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Colegio de Ciencias e Ingenierías

**Automatic Classification of *Wyeomyia* spp. and
Limatus durhamii species by Geometrical
Characterization in their wings.**

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DE TRABAJO DE TITULACIÓN**

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RESUMEN

En esta investigación se desarrolló un algoritmo implementando un proceso de análisis automatizado de aprendizaje que permite la detección y clasificación de los mosquitos en donde resalta las características relevantes en las alas de los mosquitos (morfología). El algoritmo posee: identificador de las especies, puntos de referencia, radios de las geometrías circulares. El objetivo es el de mejorar la forma manual en la cual los mosquitos son clasificados, así como de forma más didáctica implementar un software que permita interactuar con cualquier persona que desee saber más de estos mosquitos, en este caso de dos especies muy particulares como son *lydurhamil* y *wyeomya*.

Palabras clave: clasificador morfológico alas, mosquitos, *lydurhamil* y *wyeomya*.

ABSTRACT

In this paper a system for the automated classification of mosquitoes based on relevant characteristics on their wings morphology was developed. The algorithm developed allows to identify the mosquito's species by using reference points such as the radio of the circular geometries of the wing. The aim was to developed a system in order to improve the manual form in which mosquitoes are classified trough a didactic tool implemented automatically in software that allows interaction with anyone who wants to know more about these mosquitoes. For testing the system, two very particular species such as *Limatus durhamii* and *Wyeomyia* spp. were used for training and classification.

Key words: morphological classifier wings, mosquitoes, lydurhamil and wyeomya

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Automatic Classification of *Wyeomyia* spp. and *Limatus durhamii* species by Geometrical Characterization in their wings

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Abstract—In this paper a system for the automated classification of mosquitoes based on relevant characteristics on their wings morphology was developed. The algorithm developed allows to identify the mosquito’s species by using reference points such as the radio of the circular geometries of the wing. The aim was to developed a system in order to improve the manual form in which mosquitoes are classified trough a didactic tool implemented automatically in software that allows interaction with anyone who wants to know more about these mosquitoes. For testing the system, two very particular species such as *Limatus durhamii* and *Wyeomyia* spp were used for training and classification.

I. INTRODUCTION

Wyeomyia is a genus of mosquitoes, principally forest mosquitoes [1]. This specie inhabits small collections of water in bromeliads and aroids, flower bracts, bamboo stumps, tree holes, pitcher plants and occasionally artificial and other containers. Adults are active during the day. They are usually found in damp forests near larval habitats. [4]. Various species are found at all elevations in the forest canopy, but some seem to be restricted to ground level. Most of the species take blood meals and females readily feed on humans that enter their environment [1].

Limatus durhamii is another species of mosquitoes. [1] The adults of *Limatus* are unique in having a single unguis on the hind leg [1]. They generally resemble *Sabethes* in overall ornamentation, but the scutum is distinctive in bearing a striking pattern of gold, blue, and violet scales [6]. The short and broad slits of the occipital foramen and the absence of an apical tooth on the maxilla distinguishes the *Limatus* from those of *Sabethes* and the majority of species currently classified in *Wyeomyia* [6].

Researchers are seeking methods for automatically identify species of mosquitoes based on their wing’s vein patterns. Properly and fast identification of the mosquito species may allow to avoid and prevent diseases in humans. In such sense, geometry morphometric is the most common analysis method use to classify mosquitoes [3]

The actual process includes manual detection of vein pat-terns, among other morphological characteristics. This consists in identifying key patterns of the veins that may allow to recognize the mosquito [1]. The process should be repeated several times by the same user to gain consistency. Finally, the collected information must be then statistically processed for classification [10].

Therefore, these kind of geometric morphometric methods greatly depend on user’s experience and ability, therefore the obtained results may be subject to some level of subjectivity [4]. Thus, in this paper, we propose to implement and automated software system that would allow to recognize the differences between two species of mosquitoes as a first approach and that could be later easily complemented and modified to classify more species in the near future, according to certain parameters that can be identified in the wing images.

Morphometrical and morphological parameters such as intensity, shape, size, and position are very important to identify the wing pattern [7] [9]. Here we will present an automatic classifier based on digital image processing algorithms developed in previous works [8]. We have improved the image processing algorithm developed in Guerron’s previous work, using NI Labview and the IMAQ Vision Module, which focused on the wing patterns and spots. In this version, we detected the relevant information according to needs, and found important characteristics to classify the images of mosquitoes wings. We centered our focus on the characteristics of size and geometry since these are independent of the wing’s area and would allow for samples of the same species but with varied sizes.

II. IMAGE ACQUISITION

Mosquitoes samples were collected by Giovanni Ramon as part of a research project that is currently studying the biodiversity of mosquitoes in natural areas of Manabí province, in Ecuador.

First we separated all of the samples of the species that we were going to analyze. In this case, we first separated the



Fig. 1: *Limatus durhamii*. Photo credit: J. Stoffer, WRBU

Limatus durhamii and *Wyeomyia* from the mosquitoes. For both species we decided to only use the female samples, as first approximation since there may be differences between genders. We collected about 20 samples of each species, but it is important to notice that for future works, if we can get more samples, our supervised learning software will work faster and with fewer mistakes, since the model will be better constructed [8].

Once we separated the species of mosquitoes, we worked with their right wing specifically. We carefully removed the right wing with specialized equipment and put it in a slide. Hair and other residues were cleaned with a re-agent and then the sample was placed under a specialized microscope (Olympus) where the digital image was taken. This image was then used for the treatment of its subsequent classification. The preparation of the sample should be as careful as possible since any strong wind will modify the structure of the wings (possibly breaking the wings), as well as other external factors, such as the manipulation of equipment with too much force or excess reagent that would split the wing in analysis. It is important to remember that we will only work with the right wings of mosquitoes since their wings are not necessarily symmetrical, and to rule out any genetic modification, we will only use the right wings of the mosquitoes [11].

III. IMAGE IMPROVEMENT



Fig. 2: *Wyeomyia* spp. Photo credit: J. Stoffer, WRBU

Automatic Classification of *Wyeomyia* spp. and *Limatus durhamii* species by Geometrical Characterization in their wings

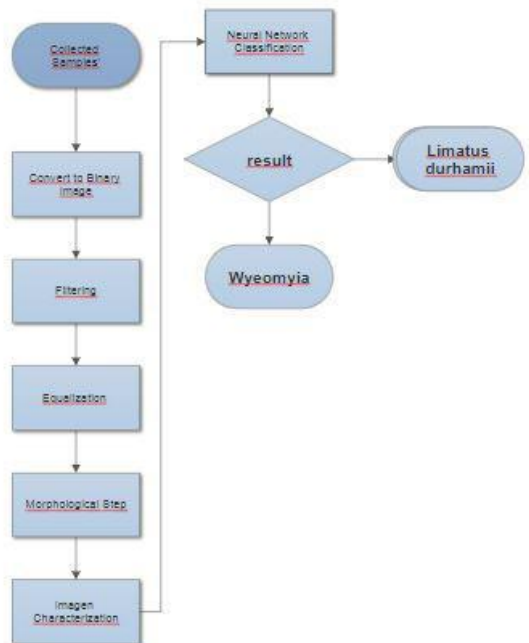


Fig. 3: Block Diagram of the algorithm.

After the original image was acquired, the Luminance plane was separated from the Hue and Saturation planes, as shown in Fig. 4. The result was an 8-bit gray-scale image in a range from 0 to 255 [19].

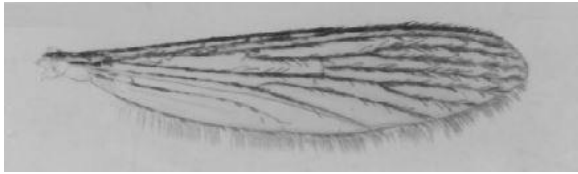


Fig. 4: Gray-scale Image. The luminance plane from Hue and Saturation planes has been extracted. *Wyeomyia* spp.

After the image quality was improved, it was then converted to a binary image. Next a spatial filter was applied to reduce the noise. The filter used was a low-pass filter that assigns the median value of its neighborhood to every pixel [10]. This filter does not take the isolated pixels, but it does not blur the edges of the objects [10].

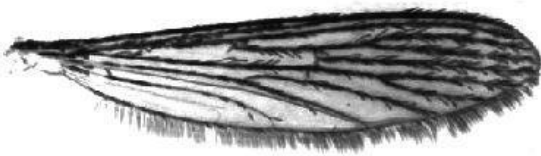


Fig. 5: Filtered Image. Spatial filter has been applied to the image of Fig. 4 in order to smooth the transitions and reduce the noise.

The binary conversion is a process in which all of the pixel intensities were changed to a one or zero depending on an estimated threshold value. To determine this threshold value, a clustering algorithm that looked for bright objects in the filtered image was applied [12]. Fig [6]



Fig. 6: Binary Image. A threshold value has been calculated using a clustering algorithm that looks for bright objects. Edges has not effectively identified

However, some information was lost when delimiting the pixels for wings characterization. To solve this problem, a “look up table” was used to spotlight characteristics in some areas, which we had lost, but may negatively affect others areas. Equalization is a special case in which each pixel is replaced by the product of its cumulative distribution value and the maximum intensity value in the image. An equalization procedure alters the image’s entropy [[14] so that the intensity values of the conforming pixels become distributed from 0 to 255 [[15]

Image equalization and thresholding were then used to recover most of the particles near the wing’s contour. It was also necessary to apply a group of morphological operations to facilitate the detection of relevant particles. [19].



Fig. 7: Equalized Image. The contrast has been increased due a differentiation of the bright and dark areas.

Touching particles were separated through an erosion and reconstruction process that eliminated the existing isthmus and separated the touching particles. Then, erosion was used to eliminate small particles that were too large to be left behind during the denoising process. The use of look up tables can also have some negative consequences. [19]. Some information was lost in comparison to the procedure in which the same morphological operations were applied without any equalization. [19]. Since all the intensities in the binary image

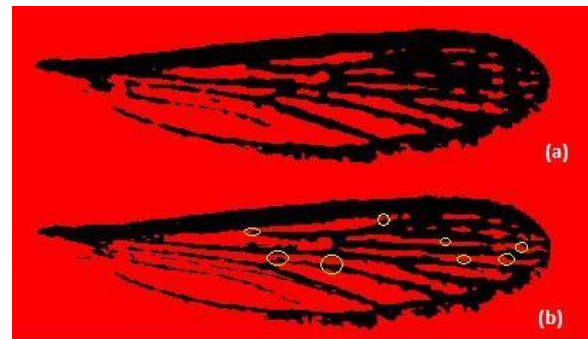


Fig. 8: Equalization and Thresholding. Thresholding preceded by an equalization. Separation procedure, border rejection and particle removal operations applied.(a) A size 5 separation procedure (b) thresholding by no-equalization, added size-7 separation procedure and removal small particles operations applied. We gain information highlighted by yellow circles on the image, as it can be seen the separation between particles has been increased.

are either 1 or 0, a logical “OR” operator was used to merge images together in order to deal with this drawback. [19]. However, when applied repeatedly or with a big structuring element kernel, this operation can erroneously divide concave particles in several parts. Therefore, an operation was also applied to the particles to compensate for this possible mistake. The opened particle put together previously divided particles, but it can make some holes appear or even show the vein forms or particles where the hair is produced. As a final morphological step, the holes within the particle are closed and a filter was applied to eliminate small particles that could have survived up to this point. [19].

We considered size and shape characteristics instead of simply counting the detected particles and vein unions, Fig 10 and Fig 12 show the results obtained.

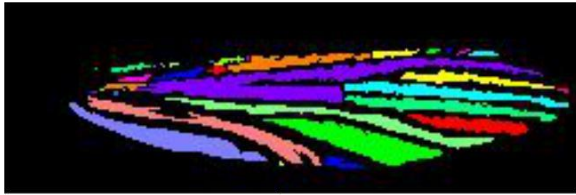


Fig. 9: Labelled Particles. Fig. 7 and Fig. 6 were combined by merging them, Hole and small particle were eliminated in the resulting image.

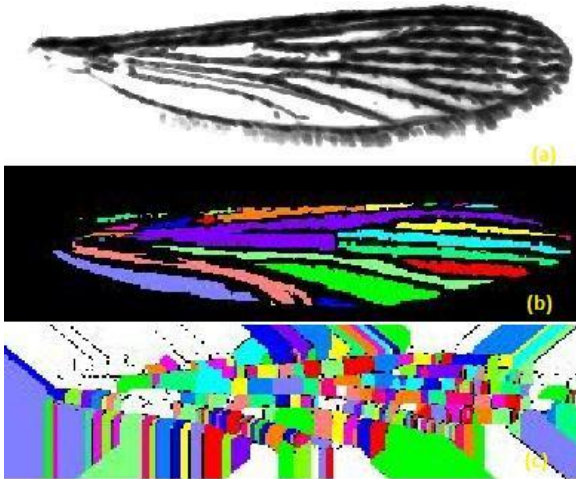


Fig. 10: Structure of the right wing of the Wyeomyia genus.(a) Source (b)Labelled particles (c) Wastershed Zones



Fig. 11: Wing example of Limatus durhamii

IV. IMAGE CHARACTERIZATION

After the particles have been identified in the image, the geometrical analysis begins. Ten parameters were extracted from the resulting images to describe two principal attributes: shape and size. [10]. The area of the particles was not considered since there may be differences in size amongst samples of the specie. Instead, the area was used as a normalizing factor for the size parameters. To analyze shape, the mean and the standard deviation amongst the particles was used. [19].

We focused on four parameters; two of them are considered as part of shape group, and the other two are considered as part of size group.

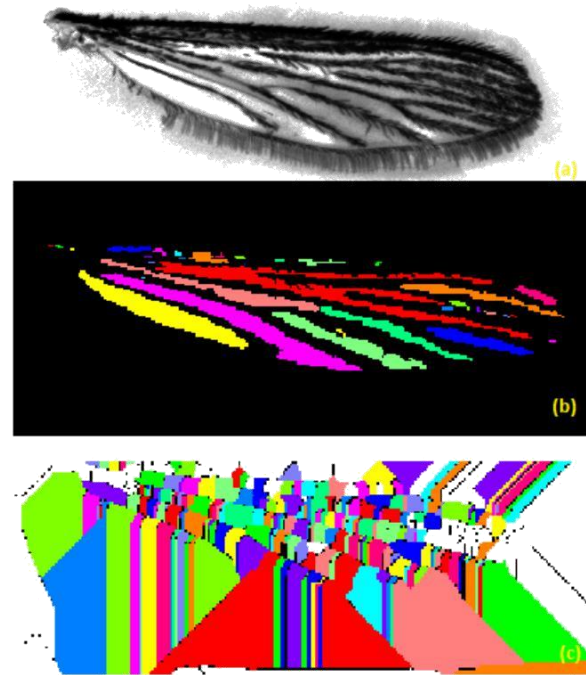


Fig. 12: Structure of the right wing of Limatus durhamii.(a) Source (b)Labelled particles (c) Wastershed Zones

The first one was the number of particles identified. Then, we have number of zones. Number of zone is the distance map which uses Danielson algorithm [17]. It consists in assigning a value to each pixel according to the distance to the border using a Watershed Transform [16]. It will show a number of non-overlapping zones present in the image. Then we focused on the next two shape parameters: elongation and Heywood Circularity Factor. Elongation Factor is the ratio of the biggest distance found in a particle Convex Hull in any direction intercepting the average value of the distances perpendicular with the particle. Finally, we have Heywood Circularity Factor which is a measure of how circular is the particle. [19].

We focused on this because, as we will see in our train neural network, these parameters will be the most important for classifying these two species of mosquitoes. Also, we have other parameters that may help in future research of other mosquitoes species such as: Centroid size is the distance between the Centroid and the landmarks normalized. Compactness is the other parameter. It represents the proportion of the particle area in relation to the smallest rectangle that encompasses the particle. This value will be represented with 1. Hydraulic radius is the ratio of the particle area to its perimeter. Type factor is the relation of the area according to the moments of inertia of the image. Eclipse ratio is the ratio of the major axis to the minor axis of the ellipse with the same perimeter and area than the particle. Rectangle Ratio is same as we discussed below, but the measure here is the ratio of the long side to the short side of the rectangle [19].

V. NEURAL NETWORK CLASSIFICATION

The human visual system is one of the greatest complexities in the world. In each hemisphere of our brain, humans have primary visual context containing 140 million neurons, with billions of connections between them doing complex image processing [6].

When you try to solve the same problems in any computer system, it is harder than it seems because algorithmically, you quickly get lost. [6]

Neural networks approach these problems in a different way. The idea is to take as many examples as possible (known as training examples) and then develop a system which can learn from those training examples. In other words, the neural network uses the examples to automatically infer rules that recognize images or patterns.

Now we wrote a Matlab program to implement a neural network that learns to recognize the difference between *Limatus durhamii* and *Wyeomyia* spp. The program is no more than 400 lines long and uses no special neural network libraries. But this short program can recognize the images and classification of two species of mosquitoes. [6]

The point of these programs was only to write a computer program that identifies two species of mosquitoes, but along the way, we developed plans with the same principle to include more species with more photographs that will be easy for the program to classify. [6]

We used an artificial neuron called a perceptron. A perceptron takes several inputs and produces a single binary output.

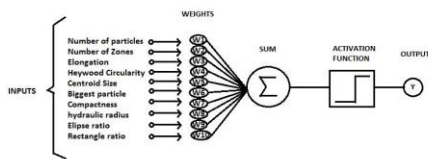


Fig. 13: Perceptrons: The Neural Networks using in our algorithm

In our case we analyzed four inputs, such as the number of particles, number of zones, elongation, and Heywood circularity. Then we introduced weights, real numbers expressing the importance of the respective inputs to the output, the neuron output, 0 or 1, is determined by whether the weighted sum is less than or greater than some threshold value In algebraic terms:

$$\text{output} = \begin{cases} 1 & \text{if } \sum_{i=1}^n W_i X_i \geq \text{threshold} \\ 0 & \text{if } \sum_{i=1}^n W_i X_i < \text{threshold} \end{cases} \quad (1)$$

where (w) is weights and (x) are the inputs.

Having defined neural networks, we used the parameters defined before. We focused on the second problem that works to choose the correct parameters. The idea and other variations can be used to solve the problem quite well. The idea is

that if the classifier is having trouble somewhere, then it is probably having trouble because the parameters have been chosen incorrectly.

A. Confusion Matrix

To use the Confusion Matrix first we need to separate our data in two parameters: - train data - test data Then we train our classification system using our train data and then we use our test data. This matrix helps us to measure the behavior of our next data .

Confusion matrix contains actual and predicted classifications that we obtained by our classification system. The following table shows the confusion matrix for a two class classifier.[20]

		ACTUAL	
		Class a	Class B
Predicted	Class A	a	b
	Class b	c	d

where:

- a is the the number of real positive cases in the data
- b is the number of incorrect predictions that an instance is negative,
- c is the number of incorrect of predictions that an instance positive
- d is the number of real negative cases in the data

Several standard terms have been defined for the 2 class matrix: The accuracy (AC) is the proportion of the total number of predictions that were correct. It is determined using the equation: [1]

$$AC = \frac{a + d}{a + b + c + d} \quad (2)$$

The recall or true positive rate (TP) is the proportion of positive cases that were correctly identified, as calculated using the equation: [2]

$$TP = \left[\frac{d}{c + d} \right] \quad (3)$$

The false positive rate (FP) is the proportion of negatives cases that were incorrectly classified as positive, as calculated using the equation: [3]

$$FP = \left[\frac{b}{a + b} \right] \quad (4)$$

The true negative rate (TN) is defined as the proportion of negatives cases that were classified correctly, as calculated using the equation: [4]

$$TN = \left[\frac{a}{a + b} \right] \quad (5)$$

The false negative rate (FN) is the proportion of positives cases that were incorrectly classified as negative, as calculated using the equation: [5]

$$FN = \left[\frac{c}{c + d} \right] \quad (6)$$

Finally, precision (P) is the proportion of the predicted positive cases that were correct, as calculated using the equation: [6]

$$P = \left[\frac{d}{b + d} \right] \quad (7)$$

The accuracy determined using equation 1 may not be an adequate performance measure when the number of negative cases is much greater than the number of positive cases. Other performance measures account for this by including TP in a product. G-mean¹ is the geometric mean of sensitivity and precision. [22] [7]

$$g \text{ mean}^1 = \sqrt[2]{\frac{TP}{P} \cdot P} \quad (8)$$

G-mean² is the geometric mean of sensitivity and specificity. [22]

$$g \text{ mean}^2 = \sqrt[2]{\frac{TP}{T} \cdot \frac{TN}{N}} \quad (9)$$

VI. RESULTS

In this section, we present the results obtained by the proposed algorithm and the classification process of these two species. Both the resulting images and the numerical values presented in Table I show the results obtained from the analysis of Fig. 10 and Fig.12 *Limatus durhamii* and *Wyeomyia* spp. that were taken as a previous example

TABLE I: Results obtained after processing a wing image of *Limatus durhamii*.

Parameter	Mean	Standard Deviation
Number of Particles	36	NA
Number of Zones	283	NA
Centroid Size	0.09	NA
Biggest Particle	0.20	NA
Elongation	6.30	5.14
Compactness	0.57	0.17
Heywood Circularity	1.78	1
Hydraulic Radius	6.88	6.48
Type Factor	0.76	0.21
Ellipse Ratio	7.97	12.01
Rectangle Ratio	10.66	18.83

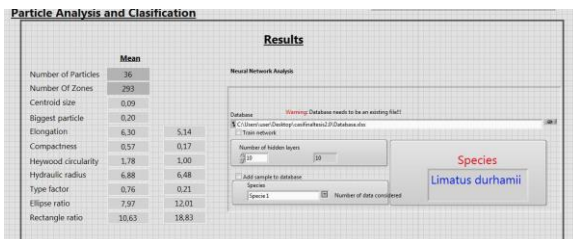


Fig. 14: data collected after the analysis of *Limatus durhamii* genus. The left side we have the values of all the parameters we discuss. The right side we have the data where our data are saved, and in blue letter the specie of the mosquitoes after the analysis

Fig. 14 and Table I show the results obtained after processing an example of a wing of *Limatus durhamii* genus. Fig. 15 and Table II, on the other hand, show the results for an example of processing a wing image from *Wyeomyia* spp. When comparing the results obtained in Tables I and II, it can

be seen that besides differences in the number of particles, the number of zones, elongation, and Heywood circularity are important differences in both size and parameters. There are also considerable differences (more than 10 percent) in the mean values of Elongation, Hydraulic Radius, Ellipse Ratio, Rectangle Ratio, and standard deviation of all seven shape parameters.

TABLE II: Results obtained after processing a wing image of *Wyeomyia* spp

Parameter	Mean	Standard Deviation
Number of Particles	25	NA
Number of Zones	580	NA
Centroid Size	0.07	NA
Biggest Particle	0.34	NA
Elongation	8.82	7.72
Compactness	0.43	0.29
Heywood Circularity	2.10	1.26
Hydraulic Radius	8.86	6.73
Type Factor	0.55	0.36
Ellipse Ratio	2.25	0.73
Rectangle Ratio	1.66	1.15

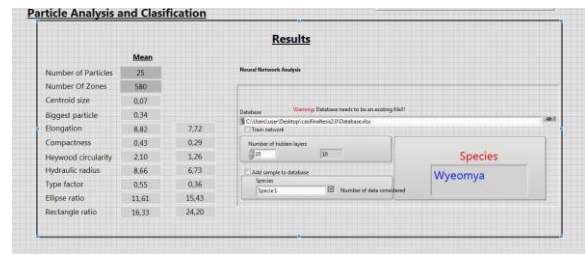


Fig. 15: Wing example of *Wyeomyia* spp. Same image as we could see in Fig 14, this time we have our example with *Wyeomyia* spp. In blue letter we see the specie of the our second sample

A. Confusion Matrix

- We used the Confusion Matrix with the next parameters.
- train data (35 %).
- test data (65 %),

Then we trained our classification system with the next results.

		ACTUAL CLASS	
		<i>Limatus durhamii</i>	<i>Wyeomyia</i>
P.C	<i>Limatus durhamii</i>	11	0
	<i>Wyeomyia</i>	1	10

$$AC = \left[\frac{21}{22} \right] \times 100 = 95.45\% \quad (10)$$

$$TP = \left[\frac{10}{11} \right] \times 100 = 91\% \quad (11)$$

$$FP = 0\% \quad (12)$$

$$TN = 100\% \quad (13)$$

$$F N = \left[\frac{1}{11} \right] \times 100 = 9\% \quad (14)$$

$$P = \left[\frac{10}{10} \right] \times 100 = 100\% \quad (15)$$

$$g \text{ mean}_1 = 95:4\% \quad (16)$$

$$g \text{ mean}_2 = 0:97 \times 100 = 97\% \quad (17)$$

if equations [17] and [16] are equal to 0, automatic software to classify is not correct. in Both cases we probed that our automatic software to classify is working correctly

VII. CONCLUSION

An algorithm for the automatic software to classify two species of mosquitoes based on particle analysis has been proposed in this paper using Labview as development tool. The analysis is based on obtaining ten parameters that describe shape and size characteristics, and we focused on four of them; as a result, an easy way of classified mosquitoes was created instead of doing it manually. Limitations are given by factors like the quality of the samples, the resolution of camera, or even the quantity of samples of any species.

The initial results obtained from the algorithm proposed are highly satisfactory and can be consider as one big step to classifying mosquitoes automatically.

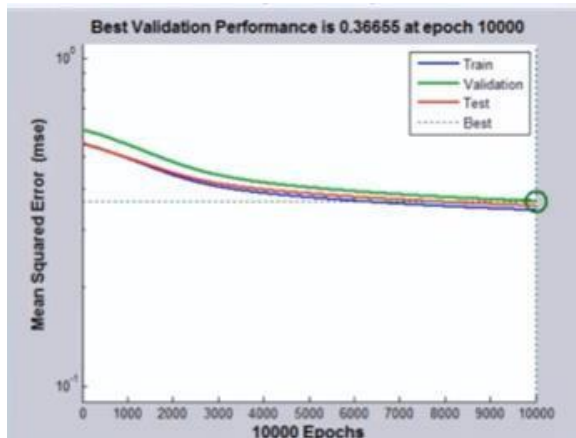


Fig. 16: Mean Squared Error of our program. In the graph we can see what modifies the performance function of the network (the error measure that minimizes the training process). By including the values of the weights and trends, the regularization produces a network that works well with the training data and offers a more fluid behavior when presented with new data.

VIII. DISCUSSION

As we have seen in the work done, and in the results; there are important differences between *Wyeomyia* spp. and *Limatus durhamii* genus according to the aforementioned parameters, and where the largest amount of data available in the laboratory was collected. We needed more samples for

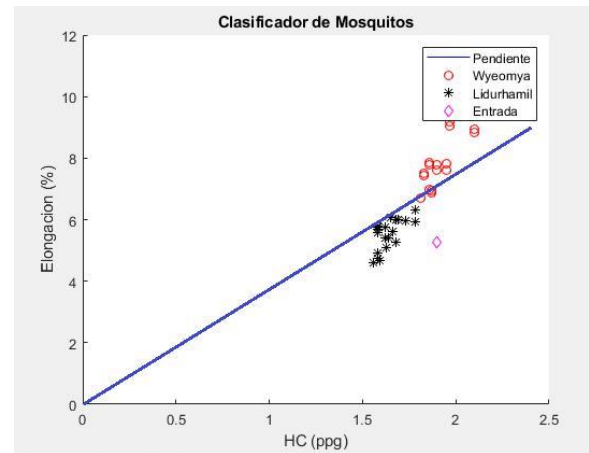


Fig. 17: We focus on our NNA. In the process we can see our data are grouped by similarity, in this case clustering we can see in the graphic the difference in HC vs Elongation and the difference in the characterization between this two species

a more optimal result, good results were established with the limitations when implementing the algorithm and subsequent classifier. One of the first parameters that we obtained was the division of samples between trained samples and the samples that we would verify that our training is carried out effectively. As we can see in the confusion matrix table, our proportion was 35 to 65, where the training samples should predominate. This will help us to train correctly according to our neural network program. Our accuracy (95.45%) indicates that we have a percentage of correct answers in our prediction, where if it exceeds 95% of efficiency it indicates that our classifier performs the desired work. Our optimal value should be 97.7% or more but according to our data samples, we won't have this class. We will need at least 100 samples more to have an optimal classify program verify. Our true positive also indicates that our proportion of positive cases were identified 91%. In other hand we have The true negatives that is 100%. If we see the balance between TP and TN are around 10% of difference. According to the theory, it said that this two data should be balance, otherwise our method is not according to what we are searching because it will be unbalance. In our case, our precision is almost perfect but this value indicate two things: the first one is that we need more samples to find the real value of our precision and according to that, we see if our system works. The second one is that if we keep as a perfect value of precision 100%. our program is not working correctly because we need an error value that indicates that our program is learning according to the samples. Not always the numbers that surpass the efficiency of 95% are the correct ones since it is only a parameter to measure if the data are sufficient. For this we take into account that the number of examples of each species must be proportional since otherwise we will obtain almost perfect values that nevertheless mean that when classifying a species they will get errors too high if our sample samples are not equal. When attempting to construct a classifier for a somewhat imbalanced data set, I

was led to the question of measuring the performance of the classifier. One of the first things I thought of was to take the average of precision and specificity. The result in our case is 95.45%. and it will call the geometric mean of sensitivity and precision .Then we have the of sensitivity and specificity with 97%.. This two values should not be more than 5%. because it measure if our program collect as same as positive and negative cases. In Fig. 16 we see the mean square error vs epoch (time), where we can see that our learning neural network takes around 0.37 seconds to learn with our training data, in that time our error is 10^{-3} . Finally in Fig. 17 we see the cluster distribution in two significant parameters that we have in our paper, Elongation vs Heywood Circularity , the distribution of our data are very clear and we see that we do not have ambiguities when we put our verify data.

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