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Automatic Detection of Spherical Virus Particle – Adenovirus and Polyomavirus

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***Abstract*—The paper describes the complete automatic detection of virus particles from transmission electron microscopy images. The approach locates two type of virus – Adenovirus and Polyomavirus. The initial detection takes place through segmentation and morphological filtering. In the second stage Gabor wavelet filtering approach is performed for particle selection in images. Decision Tree Classifier is performed in the final stage to classify the particles using various features extracted from the candidate point. The approach can locate intact particles, many damaged capsids and acceptable percentage of overlapped ones.**

Keywords—Transmission Electron Microscopy, Adenovirus, Polyomavirus, morphological filtering, gabor wavelet, decission tree classifier

I. INTRODUCTION

Manual detection of virus particles from Transmission electron microscopy images (TEM) is very time consuming because of the large number of particle views. The automatic selection becomes a topic of interest for many years. Automated methods exist but still they are restricted to specific sample condition ie. Spherical particles, high contrast and low noise. Even though current algorithms for automated selection locates the candidate point , but they are not effective upto users expectation. Most of the algorithms either select the candidate point interactively by the users from the entire image or automatic procedure edited by hand. Interactive methods solve the problem very effectively, but still a fully automated detection algorithm with efficiency greater than 75 % is the main goal.

Recently published work for automatic detection of virus particle (Maria C. Proenca et al., 2013) locates the intact candidate point with 100 % accuracy and there are some missed particles due to low contrast in the image. The approach used is based on the characteristics that entropy is low for virus particles compared to the unorganized background areas [1]. Four intensity profiles and 7 features from the profile is extracted from each candidate point and is compared with a set of test images obtained in the same condition.

The variety of clinical specimens were diagnosed in recent research(Goldsmith and Miler., 2009). They analyzed the importance of EM in virus identification. Several methods for treatment of vaccination against the virus attack are investigated. The template matching technique [3] uses a matched spatial filter which maximizes the SNR at the peak location and then a template matching model is used. Final validation is based on the correlation between the set of cross-correlation values and k^{th} correlation profile.

The local average intensity method [4] uses a ring filter to find out the peaks and several methods are carried to differentiate the actual particle centers from other peaks. In the cross point method [4] pixels of particular radius is compared with background pixels. If they are closer then that point is rejected.

This paper presents a fully automatic detection of both adenovirus and polyomavirs in TEM images prepared by negative staining methods. Negative staining is used to identify the very minute particles like viruses in the microscopy image. Virus vary from simple helical and icosahedral shape to very complex structure. The described algorithm can be extended for use of detection of any type of spherical virus.

Adenovirus (members of the family Adenoviridae) are medium-sized (90–100 nm), nonenveloped (without an outer lipid bilayer) viruses with an icosahedra nucleocapsid containing a double stranded DNA genome. Adenoviruses are unusually stable to chemical or physical agents and adverse pH conditions, allowing for prolonged survival outside of the body and water. Adenoviruses are spread primarily via respiratory droplets, however they can also be spread by fecal routes. Humans infected with adenoviruses display a wide range of responses, from no symptoms at all to the severe infections. Good hygiene, including hand washing, is still the best way to avoid picking up the adenovirus from an infected person. Most infections with adenovirus result in infections of the upper respiratory tract. Adenovirus infections often show up as conjunctivitis, tonsilitis (which may look exactly like strep throat and cannot be distinguished from strep except by throat culture), an ear infection, or croup. It can can also cause gastroenteritis.

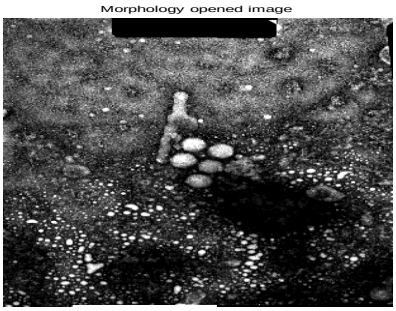
Polyomavirus are DNA based virus with 40-50 nanometer diameter. They are icosahedral in shape and do not have a lipoprotein envelope. The infection are normally acquired in childhood . Some of the symptoms are kidney problems , respiratory infection, fever, increased risk of tumors etc. It establish lifelong latency under normal cellular humoral immune surveillance.

II. ALGORITHM

The algorithm presented here has three main steps- detection of candidate point through segmentation, evaluation of candidate point through Gabor filtering, final validation of accepted candidates through decision tree classifier.The implementation was carried out in MATLAB 2009 version with a mix of proprietary function and originally developed routines in Matlab high language.

A. For detection of Adenovirus

The original negatively stained TEM image are of huge size. It should be resampled in ratio 1:3 to reduce the computation time during development. The images may contain a black label for the reference number. It is masked and replaced by the average intensity of the image. The first step in preprocessing is background irregularities compensation. It is achieved by top-hat operation i.e. morphological opening through a disk structuring element of radius 40 pixels and followed by subtraction of opened image from original image. The top-hat operation extracts small elements and details from the given image. The next step is the contrast adjustment-1% of data was saturated at high and low intensities and histogram equalization enhances the contrast through equal distributions of intensities. Histogram equalization allows low contrast areas to gain high contrast. The image is then preprocessed with wavelet decomposition followed by reconstruction with first levels of details suppressed. This preprocessed image is kept as the reference which is used in the second stage of the algorithm.



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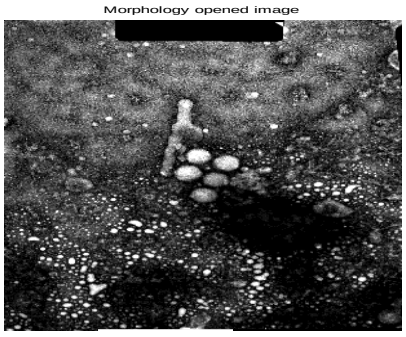
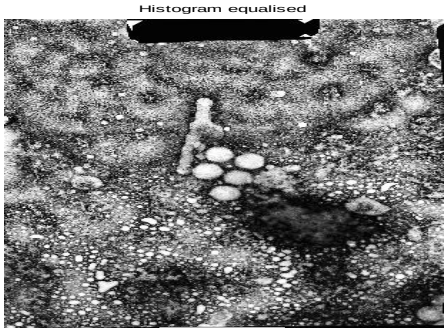
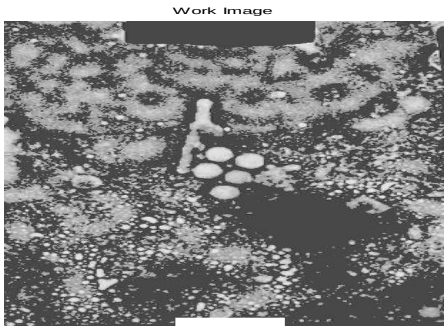


(b)

Figure 1. (a) Original Image (b) Top-Hat Filtered Image



(a)



(b)

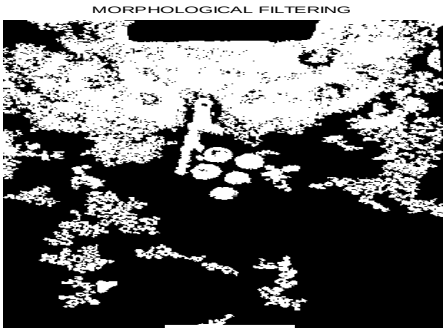
Figure 2. (a) Histogram equalized image (b) Wavelet Filtered Image

The wavelet filtered image is hereon referred to as Work image. This work image is then used for entropy filtering. The local entropy of the gray scale image is calculated and is replaced by the central pixel and the neighborhood pixels. The entropy gives statistical measure of randomness. Then the image contrast enhancement technique is performed. The image is enhanced using neighborhood pixel values via equation (1).

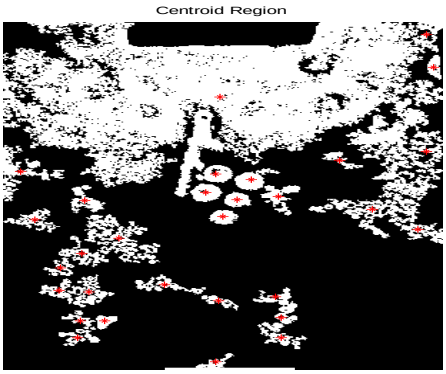
$$U_N(x,y) = \frac{U_0(x,y) - M1}{M2 - M1} \times Mg \quad \text{------(1)}$$

where U0 is the entropy filtered image , M1 is the minima and M2 is the maxima of U0. Mg is maximum gray level of the image. U_N is the contrast enhanced image.

The contrast enhanced image is then segmented based on a low threshold value . The threshold must target a low number of false negatives at the expense of high number of false positives. Morphological filtering based on area is carried out to isolate the smaller areas as these constitute the noises. Then the centroid of each cluster is obtained. The co-ordinates of each centroid is actually the co-ordinates of the candidate point.



(a)



(b)

Figure 3. (a) Morphological Filtering based on area
(b) Centroid obtained over each object.

In the second phase, the work image which was kept as reference is taken and again wavelet filtering is performed. The image is decomposed and then reconstructed with 2 levels of details suppressed followed by contrast limited adaptive histogram equalization(CLAHE) to exponential distribution. Then a pixel wise adaptive wiener filtering is carried out which is a low pass filter and removes noise followed by standard deviation filtering with a disk structuring element.

The strongly filtered image is then used for Gabor filtering . Gabor filter are band pass filters and is defined as the product of Gaussian kernel and a complex sinusoidal.

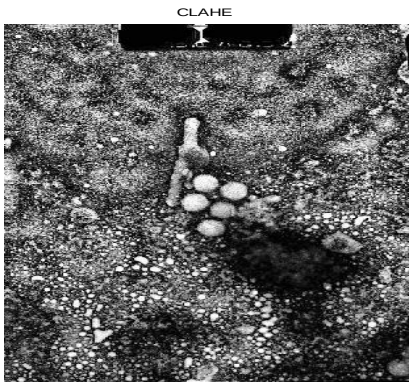


Figure 4. Contrast Limited Adaptive Histogram Equalized Image

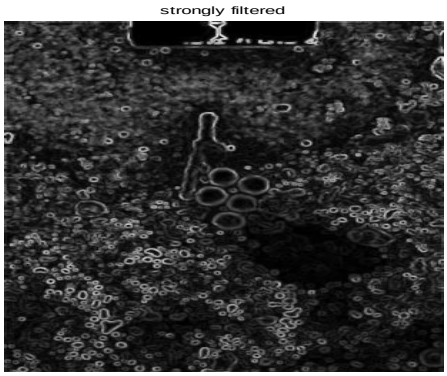


Figure 5. Standard Deviation Filtered Image

The centroid which was obtained in the first phase is overlaid onto the gabor filtered image. For each centroid certain features (Area, Eccentricity, Perimeter, Circularity, Length/ Width ratio) are extracted. The features are extracted in a circular region around the candidate point.

The features extracted are given as input to the Decision Tree Classifier. It is a popular tool for classification and prediction. DT classify based on rules. It classify starting from the root and moving through it until a leaf node. Each leaf node split into two child nodes. To start evaluating, database consist of some feature vectors are loaded. Tree building begins at the root node that consists of several features extracted from each candidate point. The algorithm determines the best possible variable to split the node into two child nodes

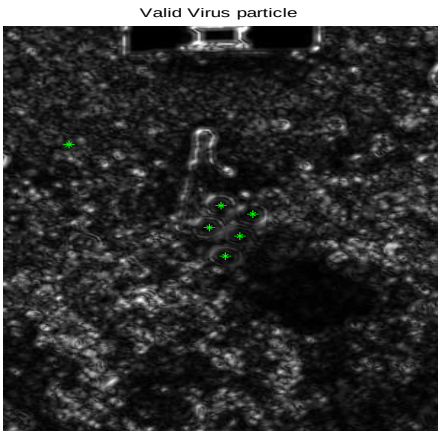
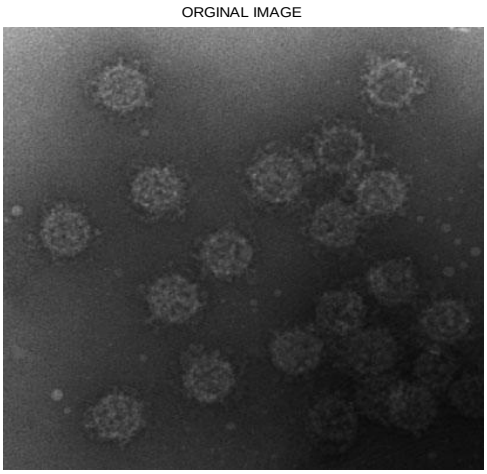


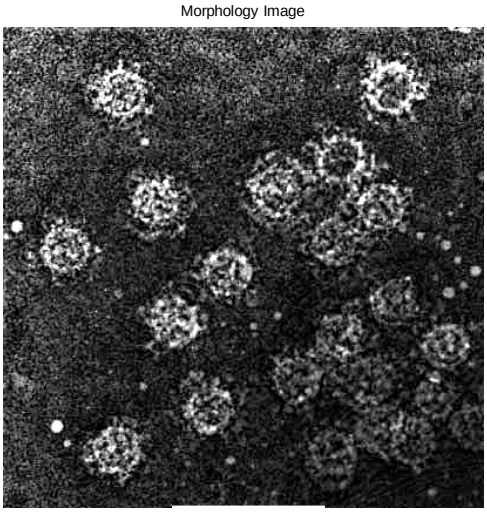
Figure 6. Valid Adenovirus Particle

B. Detection of Polyomavirus

The original TEM image is resampled in order to reduce the computational time during development. The image may contain a black reference number which should be masked and replaced by the average intensity value of the image. Same steps in the above algorithm are followed here till the centroid calculation.



(a)



(b)

Figure 7: (a) Orginal Image (b) Background Irregularities Compensated Image

To reduce the number of false positives , gray level co-occurrence matrix (GLCM) is used. GLCM analyzes pairs of horizontally adjacent pixels. It is a matrix with number of rows and columns equal to number of gray levels in image. It gives the second order statistical texture features. It provides the number of occurrence of gray level at specified angle (0,45,90,135) at a specified distance. Three measures are extracted from GLCM – energy, homogeneity, contrast. The intersection of three criteria that localizes the candidate point are low homogeneity, low energy and high contrast.

The candidate points are then evaluated on the basis of variance and mean intensity. Consider a circular region around each candidate point and calculate mean and variance of that region. Final validation is performed by considering the value of Gray Index. Gray Index is the ratio of variance to mean square at each final candidate. Based on threshold set on the gray index finally filters the candidate points.

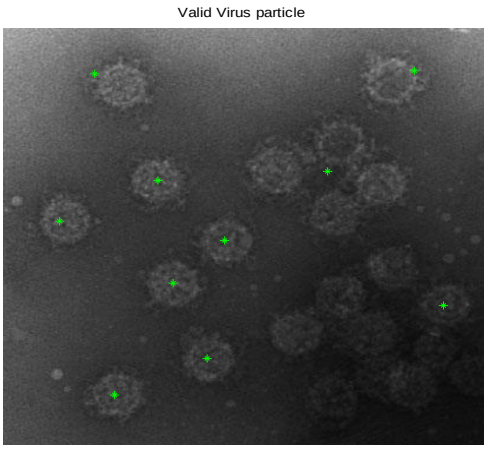


Figure 8. Valid Polyomavirus

III. PERFORMANCE EVALUATION

The algorithm detect 100% of perfect intact particles. The adenovirus detection algorithm can detect even permeated particles with acceptable percentage. Missed particles are the cases where the particle borders are on the low contrast region.

TABLE I
Algorithm Performance- Adenovirus particle

Number of Virus	Identified by algorithm	False Negatives
5	4(80%)	1(20%)

TABLE II
Algorithm Performance- Polyomavirus particle

Number of Virus	Identified by Algorithm	False Negatives
11	9(82%)	2(18%)

REFERENCES

[1] Maria da Conceicao M. Sangreman Proenca, J.F. Moura Nunes, A. P. Alves de Matos,"Automatic Virus Particle Selection- The Entropy Aprroach," *IEEE Trans. Image Processing*., vol.22, pp. 1996-2002 2013

[2] Maria da Conceicao M. Sangreman Proenca, J.F. Moura Nunes, A. P. Alves de Matos" Texture Indicators for Segmentation of Polyomaviru Particles in TEM Images"Microsc. Microanal., pp. 1-13 2013.

[3] Zhong Huang and Pawel A,PencZek, "Application of template matching technique to particle detection in electron micrographs," J.Struct.Biol.vol.145.nos.1-2.pp.168-180.2004

[4] T. Kivioja, J. Ravantti, A. Verkhovsky, E. Ukkonen, and D.Bamford,"Local average intensity-based method for identifying spherical particles in electron micrographs,"J.Struct.Biol., vol. 131,no,2,pp. 126-134,2000

[5] C. S. Goldsmith and S. E. Miller, "Modern uses of electron microscopy for detection of viruses," *Clin. Microbiol. Rev.*, vol. 22, no. 4, pp. 552–563, Oct. 2009.