Songklanakarin J. Sci. Technol.
44 (1), 112-118, Jan. - Feb. 2022

http://www.sjst.psu.ac.th

Original Article

Image technology based detection of infected shrimp in adverse environments♣

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Received: 22 March 2021; Revised: 1 September 2021; Accepted: 7 September 2021

Abstract

In recent years, countries around Japan and especially in Southeast Asia, white spot disease (WSD) is highly infectious and severely damages shrimp aquaculture. At the same time, the various diseases are occurring in shrimp farms. In the early stages of infection, shrimp shows three abnormal behaviors: (1) they appear in the shallow waters of the farm, (2) they do not move and do not eat even when feeding, and (3) they suddenly stop moving. Currently, infected shrimps are found by visual inspection, which places a burden on the farmers and delays the discovery. Therefore, in this paper, we proposed a system for detecting infected shrimp by using image processing technology in order to eliminate the delay of discovery and reduce the burden of farmers. According to our experimental results, the proposed system has 95% precision, 100% recall rate and an accuracy of 96.4% by using hold-out evaluation method.

Keywords: infected shrimp detection, image processing techniques, shrimp feeding behaviors, artificial sea water

1. Introduction

In recent years, the cultivation of whiteleg shrimp has become popular in countries around Japan and especially in Southeast Asia. It is said that 3/4 of the shrimp produced in the world are whiteleg shrimp. The growing period of black tiger shrimp is about five months while the growing period of whiteleg shrimp is about three months. The production efficiency of whiteleg shrimp is good because it took about a month to grow (Sea food aquarium, 2018). About 77.9% of whiteleg shrimp farming production is in Asia while the rest is produced from the Americas (Maeda, 2014). The profitability is the main reason behind shrimp farming industry and it is important to control various diseases that can occur at shrimp farms to maintain the production rate (Asche *et al*., 2020).

Due to the high-density culture in shrimp production, various diseases occur in shrimp farms (Inada, Mekata, & Itami, 2017). Among them, White spot disease (WSD) is causing enormous damage and high mortality rate in production of various types of shrimp (Debnath, Karim, Keus, Mohan, & Belton, 2016; Rodríguez *et al*., 2003). The WSD was caused by White Spot Syndrome Virus (WSSV) and it is the most destructive disease in shrimp farming industry (Millard *et al*., 2020). The WSD infected shrimp can reach 100% mortality in 3–7 days causing significant economic

♣Peer-reviewed paper selected from The 1st International Conference on Sustainable Agriculture and Aquaculture (ICSAA-2021)
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losses to the shrimp farming industry (Ramos-Carreño *et al*., 2014; Zhu, Twan, Tseng, Peng, & Hwang, 2019). The infected shrimp showed abnormal behaviors such as appear in the shallow area of the farm, stop eating food and suddenly stop moving (Morimoto, 2018; Seike, 2019). The WSD is also equivalent to penaeid acute viremia (PAV) which has serious impact on both shrimp farming industry and hatchery production in the Americas and Southeast Asia countries (Satoh, 2012). The WSSV is sometimes misdiagnosed as yellow head virus (YHV) because of similarity structure between these two viruses (Pantoja & Lightner, 2003).

The infection with WSD is characterized by white spots on the exoskeleton of the infected shrimp. The WSD transmission can spread out by following ways: (i) cannibalism between shrimp, (ii) transmission from parent shrimp and (iii) waterborne infection. There is no effective cure because it is caused by viral infection hence, early detection of infected shrimp is important. Therefore, we proposed a system for detection of infected and non-infected shrimp based on the shrimp behavior in the aquarium. The rest of the paper is organized as follows: section 2 presents the materials and methods used in the proposed system, section 3 describes experimental environments and section 4 describes results and discussions. Finally, the conclusions and future work are described in section 5.

2. Materials and Methods

In this paper, we proposed a system for detection of infected and non-infected shrimp based on the shrimp behavior in the aquarium. We focused on the abnormal behavior of infected shrimp not eating food. Basically, the infected shrimp are staying away from food even feeding (Sawachika, 2017). Food was placed in the container and border was covered with tape. The proposed system consisted of three functional modules: (i) extraction of feeding area, (ii) feeding confirmation of shrimp, and (iii) infected shrimp detection. The system flow diagram of proposed system is shown in Figure 1.

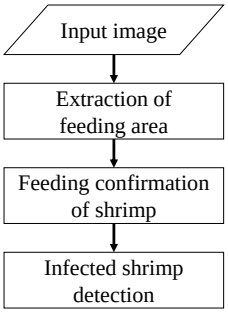

Figure 1. System flow diagram of the proposed system

2.1 Extraction of feeding area

The image without containing shrimps was taken as background image and it was converted from RGB (red, green and blue) to HSV (hue, saturation and value) color space. This color space conversion from RGB to HSV can be done by using following equations. We used MATLAB programming language to make conversion process from RGB to HSV.

$$I_{\max} = \max\{R,G,B\} \tag{1}$$

$$I_{\min} = \min\{R,G,B\} \tag{2}$$

$$V = I_{\max} \tag{3}$$

$$S = (I_{\max} - I_{\min})/I_{\min} \tag{4}$$

$$H = \begin{cases} \dfrac{G-B}{I_{\max}-I_{\min}} \times \dfrac{\pi}{3} & , \text{if } I_{\max} = R \\ \dfrac{B-R}{I_{\max}-I_{\min}} \times \left(\dfrac{\pi}{3} + \dfrac{2}{3}\pi\right) & , \text{if } I_{\max} = G \\ \dfrac{R-G}{I_{\max}-I_{\min}} \times \left(\dfrac{\pi}{3} + \dfrac{4}{3}\pi\right) & , \text{if } I_{\max} = B \end{cases} \tag{5}$$

In these equations, the I is the intensity values of input image values. This color space conversion process is shown in Figure 2(a) and (b). And then, this image was a threshold into binary image for feeding area extraction. After feeding area was extracted, image preprocessing steps such as noise removing and morphological operations (Masatoshi, 2016) were performed. Then, the filling process (Soille, 1999) was made on this binary image to fill the hole region for getting food region area. This process is shown in Figure 2(c) and 2(d).

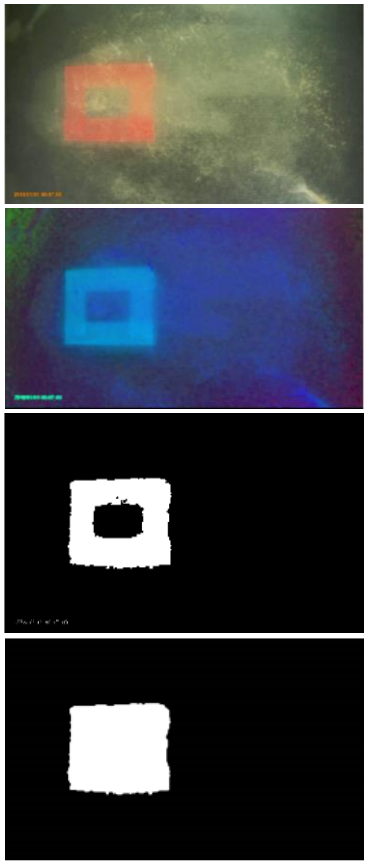

Figure 2. (a) Original RGB image, (b) HSV image, (c) Binary image after threshold, (d) Binary image after filling process

2.2 Feeding confirmation of shrimp

The shrimp comes to feeding area to eat. When shrimp comes to this area, the shape of the background subtraction image changed. To detect shrimp, the current frame was subtracted from the background image. Finally, the XOR (exclusive OR) operation was performed between the resulted image and the background image to extract the shrimp. The input image and background subtracted image is illustrated in Figure 3(a) and (b) and the XOR operation between background image and background subtracted image is shown in Figure 3(c). This process was done frame by frame and we could detect the existence or absence of shrimp in food region. After XOR operation, some noise may contain in the result image. We used morphology operation to eliminate noise contained in the image. This process is shown in Figure 4.

2.3 Infected shrimp detection

To distinguish infected and non-infected shrimp, we used feeding behavior as a feature. The number of visits and staying time in the feeding area are recorded for each shrimp. Basically, the characteristic of infected shrimp is that they are not interested in food and keep away from food area even though feeding (Morimoto, Zin, & Itami, 2018; Morimoto, 2020). The threshold values are set for visit and stay time at food region area to distinguish infected and non-infected shrimp. The effective feeding is regarded as the shrimp stays for a certain period of time. We have total of 32 recorded videos. From these videos, we used four videos of non-infected shrimp data as training data and the optimal threshold values are calculated. This information is shown in Table 1. From the training data, the least number of visits and eating time spent in food region is searched and these values are set as threshold value.

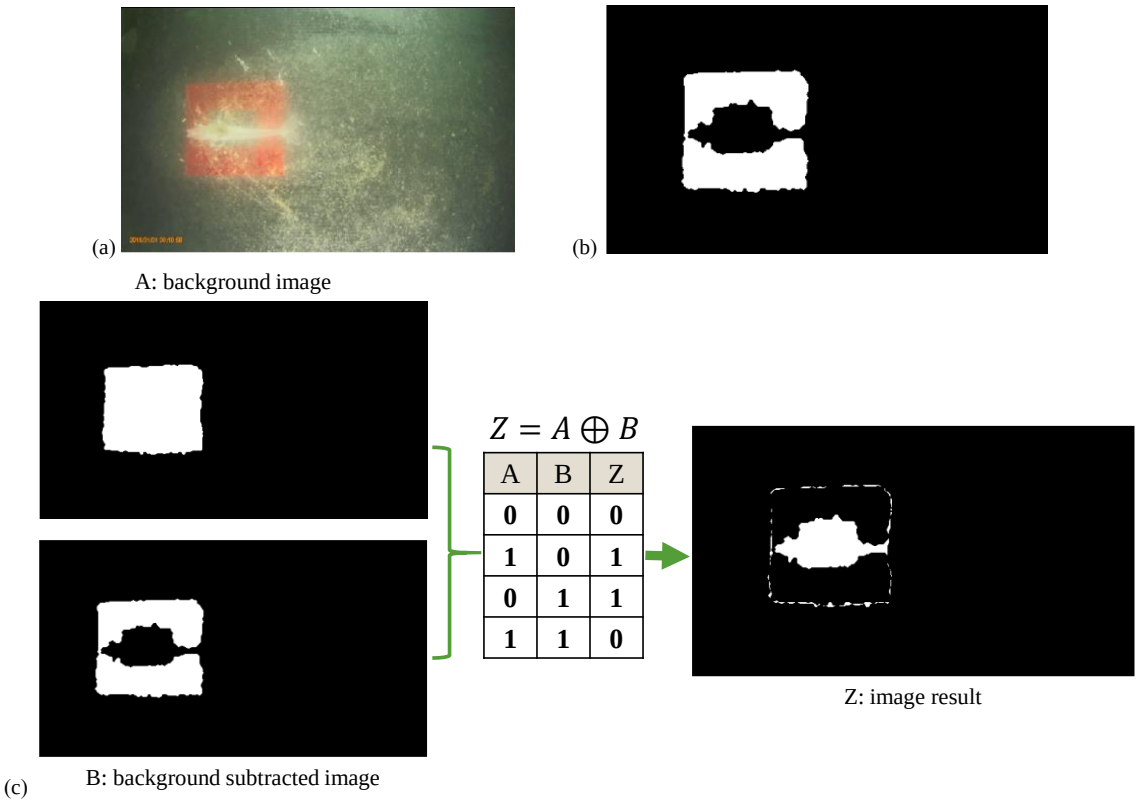

Figure 3. (a) Input image, (b) Background subtracted image, (c) The XOR operation result image

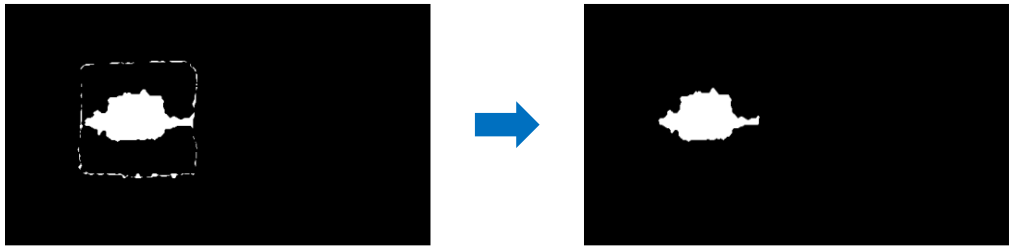

Figure 4. The final process of shrimp extraction

Table 1. Training data information

Name	Number of visits (times)	Number of stay frame in feeding area (frame)	Average number of stay frame (frame)
Day1 T1R3	2	18, 34	26
Day2 T1R3	2	3, 39	21
Day3 T1R3	6	5, 6, 9, 14, 16, 21	11.8
Day4 T1R3	5	3, 11, 14, 15, 26	13.8

From the training data, the optimal threshold value is chosen as $T_{visit} = 2$ which is the least number of time that the shrimp come to food region. In addition, the minimum number of average stay frame ($T_{avgFrame}$) for shrimp is chosen as $T_{avgFrame} = 12$. We used effective feeding (ET) as a feature to classify whether the shrimp is infected or non-infected as shown in equation 6. The ET value is incremented if the conditions are satisfied.

$$ET = \begin{cases} 1 & ,if\ (\text{number of visit} \geq T_{visit})\ and \\ & (\text{number of stay frame} \geq T_{avgFrame,otherwise}) \\ 0 & \end{cases} \quad (6)$$

If the final ET value is less than two, the system determined the shrimps are infected with virus by using the following equation.

$$\text{Result} = \begin{cases} \text{Infected} & ,if\ (ET < 2) \\ \text{Non} - \text{infected} & ,otherwise \end{cases} \quad (7)$$

3. Experiment Environment

In this paper, the experiments were performed at the Aquatic Science and Innovative Management Division, Faculty of Natural Resources, Prince of Songkla University, Thailand. The total of nine aquaria or tanks were prepared for experiments. The size of each aquarium has a dimension of 60 × 40 × 40 cm and the overhead camera is setup. We randomly selected the white leg shrimp raised in the Prince of Songkla University and put three shrimps in each aquarium. Prior to use the shrimp was confirmed to be WSSV free by PCR. After that, artificial injection of WSSV were conducted. The size of shrimp is approximately between 10 and 12 grams. A commercial shrimp feed was used in this experiment. The three aquariums, T1R1 to T1R3 contained artificial sea water with non-infected shrimp. The T2R1 to T2R3 aquaria contained artificial sea water with infected shrimp. The T3R1 to T3R3 aquaria were set up with farm water and infected shrimps. The farm water was taken from shrimp farm in Nakhon Si Thammarat Province, Kingdom of Thailand. If the experiment was carried out in a situation where the salt concentration was different between the farm water and the artificial sea water, the correct result could not be obtained. So, the experiment was conducted with the same salt concentration. The salinity of the water is 30 ppt.

The *Penaeus vannamei* (whiteleg shrimp), the main cultured species in Southeast Asia, was used. The experiment period was from 12th to 19th September, 2019. Feeding was made three times a day (8:30 am, 12:30 pm and 4:30 pm). The video recording was performed twice a day (8:30 am, 12:30 pm) 5 minutes after feeding was made. The total of 32 videos are recorded and four videos are used as training data and remaining 28 videos are used as testing data. The Figure 5 showed the experimental environment and setup.

The camera used in this experiment was MUSON MC2 Pro1 with resolution of 480 × 640 pixels. The processed frame rate was 1 fps (frame per second). Each recorded video has 300 frames. We calculated the ET value till the end of each video and calculated the one final ET value for each video. The information of testing data used in the experiment is shown in Table 2. The evaluation in this experiment was performed under the following conditions. When the shrimp has stayed in feeding area for 12 seconds (12 frames) or less, the effective number of feedings was 1 or less. When the above conditions were met, the shrimp in the aquarium were to be infected with the disease. The threshold value, ET was set with reference to the value from the training data.

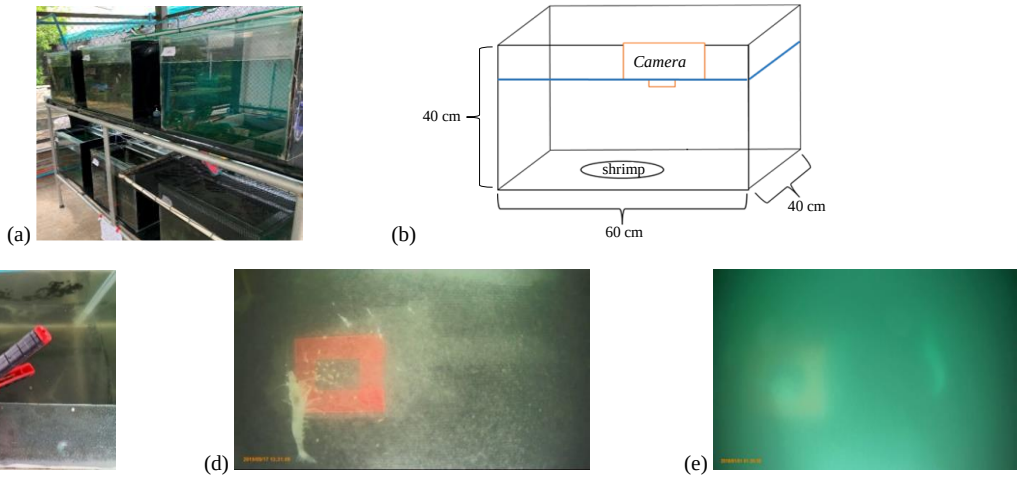

Figure 5. (a) Experiment environment, (b) Diagram of experimental environment, (c) Camera setup, (d) Artificial seawater and (e) Farm water

Table 2. Testing data information

Aquarium		Number of data (video)	Resolution (pixel)	Shooting time (seconds)
T1	R1	4	480 × 640	300
	R2	5		
T2	R2	4		
	R3	5		
T3	R1	4		
	R3	6		

4. Results and Discussions

The number of testing data used in the experiment was nine for non-infected shrimp and 19 for infected shrimp (10 of which were farm water). The experimental results on the 3rd day of recorded video of non-infected shrimp at aquarium T1R1 is shown in Figure 6 and detailed information are described in Table 3. The number of staying frames in feeding area are 33, 9, 19 and 7 and effective number of feeding is two times. Therefore, this data is determined to be non-infected condition.

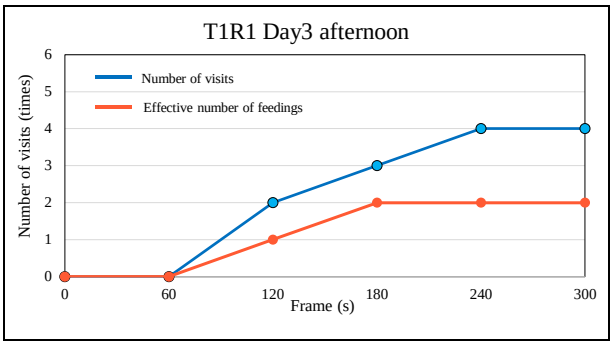

Figure 6. Information of T1R1 aquarium

Table 3. Total number of visits and eating on feeding area at T1R1 and T1R2

Aquarium			NV	ET	NSF	Result
T1	R1	Day 3 afternoon	4	2	33,9,19,7	Non-infected
		Day 4 afternoon	5	2	7,7,29,5,26	Non-infected
		Day 6 morning	7	4	28,4,3,83,44,4,21	Non-infected
		Day 7 morning	7	2	6,27,31,4,3,4,9	Non-infected
	R2	Day 2 morning	4	2	7,6,12,92	Non-infected
		Day 3 afternoon	4	2	11,3,21,12	Non-infected
		Day 4 afternoon	5	2	3,11,56,3,65	Non-infected
		Day 6 afternoon	2	1	14,120	Infected
		Day 7 morning	3	3	14,19,139	Non-infected

NV: Number of visits (times), ET: Effective number of feeding (times), NSF: Number of stay frame in feeding area (frame)

According to research data, the infected shrimp appeared their symptoms starting from two or three days. Therefore, we generally choose the data at 3rd day to illustrate the experimental condition. The experimental results on the 3rd day of recorded video of infected shrimp at aquarium T2R2 is shown in Figure 7 and detailed information are described in Table 4. Even though the shrimp visited feeding area twice, the shrimp did not stay in the feeding area sufficiently and so the effective number of feeding was zero. Therefore, this day was classified infected condition by the system.

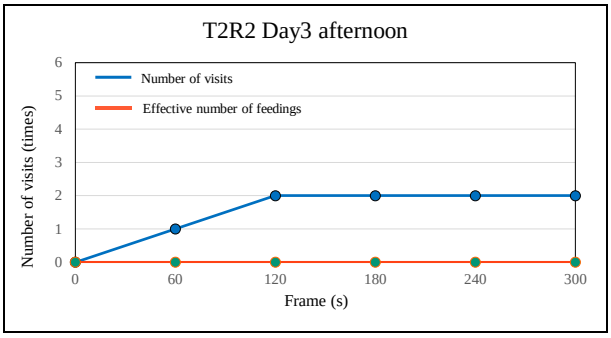

Figure 7. Information of T2R2 aquarium

Table 4. Total number of visits and eating on feeding area at T2R2 and T2R3

Aquarium			NV	ET	NSF	Result
T1	R2	Day 2 morning	3	0	5,4,3	Infected
		Day 3 afternoon	2	0	4,3	Infected
		Day 4 afternoon	1	1	16	Infected
		Day 6 morning	1	0	5	Infected
	R3	Day 2 morning	3	0	4,3,4	Infected
		Day 3 afternoon	2	0	6,9	Infected
		Day 4 afternoon	1	0	5	Infected
		Day 6 morning	1	0	3	Infected
		Day 7 morning	1	0	3	Infected

NV: Number of visits (times), ET: Effective number of feeding (times), NSF: Number of stay frame in feeding area (frame)

The experimental results on the 3rd day of recorded video of infected shrimp at aquarium T3R1 is shown in Figure 8 and detailed information are described in Table 5. In Table 5, the effective number of feeding count was zero because shrimps never visited the feeding area. By comparing the results with the data of non-infected shrimp and infected shrimp taken at the same time on the same day, there was a tendency for a large difference in the number of visits and the number of stay frames. Therefore, the characteristic that infected shrimp became less eating was remarkable from the results of experiment. Since the effective number of feedings was zero time and the system determined as infected condition.

In order to clarify the cause of the death of shrimp, the PCR (polymerase chain reaction) test was performed with the cooperation of Aquatic Science and Innovative Management Division, Faculty of Natural Resources, Prince of Songkla University. The PCR test was to check for the presence of pathogens that cannot be seen with a microscope

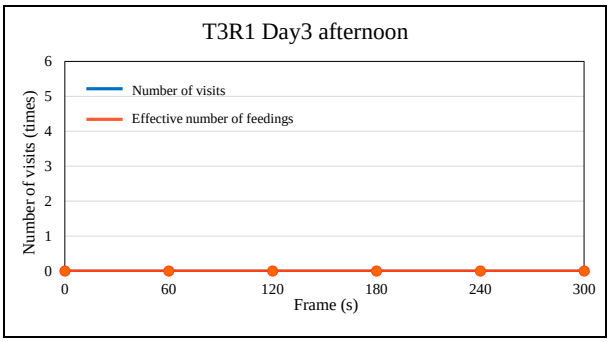

Figure 8. Information of T3R1 aquarium

Table 5. Total number of visits and eating on feeding area at T3R1 and T3R3

Aquarium			NV	ET	NSF	Result
T3	R1	Day 3 afternoon	0	0	0	Infected
		Day 4 afternoon	0	0	0	Infected
		Day 6 morning	1	1	26	Infected
		Day 7 morning	2	0	5,6	Infected
	R3	Day 2 morning	4	1	7,7,5,22	Infected
		Day 3 afternoon	1	1	12	Infected
		Day 4 afternoon	3	1	3,7,33	Infected
		Day 5 afternoon	0	0	0	Infected
		Day 6 morning	0	0	0	Infected

NV: Number of visits (times), ET: Effective number of feeding (times), NSF: Number of stay frame in feeding area (frame)

such as a virus. It is a test method that detects by amplifying the DNA of the pathogen, a reliable diagnosis can then be made. As a result of PCR examination, WSD was the cause of death for all shrimp except one shrimp. White spots were formed on the outer shell of infected shrimp. The sample image of white spots found in the death shrimp is shown in Figure 9.

The Table 6 shows the processing results and accuracy of the data used in this experiment. The system could correctly identify 27 out of 28 data. As for the data of infected shrimp, all the data could be accurately identified in both artificial seawater and farm water. To evaluate our proposed system, we calculated precision, recall and F1 score by using the following equations:

$$\text{Precision} = \frac{TP}{TP + FP} \tag{8}$$

$$\text{Recall} = \frac{TP}{TP + FN} \tag{9}$$

$$\text{F1} = 2 \times \frac{\text{Precision} \times \text{Recall}}{\text{Precision} + \text{Recall}} \tag{10}$$

where *TP* is true positive, *FP* is false positive, and *FN* is false negative. Our proposed system obtained 95% precision, 100% recall rate and F1 score of 0.97. Overall, the proposed system has an accuracy of 96.4%.

According to the literature survey, most of the experiments are processed on single captured image that is outside of water environment but our experiments are performed on sequences of image data which were operated in underwater environment. The strength of our system is that it directly monitored the shrimp behaviors to identify whether the shrimp is infected with virus or not. To the best of our knowledge, our proposed system is the first study to implement about monitoring system for infected shrimp detection using image sequences in underwater environment.

Figure 9. Typical white spots on the carapace of the infected shrimp

Table 6. Accuracy of the proposed system

Aquarium	Number of data (video)	Correct	Accuracy (%)
T1	9	8	88.8
T2	9	9	100
T3	10	10	100
Total	28	27	96.4

T1: Non-infected shrimp, T2: Infected shrimp in artificial sea water, T3: Infected shrimp in farm water

5. Conclusions and Future Work

In this study, we proposed a system for detecting infected shrimp by using image processing technology. In the proposed method, we focused on the characteristic that shrimp infected with the disease did not approach the food and did not move to the food area. The infected shrimp and non-infected were identified based on the number of visits and the staying time in feeding area. As a result of this experiment, the proposed system correctly identified 27 shrimps out of 28 samples and having the accuracy of 96.4%. The system correctly identified on infected shrimp for both artificial sea water and farm water condition. Therefore, the proposed system will be applicable and useful in shrimp farms. For the future works, the experiment will make on different environment with various conditions of shrimps such as infected and non-infected shrimp will be mixed.

Acknowledgements

This research was partially supported by Grant of Fukuyama University, Japan.

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