

## Algorithm for an Automated *Clarias gariepinus* Fecundity Estimation Technique Using Spline Interpolation and Gaussian Quadrature

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**Abstract**— Fecundity is essential in the field of population ecology, where the number of eggs is measured to get the actual reproductive rate of an organism. The estimation of fecundity is essential for an accurate study of biology and population dynamics of fish species. This estimation can be developed using the gravimetric method (weight method) to calculate the number of eggs. However, this method still requires experienced technicians and much time and effort to compute the number of eggs manually. The increasing growth in both hardware and software have led to many improvements in imaging technology. Hence, this research addresses the problem of employing constructing a computer vision algorithm. This paper introduced the automatic fecundity estimation method, which applied simple mathematic theories and image processing algorithm to estimate the fecundity of African catfish (*Clarias gariepinus*). From the image of the fish, the fish's eye was be detected using a modified Haar Cascade Classifier Algorithm and appointed axis line where the eye becomes the origin point. Next, we identify the region of interest, which reflects the fish's fecundity to obtain the pixels corresponding to the silhouette of the region as coordinates in Euclidean space, which are then represented with a function using cubic interpolation function. Using this function, we compute the region of interest using an integral numerical approach, e.g., Gaussian Quadrature. From the result, we compared with the ground truth to get the estimation of the number of eggs.

**Keywords**— fecundity; eye detection; image processing; mathematical theories; ground truth.

### I. INTRODUCTION

The steadily growing importance of fish farming has compelled improvements in the technologies necessary for securing the initial and basic requirements for productive aquaculture, namely the production of fish seed for stocking. The most crucial aspect in the reproductive biology of fish that should be understood is the fecundity of fish where it is to describe the change of production level, to make efforts to increase the number of harvests, and also useful for make estimation of the total population. Fecundity is defined as the number of eggs produced per female fish per unit of time (for example, every spawning season) [1]

This research is focused on estimate the number of eggs of African catfish species called *Clarias gariepinus*. We chose this fish because it is a robust fish species in aquaculture and high demand. *Clarias gariepinus* is one of the most important farmed freshwater fish species [2]. *Clarias gariepinus* is mostly used for fish stocking due to its fast growth rate and ability to tolerate a wide range of temperatures, low dissolved oxygen, and salinity [3]. The fecundity rate for female catfish is often related to her size

throughout her reproductive lifespan. Several factors affect egg size quality, such as different genetics, type of food, and water temperature. *Clarias gariepinus* air-breathing catfish is a typical scaly, bony elongated body with long dorsal and anal fins. Dorsally have different color from dark to light brown and often mottled with shades of olive and grey and lower part is pale cream to white. This fish can growth very large where it can reach a maximum length of 170 cm and weight 60 kg.

Several methods have been introduced and used to estimate the fecundity of fish, which are the gravimetric method (weight method), volumetric method, auto-diametric method, and stereometric method [4]. However, this method is time-consuming. Hence, this research addresses the problem of employing constructing a computer vision algorithm. The following section describes the ways that we use in our study to estimate the number of eggs.

### II. MATERIALS AND METHOD

This section describes previous methods that have been introduced and used to estimate the fecundity of fish.

### A. Gravimetric Method

It is the most common method used to estimate the number of eggs because of a beneficial technique for batch fecundity estimation [4], and it is more accurate and inexpensive low technology approach [1], [5]. However, this method is time-consuming due requires the whole ovaries being returned to the laboratory—this method is based on the relation between ovary weight and oocyte density in the ovary. The mature fish's length, weight, and ovaries weight were taken for gonadosomatic index (GSI). Three subsamples were taken from the front, mid, and rear sections of each ovary and weighed. Then, one by one count these eggs of the whole slide.

$$\text{GSI} = (\text{weight of ovary} / \text{weight of fish}) \times 100$$

This value was proportional to the total ovary weight; the number of eggs (F1) for the subsample was estimated by using the following equation:

$$\text{Fecundity (F1)} = (\text{no. of eggs in sub-sample} \times \text{gonad weight}) / \text{weight of subsample}$$

Then, by taking the mean number of three sub-sample fecundities (F1, F2, and F3), the individual fecundity for each female fish was calculated using the formula below:

$$\text{Fecundity} = (\text{F1} + \text{F2} + \text{F3}) / 3$$

### B. Volumetric Method

The volumetric method uses the same principle as the gravimetric method but uses ovarian volume and subsample volume instead of ovary weight [4]. The volumetric method is necessary to estimate the mean value of the egg volume [6]. The mean volume of eggs was estimated after immersion in distilled water (immersed condition) and after removing the water utilizing a vacuum pump at 0.50 atm (humid condition). Egg loss by suction was prevented by employing a piece of phytoplankton mesh.

### C. Stereometric Method

The stereometric method is unique due it is subject to analyse histological images of the fish ovary to estimate the fecundity. A hexagonal grid is overlaid on the histological image and the number of grid points associated with each oocyte (reproductive cells) category. In each category, the number of oocytes is counted in this method [7]. By using off-the-shelf software, this process is done manually, but it is very time-consuming, requires specialized technicians, and does not allow to review the calculations.

### D. Automated Eggs Counting and Sizing from Scanned Images

An imaging-based technique was developed to count and measure oocytes from a gravimetric gonadal sub-sample [8]. Sub-samples were preserved in a non-toxic formulation of Gilson's solution, which offers an alternative to other preservatives commonly used in fecundity studies. The technique uses high-resolution optical scans of plated oocytes, imaging software, and user-defined object

classifications to separate oocyte from ancillary material likely to be present in a processed sample.

Thus, the first step was to set the threshold for an image. This is an important step because this setting defines the position of an object's edge, in this case, the outline of an egg. At this point, the counting routine would find all objects—eggs and debris—and would classify touching eggs as a single object, as shown in Figure 1. To reduce the number of touching eggs misclassified and counted as single objects, the image was first binarized, and an erosion-dilation filter utility in the imaging software was applied. They are effective in separating most touching eggs from a single cluster to the component individual eggs, as shown in Fig. 2.

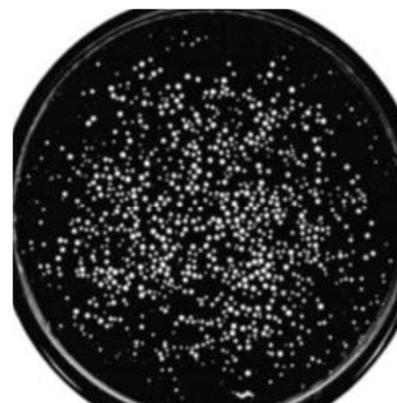


Fig. 1 Image of scanned sub-sample of American shad eggs suspended in agarose in a petri dish

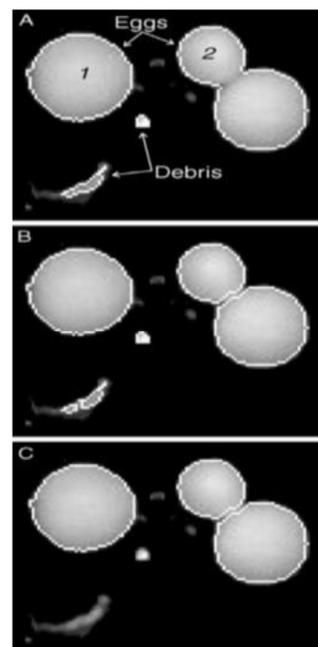


Fig. 2 Image of a portion of scanned gravimetric sub-sample. White perimeters identify objects automatically designated by the imaging software as foreground objects. (A) Single oocyte ('1'), touching oocytes ('2'), and debris. (B) Applying the erosion-dilation filter results in the separation of touching oocytes. (C) Using the egg-object-class classification results in debris sent to the image background.

### E. Estimating Fish Fecundity Based on Digital Analysis of Histological Images

The software Govocitos is evaluated to offer an easy and automatic way to estimate fecundity using the stereological

method [7]. Firstly, it uses a grid of points defined by the user; second, counting of points and objects inside the grid; and third, estimation of stereological parameters, partial areas/volumes, potential and partial (for each development stage) fecundity. Govocitos automatically recognizes and classifies oocytes based on the presence/ absence of nucleus (as shown in Fig. 3), as well as on the developmental stage; it calculates cell diameter, area, and roundness; it builds diameter frequency histograms.

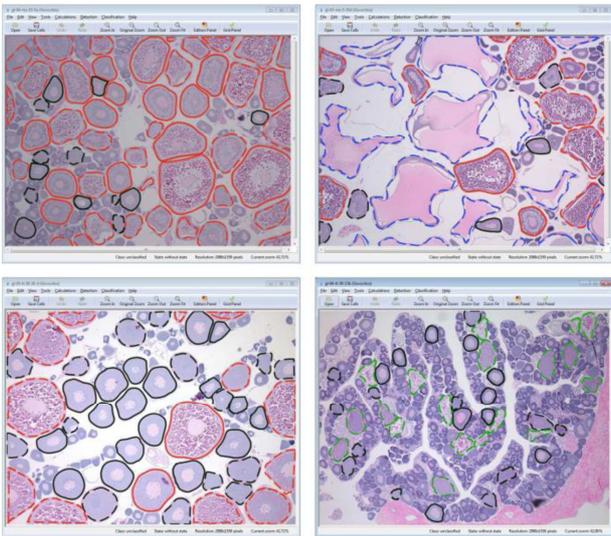


Fig. 3 Some typical histological images of fish ovaries from two species: European hake (upper panels) and pouting (lower panels). The true contours of mature oocytes are overlaid (the color and line type show the stage and class of each of them)

The recognition step is based on a multi-scalar Canny filter, which automatically detects the oocyte outline. It achieves an accuracy of 64% in an unsupervised way, which increases up to 80% when the expert marks only one point on the unrecognized oocytes using the GUI. The classification uses Support Vector Machines (SVM) combined with texture (Local Binary Patterns) and color features, achieving an accuracy of 84% to discriminate between oocytes with or without a nucleus and 87% to distinguish among three development stages (cortical alveoli, hydrated and vitelline/atretic). It can see in Fig. 4 below:

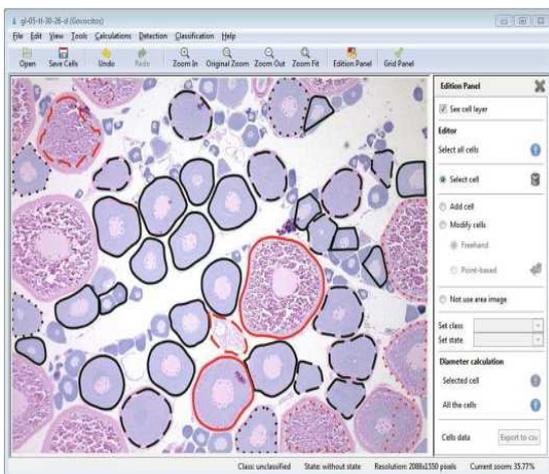


Fig. 4 The editing view after automatic recognition and classification steps

#### F. Automatic Counting of Aedes

Automatic counting of *Aedes aegypti* Eggs in Images of Ovitrap is the method for mosquito eggs counting, as shown in Fig. 5. To achieve more difference between the eggs and the trap, the images are converted from RGB to HSL color model (Hue, Saturation, and Lightness) [9], as shown in Fig. 6.



Fig. 5 Samples of mosquito eggs

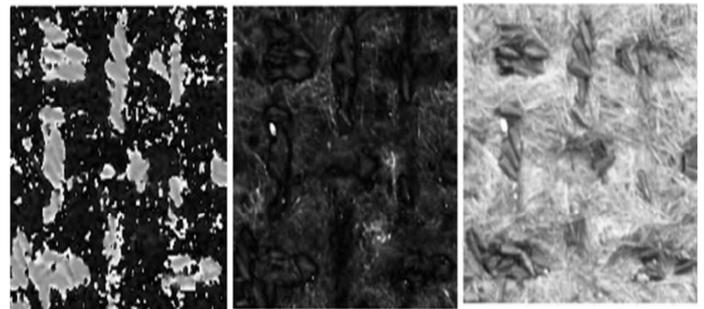


Fig. 6 Image with Hue, Saturation and Lightness Components

The hue image is then binarized using Huang thresholding algorithm, as shown in Fig. 7. With the bi-level image, a connected components algorithm is applied to the label at each connected white area of the image. Small white areas can be deleted as they could not contain an egg. The image is filtered using the morphological operation of closing to focus on a structural element in the form of an egg, as shown in Fig. 8. The number of eggs is the total amount of white pixels divided by the average area. From this research, they considered that an egg occupies an area of 170 pixels. So, the number of eggs is the total amount of white pixels divided by the average area. In this case, the method registered an amount of 33 eggs against the correct value of 34 eggs that the image contains.

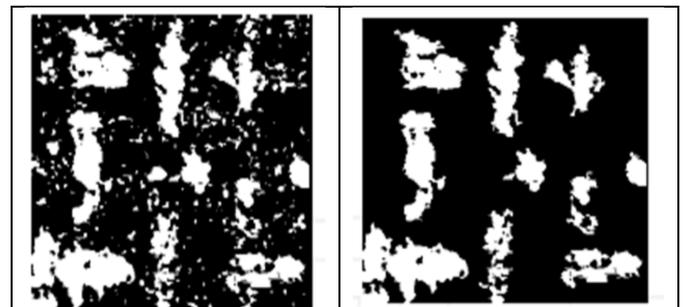


Fig. 7: Bi-level hue image after elimination of small connected areas and hue image after binarized by Huang's thresholding algorithm

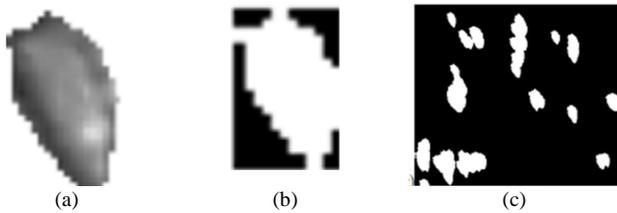


Fig. 8: (a) Average egg that was used to define, (b) The structural element, (c) Image after application of closing operator with the structural element

The comparison between the previous researches has been discussed in Table I below:

TABLE I  
COMPARISON BETWEEN EXISTING METHOD AND PREVIOUS RESEARCH

Method/Previous research	Main features
Gravimetric method	Used ovary weight.
Volumetric method	Use ovarian volume and the subsample volume.
Stereometric method	Analyze histological images of the fish ovary.
Automated eggs counting and sizing from scanned image	Count and measure oocytes from a gravimetric gonadal subsample. Using the method of erosion-dilation filter to separate the eggs in contact, distinguish eggs of other materials.
Estimating Fish Fecundity based on Digital Analysis of Histological Images	Automatic way to estimate fish fecundity based on the traditional stereological method. Automatically recognizes and classifies oocytes based on the presence/absence of the nucleus and the development stage; it calculates cell diameter, area, and roundness.
Automatic Counting of Aedes aegypti Eggs in Images of Ovitrap	Two methods were used, which converts the image from RGB to HSL and changes the image from RGB to YIQ. The image filtered using the morphological operation to focus on the structural elements in the form of eggs.

### III. RESULTS AND DISCUSSION

The main objective of the study is to calculate the area of fecundity and evaluate the eggs count values of that interest region. To do this, we maintain the fish image capture at 20cm from the base ground. After that, the overall process is accomplished in three phases in Fig. 9 below:

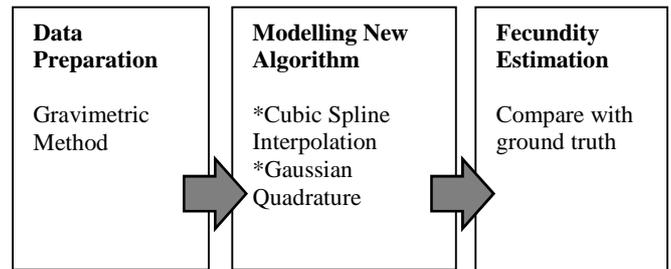


Fig. 9 Three phases involved in the process of fecundity estimation

#### A. Data Preparation

The proposed model requires data preparation and ground-truth development. This data preparation used a manual method, which is a gravimetric method to get the number of eggs. The technique has been discussed in section II. From that data, the Catfish Fecundity Database was created. Adult females of catfish were chosen alive and in good condition from the tanks. Then, the sample fishes were precisely measured by collecting all the body weight (g), body depth (cm), and body width (cm). The images collection process requires a person to capture the fish images in 3 positions where the distance between the camera and the catfish is fixed at 20cm, as shown in Figure 10 below. Although, before the process of measurement and capturing the images, the catfish went through the process of pitting.

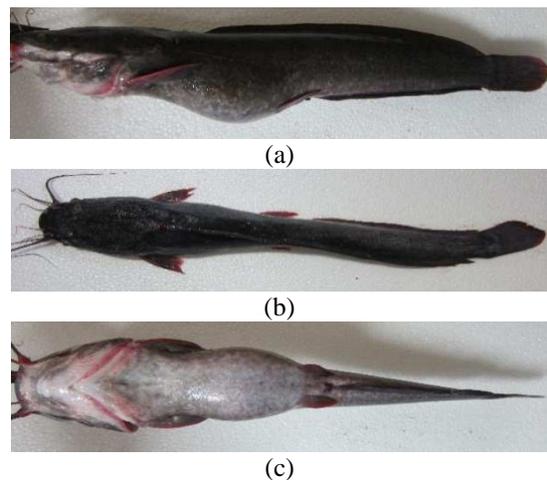


Fig. 10: Position image of fish (a) hanging position (b) top position (c) under position

Fig. 11 shows the process of data preparation using the gravimetric method. Once the training images and ground truth are ready, the properties of the algorithm can be identified

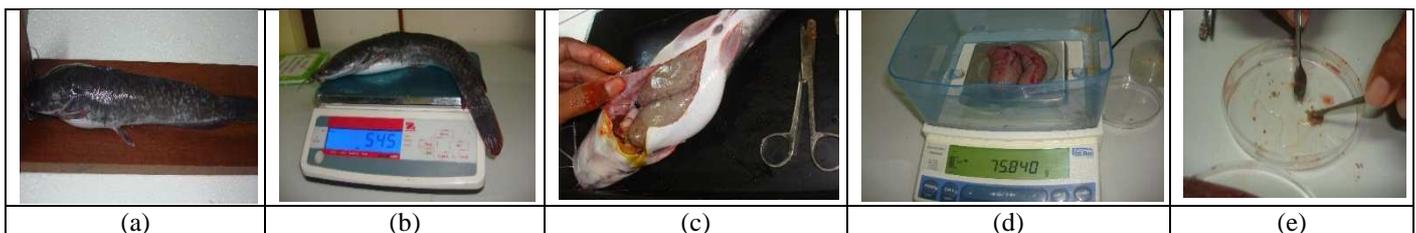


Fig. 11: Process data preparation using Gravimetric method (a) measure length of fish (b) measure the weight of fish (c) removing the ovary (d) the ovary weighting process (e) process of counting fish eggs

## B. Modeling New Method

The images were enhanced digitally to improve visibility to enable a clear view of the fish's eye. A new fecundity estimation was designed. We started the process by detecting the fish eye using a modified Haar Cascade Classifier Algorithm. The core basis for object detection is the Haar-like features [10], [11]. These features have used the change in contrast values between adjacent rectangular groups of pixels. To determine relative light and dark areas, the contrast variance between two pixels groups is used to identify. Each Haar feature has a value that is calculated by taking the area of each rectangle, multiplying each by their respective weights, and then summing the results. The area of each rectangle is easily found using the integral image. The integral image generation is illustrated in Fig. 12 below:

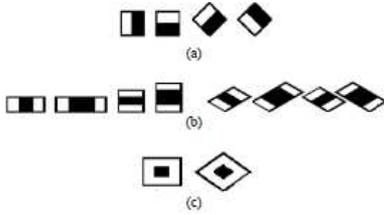


Fig. 12: Common Haar Classifier (a) Edge Feature (b) Line Features (c) Centre Surround Features

The value at any location  $(x, y)$  of the integral image is the sum of the image's pixels above and to the left of the location  $(x, y)$ . So, if  $A[x, y]$  is the original image and  $AI[x, y]$  is the integral image, then the integral image is computed as shown in equation 1 and illustrated in Fig. 13.

$$AI[x, y] = \sum_{x' \leq x, y' \leq y} A(x', y') \quad (1)$$

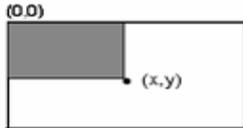


Fig. 13 Summed area of the integral image

The features rotated by forty-five degrees, like the line feature shown in Fig. 12 (b), require another intermediate representation called the rotated integral image or rotated sum auxiliary image [12]. The rotated integral image is calculated by finding the sum of the pixels' intensity values that are located at a forty-five-degree angle to the left and above for the  $x$  value and below for the  $y$  value.

So, if  $A[x, y]$  is the original image and  $AR[x, y]$  is the rotated integral image, then the integral image is computed as shown in equation 2 and illustrated in Fig. 14.

$$AI[x, y] = \sum_{x' \leq x, x' \leq x - |y - y'|} A(x', y') \quad (2)$$

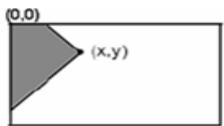


Fig. 14 Summed area of the rotated integral image

By picking up the eye as the focal point, we generated the axis line where the fish's eye was set as the origin point  $(x_0, y_0)$  as illustrated in Fig. 15 below:



Fig. 15 Axis line; fish's eye as a point of origin  $(x=0, y=0)$

Then, we identified the start and ending point (yellow line)  $(x_1, x_n)$  of the interesting region where it started from pectoral fin to anal fin (as shown in Fig. 16).

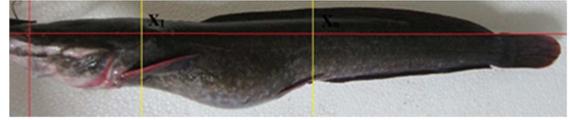


Fig. 16 Start and endpoint (yellow line) of the interesting region

All the potentially related points are detected. At this stage, the selected points produced the stable function  $f(x)$  using Cubic Spline Interpolation. The most common piecewise-polynomial approximation uses cubic polynomials between each successive pair of nodes called Cubic Spline Interpolation. It is a particular case for spline interpolation that is used very often to avoid the problem of Runge's phenomenon. It gives an interpolating polynomial that is smoother and has a smaller error than some other interpolating polynomials. If there are  $n$  data points, then the spline  $S(x)$  is the function

$$\begin{aligned} C_1(x), \quad x_0 \leq x \leq x_1 \\ S(x) = C_i(x), \quad x_{i-1} \leq x \leq x_i \\ C_n(x), \quad x_{n-1} \leq x \leq x_n \end{aligned} \quad (3)$$

Each  $C_i$  is a cubic function. The most general cubic function has the form

$$C_i(x) = a_i + b_i x + c_i x^2 + d_i x^3 \quad (4)$$

To determine the cubic spline  $S(x)$ , need to decide  $a_i$ ,  $b_i$ ,  $c_i$  and  $d_i$  for each  $i$  by:

$$\begin{aligned} C_i(x_{i-1}) = y_{i-1} \text{ and } C_i(x_i) = y_i, \quad i = 1, \dots, n. \\ C'_i(x_i) = C'_{i-1}(x_i), \quad i = 1, \dots, n-1. \\ C''_i(x_i) = C''_{i+1}(x_i), \quad i = 1, \dots, n-1. \end{aligned} \quad (5)$$

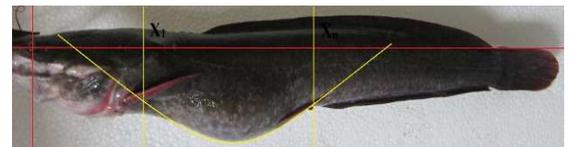


Fig. 17 Example of interest points

After getting all the interest points and the stable functions  $f(x)$  has been produced as illustrated in Fig. 17, the area of the interested region was estimated by using Gaussian Quadrature. Gaussian Quadrature is an accurate and effective method to deal with the definite integral of a

function, which is usually illustrated as a weighed sum at specified points of function values. The general formulation of Gaussian Quadrature as shown in the equation below:

$$\int_a^b f(x)dx \approx c_1 f(x_1) + c_2 f(x_2) + \dots + c_n f(x_n) \quad (6)$$

n=2: two-point method:  $c_1 = 1, c_2 = 1, x_1 = -0.5777\dots$   
 $x_2 = -0.5777\dots$

n=3: three-point method:  $c_1 = 0.555\dots, x_1 = -0.77\dots$   
 $c_2 = 0.888\dots, x_2 = -0.00\dots$   
 $c_3 = 0.555\dots, x_3 = -0.77\dots$

Firstly, n must be chosen whether n=2 for the two-point method or n=3 for the three-point method. Then,  $c_i$  and  $x_i$  that need to use must be identified. The boundaries from [a,b] to [-1,1] must be converted as follows:

In the form of x:

$$x = \frac{b-a}{2}t + \frac{b+a}{2}, dx = \frac{b-a}{2}dt \quad (7)$$

Becomes:

$$\int_a^b f(x)dx = \int_{-1}^1 f\left(\frac{b-a}{2}x + \frac{b+a}{2}\right) \frac{b-a}{2} dx \quad (8)$$

Therefore, the formula of Gaussian Quadrature used as illustrated below, was evaluate based on chosen  $n, c_i$  and  $x_i$  to get the area of the interesting region, as shown in Fig. 18 and the results from the tested images is presented in Table II.

$$\int_{-1}^1 g(x)dx \approx \sum_{i=1}^n c_i g(x_i) \quad (9)$$

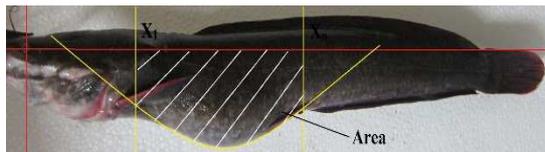


Fig. 18: Area of an interesting region

TABLE II  
THE ACCURACY RATE FOR FECUNDITY ESTIMATION

Sample Images	Total eggs (Groundtruth)	Total eggs (Autocount)	Percentage of accuracy
1	71065	78531	90.49%
2	81278	88190	92.16%
3	95415	103215	92.44%
4	59288	62473	94.90%
5	72386	80315	90.13%
6	134376	135076	99.48%
7	101385	105439	96.16%
8	135861	136980	99.18%
9	157859	159784	98.80%
10	64781	68378	94.74%

Overall, Table II provides the results from the tested images in this study. Several images were taken for being process to find the percentage of accuracy using the proposed method. The details of the percentage accuracy are presented in Table II below. The major inaccuracy factors in the calculation because of the unhealthy fish or stomach swollen syndrome in the abdominal area. In Table II, the result shows the accuracy mean of 94.85% with a standard deviation of 3%. Since in the standard practice of estimation, that any accuracy rate above then 85% is considered good, thus 94.85% accuracy derived from the proposed method is acceptable for real-life applications.

#### IV. CONCLUSION

In conclusion, the common method used to estimate the number of eggs is time-consuming due requires the whole ovaries to return to the laboratory. Therefore, we address the problem by utilizing constructing a computer vision algorithm. We have successfully demonstrated the feasibility of using an algorithm for an automated *Clarias gariepinus* fecundity egg estimation technique. In the upcoming study, it is essential to develop a mobile application to support this research further and to test a new or another mathematical model for improvement and as a comparison with our current results.

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