# Correlation between Serum Vitamin A Level and Pupil Color of Beef Cattle

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*Abstract*— Farmers usually control cattle's serum vitamin A level to improve beef meat fat quality. They sometimes conduct blood tests, but that is invasive and another measurement system is desirable. The objective of this study was to investigate the relationship between serum vitamin A level and pupil colors of beef cattle for developing a vitamin A monitoring system by machine vision. Images of cattle eyes were collected and chromaticity values of pupil color were calculated. Although the values had variability, it was found that serum vitamin A had a negative correlation with red component, while it had a positive correlation with blue component.

#### I. INTRODUCTION

Japanese people are particular about food quality and there is a big demand for high-quality beef.

For judging beef quality, Japan meat grading association provides 4 indexes-- marbling score, beef color, color and quality of fat, and texture and firmness of meat. The most important factor is marbling score because it affects taste very much. Therefore we can say that high marbling score means high-quality.

To improve marbling score, controlling cattle's serum vitamin A is very effective [1][2][3]. In fact, some fattening farmers maintain cattle's vitamin A at low level, about 30 IU/dl, in middle fatting phase (16 to 24 months of age) (Fig. 1).

Farmers sometimes conduct blood test for monitoring vitamin A level. However, it has many disadvantages— it is time-consuming, expensive, and stressful for cattle. That's why another measurement system is desirable.

Recently the relationship between vitamin A and eye ground color has been reported. If vitamin A level becomes lower, blue part of retina sometimes became red or yellow [4]. From that study, we assumed that it is possible to estimate serum vitamin A level from retina colors (Fig. 2).



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Fig. 1 Ideal vitamin A level control in fatting phase



Fig. 2 Cattle's eye diagram

However retina is difficult to take images. On the other hand, pupil area is easy to take images and there is a possibility that pupil color is influenced by retina color. Therefore we started to take pupil images and investigate its color.

The objective of this study is to develop a simple and non-stressful method of serum vitamin A level measurement by analyzing pupil color images of cattle.

#### II. MATERIALS AND DEVICES

# A. Materials

This experiment started from December in 2010 at Hyogo Prefectural Hokubu Agricultural Institute with 24 heads of Japanese black cattle in early fatting phase (9 to 11 months of age). Serum vitamin A levels of these cattle were 44 to 126 IU/dl. From March in 2011, another 18 cattle in early fatting phase (10 to 12 months of age) were added and the total number of cattle became 42.

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## B. Experimental devices

The device we used is shown in Fig. 3. It was composed of 2CCD camera (JAI, AD-080CL), 12mm Lens (EDMUND, 63240-L f-number is 1.8), White LEDs (MORITEX, MDRL-CW5), Near-infrared (NIR) LEDs (MORITEX, MDRL-CIR31), and Polarization (PL) filter (MORITEX, ML-PL270LB). We could take color image and NIR image at the same time. PL filters were attached to reduce the halation caused by the specular reflection. The tube was set with the front edge of camera to prevent external light and maintain the same distance during the experiment. Inside the tube, non-reflect cloth was attached.

# III. METHODS

# A. Experimental Method

The camera condition is shown in Table 1. The values of shutter speed, f-number, and master gain were fixed as bright as possible.

TABLE. 1

CAMERA CONDITION		
Shutter speed [s]	1/30	
F-number	1.8	
Master gain	336	
The brightness [lx]	1700	
The length of tube [mm]	100	

Before starting to take images, white balance was checked every time using color chart (Munsell Color Checker, X-rite). After that, we covered the cattle eye with black cloth and waited more than 120 s in order to dilate pupil. Next, we put the NIR LED on and set the camera to pupil position. Later, put white LEDs on and started to collect images. Images were taken continuously for 3 s with the interval of 1/ 30 s. Right eye images were taken in this study.

Within a few days before the experiment, serum vitamin A level was measured with blood test. These vitamin A data were also used for analysis.



Fig. 3 Color camera used in the present study

# B. Method of Analysis

Images taken from December in 2010 to July in 2011 were processed manually by using image processing software (WinROOF, Mitani Corporation, Japan).

After that, we made image processing software using C language. Here is the method of image processing with that programming (Figs. 4 to 13).



Fig. 4 Flow chart of image processing



Fig. 5 Color image of cattle's right eye



Fig. 6 NIR image of cattle's right eye



Fig. 7 Image before reducing noise



Fig. 8 Image after Salt-and-pepper reduction



Fig. 9 Image after filling missing pixels caused LED



Fig. 10 Noise was reduced again



Fig. 11 Halation part was detected by (Fig. 10) – (Fig. 8)



Fig. 12 ROI was Determined



Fig. 13 ROI was adopted to color image

First, the image was binarized using R, B, Hue, IR components. Second, Salt-and-pepper noise reduction (Fig. 8) was done. Later, fill missing pixels caused by LED (Fig. 9) and reduced noise (Fig. 10) again. After detecting halation part (Fig. 11) by (Fig. 10) – (Fig. 8), only this part was binarized using R, B, Hue components. Through this process, ROI was determined (Fig. 12). Finally, we got the mean RGB values and calculated chromaticity values and HSI values. RGB is an additive color model and each color has 8 bits per 1 pixel. Chromaticity values and HSI are calculated as below.

$$r = R/(R+G+B)$$
  

$$g = G/(R+G+B)$$
  

$$b = B/(R+G+B)$$
(1)  

$$M_{I} = \frac{2}{\sqrt{6}}R - \frac{1}{\sqrt{6}}G - \frac{1}{\sqrt{6}}B$$
  

$$M_{2} = \frac{1}{\sqrt{2}}G - \frac{1}{\sqrt{2}}B$$
  

$$I_{I} = \frac{1}{\sqrt{3}}R + \frac{1}{\sqrt{3}}G + \frac{1}{\sqrt{3}}B$$
  

$$H = \tan^{-1}(M_{I}/M_{2})$$
  

$$S = (M_{I}^{2} + M_{2}^{2})_{2}^{1}$$
  

$$I = \sqrt{3}I_{I}$$

Where,	<i>r</i> : red component	g : green component
	<i>b</i> : blue component	H: Hue
	S: Saturation	I : Intensity

Using *r*,*g*,*b*, HSI and the data of vitamin A collected in advance, graphs of time-series data were made about each cattle.

## IV. RESULT AND DISCUSSION

#### A. Result of Present Study

As shown in Figs 14 and 15, February data and March data are omitted because serum vitamin A levels in February were abnormal and we didn't take images of right eye in March.

The relationship between r and vitamin A level is Fig. 14. When vitamin A level became lower, r became higher. When vitamin A level became higher, r became lower. It is a negative correlation. This tendency was found in 10 cattle among 21.

The relationship between b and vitamin A level is shown in Fig. 15. When vitamin A level became lower, b became lower. It is a positive correlation. This tendency was found 11 cattle among 21.



Fig. 14 Relationship between *r* and vitamin A level (Left vertical axis: vitamin A level, Right vertical axis: *r*)



Fig. 15 Relationship between *b* and vitamin A level (Left vertical axis: vitamin A level, Right vertical axis: *b*)

## B. Result of Previous Study

We compared the experimental results to those of the same experiment that was carried out from July to November in 2010 with 43 cattle in the middle fatting phase (15 to18 months of age). Serum vitamin A levels of these cattle were under 50 IU/dl. The camera condition is as below (Table 2). F-number was different from present study because the lens used in previous study was a different type.

TABLE. 2		
THE CAMERA CONDITION		
Shutter speed [s]	1/30	
F-number	2.2	
Master Gain	336	
The brightness [lx]	1700	
The length of tube [mm]	100	

The time-series data of one cattle (No.226) are as shown in Figs.16 to 18. There is a negative correlation between vitamin A and r, and positive correlation between vitamin A and b. We can find the same tendency as Fig. 14 and 15. In addition, S also has a positive correlation with vitamin A.



Fig. 16 Relationship between *r* and vitamin A level (Left vertical axis: *r*, Right vertical axis: vitamin A level)



Fig. 17 Relationship between *b* and vitamin A level (Left vertical axis: *b*, Right vertical axis: vitamin A level)



Fig. 18 Relationship between S and vitamin A level (Left vertical axis: S, Right vertical axis: vitamin A level)

## C. Discussion

In both the early and middle fatting phases, the same tendency was found. There is a negative correlation between r and vitamin A, and there is a positive correlation between b and vitamin A. It is also suggested that the changes of pupil color are influenced by change of retina color.

However, these tendencies were not found in all cattle. Serum vitamin A level of some cattle has no relationship with pupil color. We need to solve this problem to get more precise results. We also need to study cattle in the final fatting phase.

#### V. CONCLUSION

We got the positive or negative correlation between serum vitamin A level and pupil color with about the number of samples in the present study.

## ACKNOWLEDGMENT

This study was supported by Grants-in-Aid for Scientific Research (23380153). We gratefully acknowledge the financial support of Ministry of Education, Culture, Sports, Science and Technology. We also want to thank the staff of Hyogo Prefectural Hokubu Agricultural Institute for their help and support with the cattle. The authors gratefully acknowledge the useful suggestions and technical assistance of Prof. Malcolm Fitz-Earle in the preparation of this paper.

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