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# A Review on Methods of Identifying and Counting *Aedes Aegypti* Larvae using Image Segmentation Technique

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## Abstract

*Aedes aegypti* mosquitoes are a small slender fly insect that spreads the arbovirus from flavivirus vector through its sucking blood. An early detection of this species is very important because once these species turn into adult mosquitoes a population control becomes more complicated. Things become worse when difficult access places like water storage tank becomes one of the breeding favorite places for *Aedes aegypti* mosquitoes. Therefore, there is a need to help the field operator during the routine inspection for an automated identification and detection of *Aedes aegypti* larvae, especially at difficult access places. This paper reviews different methodologies that have been used by various researchers in identifying and counting *Aedes aegypti*. The objective of the review was to analyze the techniques and methods in identifying and counting the *Aedes Aegypti* larvae of various fields of study from 2008 and above by taking account their performance and accuracy. From the review, thresholding method was the most widely used with high accuracy in image segmentation followed by hidden Markov model, histogram correction and morphology operation region growing.

**Keywords:** *Aedes Aegypti* larvae, Water Storage Tank, Image pre-processing, Image processing

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## 1. Introduction

*Aedes aegypti* mosquitoes are small slender fly species that suck the blood of mammals as their essential feeds [1]. *Aedes aegypti* mosquitoes are one of the most dangerous living organisms in the world which have contributed to a huge number of death annually. These mosquitoes are agents which spread the flavivirus vector that is related to arboviruses such as dengue fever, chikungunya fever, yellow fever and Zika fever [2, 3]. An early detection of this species is very important because once these species turn into adult mosquitoes, a population control becomes more complicated. Urbanization, demography, and environment are the main factors that contribute to the global distribution of this arbovirus [4]. The main problem arises when difficult access places like water storage tank become one of the breeding favorite places for *Aedes aegypti* mosquitoes. Things become worst when field operators from the health sector face a difficulty to do a routine inspection to identify the larvae, especially at difficult access place [5].

In order to curb the spread of flavivirus vector, the agents need to be eliminated. Thus far, several practices are implemented to control the widespread of this vector such as environmental management, chemical control and also biological control [5]. However, studies that involve technology in this field are scarce. *Aedes Aegypti* larvae identification and segmentation using technology are rarely studied in the field of entomology. In the world of signals, signals can be categorized dimensionally like one-dimensional (1D), two-dimensional (2D), three-dimensional (3D) and also multidimensional signals based on their characteristics and behavior [6].

A 1D signal characterized has only one independent variable and is often in time waveform form. Acoustics is one example of communication and it poses the 1D signal [7]. Moreover, 2D signal consists of two independent variables. This 2D intensity signal is commonly known as an image. Image processing method is widely used all over the world in many fields of study [8]. In entomology field, this method has also been implemented in the counting of *A. Aegypti* eggs in the image of ovitraps [9]. When an image is added with a time variable, the signal represents 3D and it is also known as video signals.

In this context, it is found that the best way to identify and count the *Aedes aegypti* larvae is by posing the 2D signal of image processing. The significance of choosing this method is due to its wide-spread and deep exploring techniques usage by many researchers. Nevertheless, only digital image can convert the image to numerical representation which allow it to be analyzed compared with the analog picture which uses film that can be only printed [10].

## 2. Pre-Processing

Pre-processing techniques used are briefly discussed in this section. In image processing, pre-processing is the most vital step that must be taken to make sure that the image information content and quality are prepared for further processing [11]. As this project is dealing with the underwater organism, thus a camera will be placed under water. In underwater image processing study, image enhancement and image restorations are very important because the medium of water itself causes the light absorption and scattering which make the visibility of underwater is only within 20 meters [12]. The underwater hazy image formation model that is contributed by Koschmieder can be simplified as [13]:

$$I_{\lambda}(x) = J_{\lambda}(x) * t_{\lambda}(x) + (1 - t_{\lambda}(x)) * B_{\lambda}, \lambda \in [\text{red}, \text{green}, \text{blue}] \quad (1)$$

where  $I_{\lambda}(x)$  is input image captured by the camera,  $J_{\lambda}(x)$  is radiance scene at point  $x$ ,  $t_{\lambda}(x)$  is reflecting residual energy ratio  $J(x)$  from point  $x$ ,  $B_{\lambda}$  is homogenous background light and  $\lambda$  light wavelength of the red, green and blue spectrum.

Many methods can be implemented for underwater image enhancement and restoration. However, it is found that the traditional method of generalized unsharp masking, probability-based method and histogram equalization cannot effectively adapt the degradation of underwater images [14]. The studies of [15] and [16] have proposed a method of the dark channel prior (DCP) in which this approach is physically able to handle heavily hazy image. It is found that at least one color channel of red green or blue (RGB) has very low intensity except for the region of sky image. The dark channel concept can be formally described as:

$$J^{dark}(x) = \min_{y \in \Omega(x)} (\min_{c \in \{r, g, b\}} J^c(y)) \quad (2)$$

where  $J^{dark}$  is a dark channel of the image,  $J^c$  is the color channel of the image (red, green, and blue) and  $\Omega$  is local patch centered. The dark channel prior technique has been improved [17] in order to make it capable as an underwater environment. The author has found that the red channel can be a variant for the dark channel prior which restored the contrast lost in an image.

In addition, instead of the dark channel prior, there is another method called Unsupervised Colour Correction (UCM) method [18]. This technique has basically improved the contrast of RGB color model and hue-saturation-intensity (HSI) color model. It also effectively removes the bluish color in the image based on color balance and histogram stretching. This approach also improves the low illumination and low red problem in order to obtain better results in segmentation. Improving the red channel can be referred as contrast enhancement technique. In another study, UCM has been extended and modified [18]. This study has applied histogram stretching to red, green and followed by blue which significantly minimized over enhanced and under enhanced contrast in an image [19]. The histogram stretching in the RGB colour mode can be defined as:

$$P_{out} = (P_{in} - i_{min}) \left( \frac{o_{max} - o_{min}}{i_{max} - i_{min}} \right) + o_{min} \quad (3)$$

where  $P_{in}$  and  $P_{out}$  are the input and the output pixels of the image,  $i_{min}$  and  $i_{max}$  are the minimum and maximum intensity values for input images meanwhile,  $o_{min}$  and  $o_{max}$  are the minimum and maximum intensity values for output image respectively.

Acquiring clear images in underwater is very important. A study has proposed an enhancement method by wavelength compensation and dehazing which eliminates the distortion that is caused by colour change and light scattering [13]. This algorithm is a derivation work from the dark channel prior technique. Besides, this algorithm is designed to remove the presence of artificial light in the image which increases the red channel in the picture. As the red colour has the longest wavelength, the high intensity of it causes the noise in the image. In another study, there is also another technique called single image dehazing which estimates the depth prior of a scene from the image which exploits the wavelength-dependent attenuation of light in water [20]. This technique filters all the turbid water, fog and large suspended filter that exist in the transmission medium.

Nevertheless, Laplacian and Median filters have applied 3x3 kernels to the squared image [21, 22]. The purpose of applying this filter is to improve the contrast of the image that occurs due to imbalanced illumination so that the edges of the region are highlighted. The Laplacian filter of the image can be described as:

$$L(x, y) = \frac{\partial^2 I}{\partial x^2} + \frac{\partial^2 I}{\partial y^2} \quad (4)$$

where  $\frac{\partial^2 I}{\partial x^2}$  is a derivative of x Cartesian and  $\frac{\partial^2 I}{\partial y^2}$  is a derivative of y Cartesian. Meanwhile,

the median filter operation can be described as:

$$y[m, n] = \text{median}\{x[i, j], (i, j) \in w\} \quad (5)$$

where  $w$  represents the neighbourhood defined by users [3x3 kernel] and  $[m, n]$  is the central location in the image. Hence, the image processing can be performed effectively and efficiently in segmenting the region of interest. Both papers, shows that the authors have to repeat the filter several times to ensure that the image is enhanced enough to proceed for thresholding. From the review that has been made, it is observed that various techniques exist in image enhancement and restoration.

### 3. Processing

Image processing is part of pattern recognition which is widely applied in various fields including agriculture, biomedical, engineering, rehabilitation and many others [23-25]. Only few studies in entomology field implement the image processing technique, thus, the best way of selecting is by reviewing the technique and method that are related and similar to the shape of *Aedes aegypti* larvae. This section explores on several methodologies which are significantly implemented by different authors in identifying and counting *Aedes Aegypti* larvae. The most challenging part in the review is that the technique that is selected must have high accuracy. The significant fields that relate to this project and widely implement the image processing are agriculture and biomedical field. The review of the technique used is briefly discussed.

In agriculture, image processing is widely used as a survey method in controlling the population of pests in the farm. Pest identification, segmentation, and automated counting are often discovered and developed by many researchers in algorithm evolution. A study has implemented a detection and count system of pests population at a paddy field in India [26]. This study has used the region growing method in order to provide the clear edge and good segmentation of images. The region growing technique is a technique which classifies the same

property of connected pixel as one identified region. This technique has first converted the RGB colour model into the greyscale. Then, the horizontally and vertically coordinates of each insect in the image are determined and the matrix is noted. The matrix noted is extracted as the detected pest and the accuracy rate of this technique is 80%.

A quantitative survey is very important in agriculture. In China, automatic planthopper counting has been used in open paddy field based on image processing [27]. The system used by the author is a portable and mobile system which only uses camera and smartphone during the surveyed-on field. The automated count planthopper system has adopted 3-layer detection. Figure 1 shows the flowchart of the layer 3-layer detection.

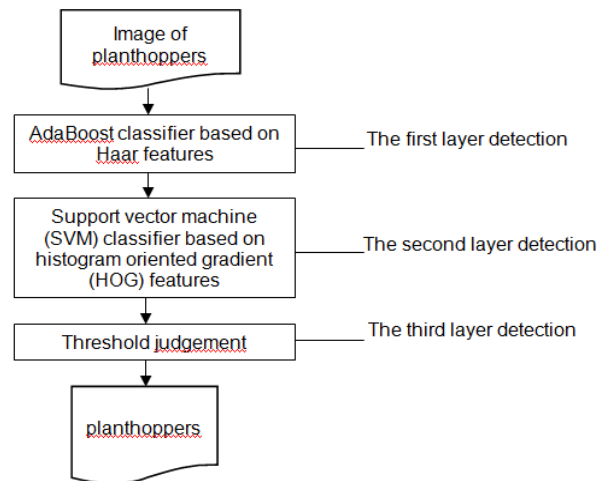


Figure 1. Flowchart of 3-layer detection for automated counting of white back planthoppers

The accuracy of this system of counting whiteback planthoppers is up to 85.2 % detection and 9.6% of false detection rate from 5800 images of trained data. The high accuracy of this algorithm is because of the population density of the planthoppers.

In another study, an adaptive algorithm has been developed based on hidden Markov model (HMM) in order to identify the images of the stored-grain insect [28]. The algorithm has improved the k-mean method which is used to initialize the HMM. k-mean is a classical technique for clustering in which its iterative method can minimize the quantization error by repeatedly moving all code vectors to their Voronoi sets of the arithmetic mean [29]. The Voronoi sets is defined as:

$$V_k = \{x_i \in D \mid k = \arg \min_{j=1, \dots, K} \|x_i - w_j\|\} \quad (6)$$

Where  $D = \{x_i\}_{i=1}^l$ ,  $W = \{w_k\}_{k=1}^K$ ,  $w_k \in \mathbb{R}^N$  and  $K \ll l$ . Code vector of  $w_k$  is set vector of  $D$  in which  $w_k$  is:

$$w_k = \frac{1}{|V_k|} \sum_{x_i \in V_k} x_i \quad (7)$$

where  $|V_k|$  is the cardinality of  $V_k$ . Thus, k-means can allocate quantization error by finding the local minimum. Improving the K-mean can increase the efficiency and stability of the algorithm. The algorithm that has been proposed is capable to segment and detect 98% of the normal image and 85% of lateral image of the stored-grained insect. The number of images that has been trained is about 100 pictures.

In Brazil, digital image processing has been used to quantify the nymph and the adult of whiteflies on soybean leaves [30-31]. Before threshold value can be obtained, the author has first analyzed the best colour model transformations which includes Cyan-Magenta-Yellow-Key (CMYK), CIE Lab, hue-saturation-value (HSV) and XYZ. Then the best colour model transformation is selected. CMYK has been selected and only channel C and Y colour are binarizing for further analysis. The best thresholding value that is selected to isolate the region of interest is between 150 and 100 in 8 bit. The algorithm is capable of quantifying the numbers of adult whiteflies within -1% of deviation from 748 images.

Thailand has become the world-leading shrimp exporter. Besides automated counting for the insect in field application, image processing method has been implemented in the counting of shrimp larvae by capturing the image above the tank [21]. This study has proposed the method of the co-occurrence colour of the histogram. The algorithm developed can be categorized into several distinct parts which include separation of larvae from the background, shape classification, and also the larvae count in the image. The larvae image can be classified to those connected to each other and not connected to each other. The algorithm cannot differ the false negative and true positive precisely because it is developed on the shrimp larvae count and not on the larvae position. From the statistics, the system has 97% precision on automated counting compared to manual counting.

Apart from shrimp, a biologist from Peru has acquired a sample of Peruvian Scallop in order to identify, count and establish its size by image processing through microscope [22]. The task was conducted in the lab in which the image of the scallop is captured within Sedgewick Rafter Cell slide in order to make it easier for thresholding process by establishing a region of interest. Likewise, a study has classified the image captured by two groups, those who are connected with each other and also to those which are not connected in way of border edges [21]. Thus, the algorithm counts the scallop as one when the scallop is attached to each other. Statistic in the spectral analysis by Fourier Transform of the histogram gives the main frequency of the signal. The number of scallops can be attained by counting the peak number of the FFT graph. The radius of the scallop can also be determined by assuming its circular shape.

Medical is one of the oldest fields that has been discovered by many researchers. A lot of application has involved image processing in the medical fields such as MRI, CT scan, x-ray, microbiology and many others. Although various fields exist in medical, only relevant applications that are significant to the task will be reviewed. It is observed that the medical application that fits the most is automated blood detection and count because it is almost identical to the *Aedes aegypti* shape. The enhanced threshold technique of Otsu's method has been used in segmenting the foreground and background of the blood cell [32, 33]. By converting the greyscale into a binary image, the author has successfully detected and counted the number of blood cell within 80% to 90% of overall accuracy.

In another study, a method of count colonies has been suggested in microscopic images using morphological operation [34]. Morphology operation involves geometric analysis which implements structuring element as an input and generating an output image of the same size. The approach of this operation is based on correlations between pixels instead of arithmetic correlations. The geometric features of the blood cell are extracted by choosing the best shape as a probe. Author has first performed the top-hat filtering to extract small elements before thresholding. From the review, the method has capabilities to identify more than 90% correctly from the cases. 8% overall deviation is accounted for.

In another study, an auto adjustable observation model has been proposed in which this method has combined different observation models in a particle filter framework [35]. This auto adjustable observation model has been tested with mosquito larvae which significantly move randomly and relate to the animal behaviour. By applying the k-means to cluster, the pixels in the merged blob. This method can track the mosquito larvae that overlap. Moreover, by marking the foreground of the segmented image and traitiong it to the closest centre using Euclidean distance, the processing cost can be lessened. This method provides a better and correct classification by achieving 85% of mean classification rate using 500 particles. Summary of technique used in various applications is shown in Table 1.

Table 1. Summary of technique used in various applications

| No. | Author             | Technique   | Result   | Description  |
|-----|--------------------|---|--|--|
| 1   | Martin [26]        | Region growing  | 80% accuracy of pest detection   | RGB is transformed into greyscale first and the test is conducted in lab   |
| 2   | Yao [27]           | 3 layers of detection include AdaBoost Classifier, SVM and Thresholding | 85.2 % detection and 9.6% of false detection rate from 5800 images                                       | The test is conducted at open paddy field  |
| 3   | Lu [28]            | Improve K-means via HMM   | Detect 98% of the normal image and 85% of lateral image of stored-grained insect from 100 images.        | The test is conducted in the lab.  |
| 4   | Barbedo [30], [31] | Thresholding  | Quantify the numbers of adult whiteflies within -1% of deviation from 748 images                         | It has been tested with CMYK, CIELab, HSV and XYZ transformation before CMYK is selected to proceed for thresholding. The test is conducted at open soybean farm |
| 5   | Wongnaret [21]     | Co-occurrence colour of histogram                                       | 97% precision on automated counting compared to manual counting.   | RGB is transformed into greyscale first and the test is conducted in lab   |
| 6   | Andres [22]        | Thresholding  | The scallop quantifies by counting the peak number of the FFT graph. The numbers of images tested is 20. | RGB is transformed into greyscale first and the test is conducted in lab   |
| 7   | Kanti Das [32]     | Thresholding  | The edge of the blood cell is 85% accurately detected from 20 images                                     | RGB is transformed into greyscale first and the test is conducted in lab   |
| 8   | Mohamed [33]       | Thresholding  | The nucleus of red blood cell is 80.6% accurately segmented from 365 images                              | RGB is transformed into greyscale first and the test is conducted in lab   |
| 9   | Barbedo [34]       | Morphology operation  | Identify more than 90% correctly from the cases and the overall deviation is 8%                          | After greyscale transformation, it performs the top-hat filter before thresholding   |
| 10  | Pistori [35]       | Auto adjustable observation   | 85% of mean classification rate using 500 particles.   |  |

### 3. Discussion

From the review, various techniques have been discovered that are suitable in identify and counting the number of *Aedes aegypti* which includes image pre-processing and image processing. Pre-processing is an important step that needs to be performed before segmenting an image. In this review, the best pre-processing techniques which are suitable to be suggested in image dehazing using DCP technique that has been proposed by the [15-17] and also Laplacian with median filter that is proposed by [21, 22]. The significance of suggesting this method is because experimental constraint; conducting in nominals dark in a room. The attenuation of the red colour channel causes the degradation of image quality because this experiment is carried out in a dark room. Thus, the DCP technique will restore and enhance the contrast of images. Meanwhile, the Laplacian and median filter can reduce noise and preserve the image information so that the contour of the edge can be clearly seen when it is segmented.

Many studies have been conducted in segmentation method, where thresholding is one of the simplest methods of segmentation that converts a greyscale image into a binary number. It has been reviewed the best technique to be suggested is one proposed by [30] and [31] for it has been tested by the authors with several methods of greyscale transformation before the thresholding process. The greyscale transformations that have been tested include CMYK, CIELab, HSV and XYZ colour model. From the analysis, it has been found that CMYK colour model has produces the best thresholding accuracy. Besides, the algorithm has tested at the open soybean field which portrays the stableness and robustness. Figure 2 shows the structured of the pre-processing and processing technique in identify and counting *Aedes Aegypti* larvae.

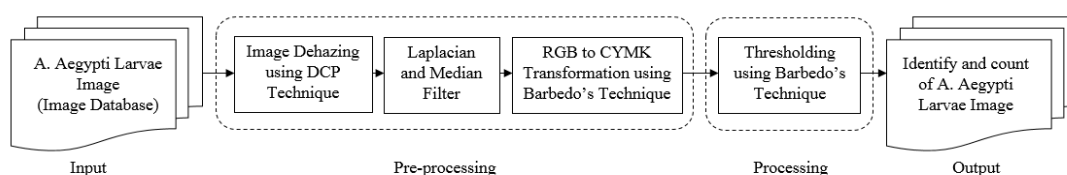


Figure 2. Structure of the pre-processing and processing technique suggested

The technique in identifying and counting *Aedes Aegypti* larvae is suggested literally. However, future work is recommended in order to cooperate with any pivots that may occur during the experiment.

#### 4. Conclusion

A review of different methodologies that have been used by various researchers in identifying and counting *Aedes Aegypti* has been conducted. The best technique of image processing is a technique which has high efficiency with less time of processing. Pre-processing is an imperative step that is required in any application that involves image processing. An important thing that needs to be highlighted is the best method to be suggested in identifying and counting of *Aedes Aegypti* larvae. The success in identifying and counting of *Aedes Aegypti* larvae relies mostly on the algorithm and technique selected. Although the best technique has been suggested, it is unethical to criticize other techniques because the technique has been suggested based on the application inquiries. Nevertheless, other techniques also have their own drawbacks.

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