can be written as:

$$S(Q) = \frac{\sum_{q=1}^{Q}\|\sigma_{\mu_q}\|}{Q\|\sigma\|} \tag{10}$$

In which the members of vector σ_{μ_q} are obtained as:

$$\sigma_{\mu_q}^{y} = \frac{1}{M}\sum_{m=1}^{M} p_{qm}(f_m^y - \mu_q^y)^2, y = 1,2,\dots,Y \tag{11}$$

In above equations, μ_q^y and f_m^y are the y'^{th} members of average vector of cluster q and feature vector, respectively. Furthermore, Y and $||\sigma||$ show the length of each feature vector and variance, respectively. Now, $D(Q)$ can be obtained for X_l which is segmented to $1 < q < Q$ fuzzy clusters as:

$$D(Q) = \frac{D_{max}}{D_{min}}\sum_{q=1}^{Q}(\sum_{q'=1}^{Q}\|\mu_q - \mu_{q'}\|)^{-1} \tag{12}$$

In which $D_{\max}$ and $D_{\min}$ are obtained as:

$$\begin{aligned} D_{max} &= \max(\|\mu_q - \mu_{q'}\|) \\ D_{min} &= \min(\|\mu_q - \mu_{q'}\|) \\ &\forall\, q, q' \in \{1,2,3,\dots,Q\} \end{aligned} \tag{12}$$

To construct or remain the best clusters (e.g. fuzzy objects) in such way that they have the maximum difference between each other and maximum unity in each object, the combination of (10) and (12) has been used as decision function:

$$\Lambda(Q) = \lambda.S(Q) + D(Q) \tag{14}$$

In which λ regulates the weight of each part of decision function. In our research, several experiments show that the best results can be obtained when these terms have same weights (i.e., λ=1). The above equation leads to obtaining Q as:

$$\begin{aligned} \gamma &= \lfloor \Lambda(Q_{\min}),\dots,\Lambda(Q'),\dots,\Lambda(Q_{\max}) \rfloor \\ Q &= Q' \mid \Lambda(Q') = \min(\gamma) \end{aligned} \tag{15}$$

Using this result, X_l may be clustered as Q fuzzy objects which they can be shown as:

$$Q'' = \{O_1'', O_1'', \dots, O_q'', \dots, O_Q''\} \tag{16}$$

Now, by combining equations (2) and (16), dependence of each pixel to artifact and noise (H_0) or to a sperm (H_1) is determined as:

$$\begin{cases} H_0: I_{lj} \notin Q_q^{"} \Rightarrow I_{lj} = |c_{lj} + n_{lj}| \\ H_1: I_{lj} \in Q_q^{"} \Rightarrow I_{lj} = |r_{lj} + c_{lj} + n_{lj}| \end{cases} \quad (17)$$

Based on the aforementioned equations our strategy for determining sperms contains following steps: i) interpret *I* as a topographic surface, ii) grow catchment basins by assigning pixels to them, iii) consider the segmented image as a group of fuzzy sets and iv) qualify results by using fuzzy entropy decision. The following experiments and results show how such a strategy may be effective in increasing the performance of the sperm detection.

3. Results

The proposed algorithm was applied to real data. The data set was various microscopic images of human semen which captured by an Orca ER Digital CCD Camera mounted on a Nikon invert microscope using a 100x zoom lens. The proposed method was implemented using Matlab 2009. Additionally watershed segmentation algorithm (WSA) [14] was implemented to compare with the proposed algorithm. Tests were carried on two different scenarios. In the first scenario, the semen specimens had low densities of sperms but the second scenario contained high densities of sperms. Specifications of both of scenarios have been shown in table 1. The captured videos were first processed using manual detection to obtain a ground-truth detection to compare the automatic methods with. Then, sperms were detected by applying the proposed and WSA algorithms and finally performance of each algorithm was determined by comparing its results with manual detection results.

A. First scenario

In the first scenario, the captured images had been obtained from semen specimens with densities bellow 2×10^6sperms per milliliter. Some sampled images for this scenario have been shown in Fig. 1 and Fig. 2. Moreover Fig.3 show obtained results utilizing the proposed and WSA methods on above sampled image. For instance Fig. 2 shows that the proposed method has extracted 59 sperms of total 61 sperms which have been shown in Fig. 1 without any false detection. It is obvious in Fig. 3 that WSA has extracted only 55 sperms correctly from the sampled image without any false detection. The above example showed the better performance of the proposed method compared to WSA in detecting sperms in the first scenario. Based on this fact that the sperm parameters may be used as indicator of fertility [3], the correct detection has been considered as a sperm that at least 90% of its pixels had been extracted correctly, otherwise it has been considered as a false detection.

B. Second scenario

In this scenario images were obtained from high density (2×10^6 sperms per milliliter) semen specimens. An example for this scenario has been illustrated by Fig. 4. In addition, Fig. 5 and Fig. 6 shows the obtained results from sampled image of figure (4) utilizing the proposed and WSA methods respectively. Fig. 5 shows that the proposed method extracted 181sperms from total 210 sperms in Fig. 4 plus 7 false sperms. Fig. 6 shows the above values have been obtained equal with 150 correct and 8 false sperms. These results show like the first scenario that WSA has still had weaker results than the proposed method.

Table 1:
Specifications of Test Scenarios

Specification	Value	Specification	Value
Min and Max sizes of sperms	40 , 190 pixels	Number of captured frames	Scenario1: 180 Scenario 2: 180
Frame size (pixels)	1344* 1024	Min-Max population of sperms per frame	Scenario1: 41-73 Scenario 2: 136-221
Number of samples	11 persons	Average contrast	17%

4. Discussion

Real data which had been obtained from semen microscopy were analyzed. The proposed and WSA methods were applied on data and the results were compared by using ROC. In detection theory, a receiver operating characteristic (ROC), is a graphical plot which illustrates the performance of a detection system as its discrimination threshold is varied. It is created by plotting the indicator of detection rate (i.e., true positive rate) vs. the indicator of false detections (i.e., false positive rate) at various threshold settings. The True Positive Rate is defined as the ratio of correctly identified sperms (i.e., true positives) to sum of correctly identified and incorrectly rejected sperms (i.e., false negatives).

$$TPR = \frac{TP}{TP + FN} \tag{18}$$

In which TP and FN represent true positives and false negatives, respectively. The False Positive Rate (FPR) is defined as the ratio of incorrectly identified sperms (i.e., false positives) to sum of incorrectly identified and correctly rejected sperms (i.e., true negatives).

$$FPR = \frac{FP}{FP + TN} \tag{19}$$

In which *FP* and *TN* represent false positives and true negatives, respectively. Figures (7-8) show ROC for the first and second scenarios, respectively. These figures show clearly the superiority of the proposed method compared to WSA in both of scenarios. To evaluate the performance of algorithms, firstly 3% was considered as a typical acceptable value for false detections (i.e., FPR). As shown in Fig. 7, the proposed and WSA algorithms achieved detection rates (i.e., TPR) equal to 95% and 87%, versus this amount of false detections in the first scenario. In the same manner, Fig. 8 shows detection rates of 85% and 70% at the same false detection parameter for the second scenario. These results showed that the detection rate of the proposed method has been considerably (e.g. 8%, 15% in the first and second scenarios, respectively) higher than WSA.

Now, to evaluate the performance of algorithms versus false detections, the minimum acceptable value for detection rate (i.e., TPR) was considered equal to 90%. This led to false detection (i.e., FPR) equal with 0% and 12% for the proposed and WSA methods in the first scenario (Fig. 7). These values were 8% and 29% in the second scenario (Fig. 8). These results showed the false detections of the proposed method have been considerably (e.g., 12%, 21% in the first and second scenarios, respectively) higher than WSA.

Finally, ROC curves show that in the second scenario detection rates have been decreased typically 10% and 17% compared to the first scenario. In the same manner, false detections have been increased in the second scenario by 8% and 17% compared to the first scenario. In spite of these degradations, the superiority of the proposed method versus WSA has been more pronounced in the second scenario.

The superior performance of the proposed algorithm in detection rate in the second scenario had been 7% larger than its improvement in the first scenario. In the same manner, the Superior performance of the proposed algorithm versus WSA in false detection parameter in the second scenario had been 9% better than its improvement versus WSA in the first scenario.

5. Conclusion

In this paper a new method was introduced for sperm detection in microscopic images of human semen. In the proposed method, firstly some regions of image were indicated as "candidates" using a watershed-based segmentation algorithm. In the second, step a fuzzy decision procedure was utilized to confirm only correct sperms. To evaluate the performance of the proposed algorithm, two scenarios were carried based on real microscopic images containing different densities of sperms. The first scenario belonged to semen specimens with low density of sperms and the second one dealt with specimens with high density of sperms. In both of scenarios, the performance of the proposed algorithm was compared to WSA method using their ROC curves. By exploiting the obtained ROC curves, it was shown that the proposed algorithm has extracted sperms, 8% (in first the scenario), and 15% (in the second scenario), higher than WSA in the presence of a typically low false detection parameter equal with 3%.

Furthermore, it was shown that false detections of the proposed algorithm were 12% (in the first scenario) and 21% (in the second scenario) better than WSA considering the minimum acceptable detection rate equal with 90%. These results showed that better sperm detection obtained by the proposed algorithm hasn't led to more false detections.

Although the proposed algorithm has shown the better performance compared to WSA, but this superiority is more considerable in the second scenario. The superiority of the proposed algorithm versus WSA in detection rate in the second scenario was 7% however this superiority in false detections was 9%. Consequently, it can be concluded that the proposed method may be used as a suitable alternative for detecting sperms in microscopic images especially in semen specimens with high density of sperms.

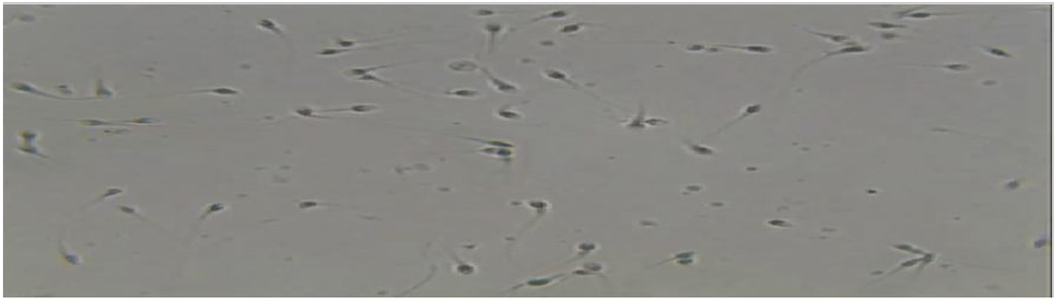

Figure 1: Captured microscopic image for a semen specimen containing low density of sperms (first scenario)

Figure 2: Extracted sperms using proposed algorithm in image shown in Fig. 1

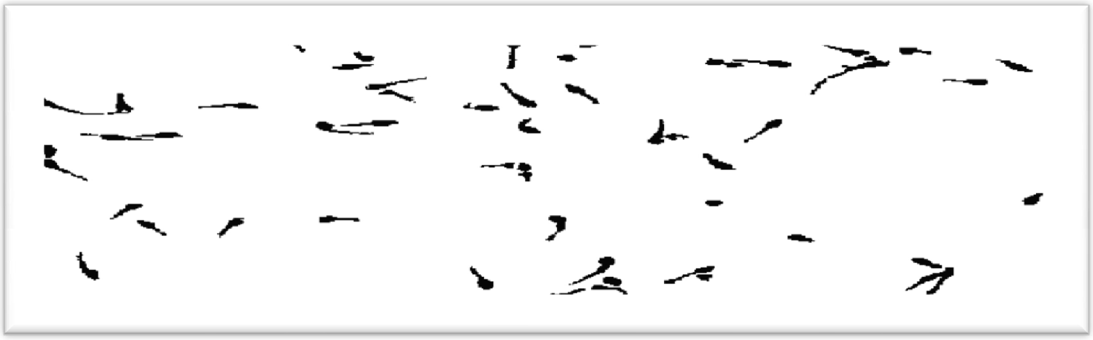

Figure 3: Extracted sperms using WSA algorithm in image shown in Fig. 1

Figure 4: Captured microscopic image for a semen specimen containing high density of sperms (second scenario)

Figure 5: Extracted sperms using proposed algorithm in image shown in figure 4

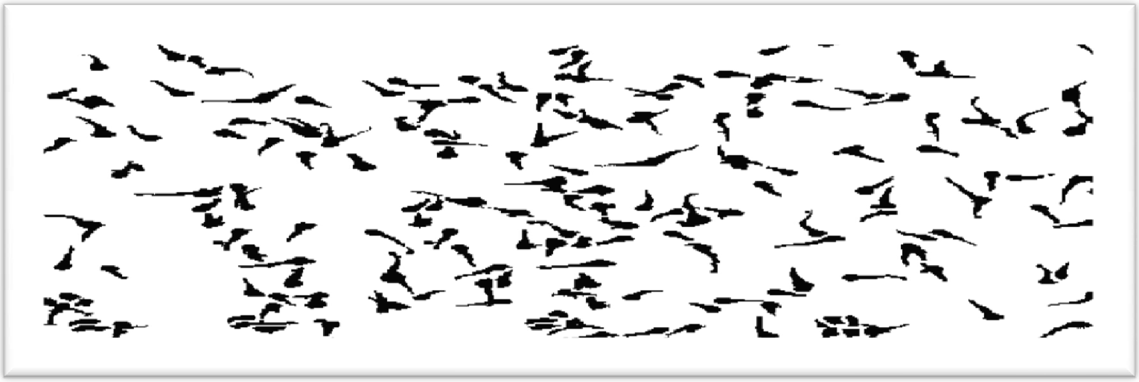

Figure 6: Extracted sperms using WSA algorithm in image shown in figure 4

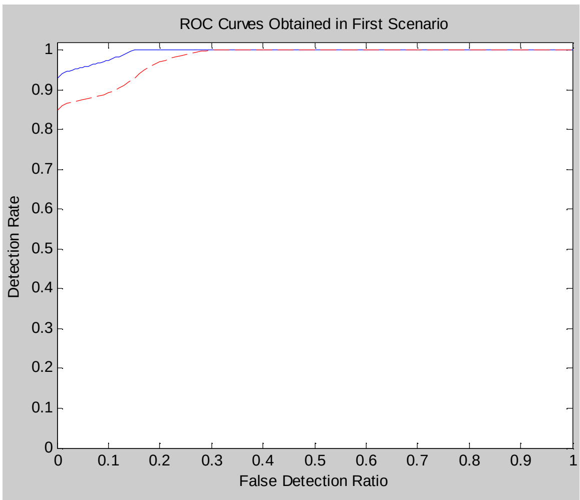

Figure 7: ROC curves in the first scenario for the proposed (solid-line) and WSA (dashed-line) algorithms

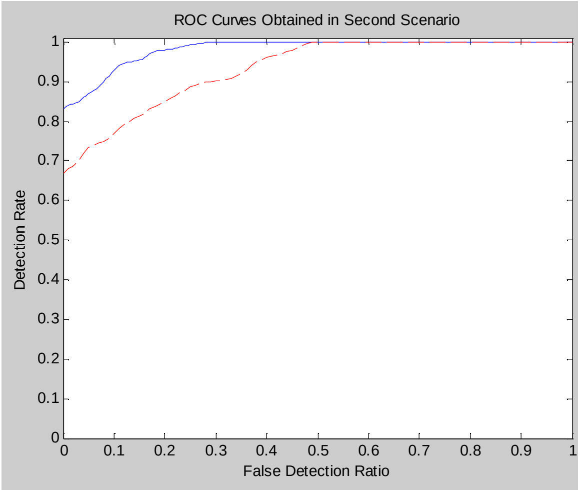

Figure 8: ROC curves in the second scenario for the proposed (solid-line) and WSA (dashed-line) algorithms

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