

## Immune Response of Dry Holstein Vaccinated by Killed Avian Influenza H5N1 Vaccine

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### INTRODUCTION

The establishment and spread of highly pathogenic avian influenza (HPAI) viruses of the H5N1 subtype in birds and coincident infections in humans since 2003 have raised concerns that we may be facing an influenza pandemic caused by an H5N1 influenza virus [4]. Globally, from January 2003 to 2 March 2017, there were 860 cases of human infection with avian influenza A(H5N1) virus reported from 16 countries worldwide. Of these 860 cases, 454 were fatal (CFR of 53%). The last case was reported from Indonesia on 26 September 2017 [7]. Although the human cases of H5N1 in Indonesia has decreased significantly since 2010, according to WHO until 2017 there have been reported 200 cases H5N1 in human with 168 cases of deaths. The last case was reported from Indonesia on 26 September 2017. This is indicate Indonesia as the highest fatal case of H5N1 globally. With continued incidence of avian influenza due to existing AI H5N1 viruses in poultry, the avian influenza H5N1 has been believed remain to threaten Indonesia [6].

Passive immunization using specific antibody against AI H5N1 from bovine colostrum is one of an alternative to control H5N1 virus infection due to lack of H5N1 vaccine production for human. Bovine colostrums consider an ideal alternative antibody source, as the antibody in the bovine's blood is transported to mammary gland easily and accumulates in the colostrum in large quantities. As a "biological factory" and the source of natural antibody, bovine colostrum could be designed to produce the specific antibody against certain disease for animal and human by immunizing the dry cow (with the antigen of interest).

Vaccine is an antigenic material used to produce active immunity against diseases. Vaccination is the administration of vaccine to the individual to generate immunity against a disease [2]. According to [1], the exposure of a dry cow against antigen (vaccine) will produce specific antibody in their blood circulation. Therefore, evaluation on the status of antibody against AI H5N1 following the vaccination of dry Holstein cows with killed AI H5N1 vaccine is needed.

### MATERIALS AND METHODS

Dry Holstein cows, on 2<sup>nd</sup>-3<sup>th</sup> lactation, clinically healthy, were divided into 2 groups, control (n=3) and treatment (n=7). The treatment group were injected by commercial killed avian influenza (AI) H5N1 vaccine subcutaneously, double doses/head, three times every two weeks. Blood samples were collected from coccygeal vein, starting before the 1<sup>st</sup> vaccination (0 week), repeated every two weeks until calving for serum antibody against AI H5N1 analysis. Serum antibody against AI H5N1 were detected using haemagglutination inhibition test [8].

### RESULT AND DISCUSSION

Results of this study showed that the administration of avian influenza H5N1 killed vaccine double doses per head, three times every two weeks may induce the formation of antibody against AI H5N1 in their blood. However, the ability of antibody induction are various among individual cows. Out of seven cows vaccinated, one cow did not show antibody against H5N1 AI in her blood. The remaining six cows expressed enough titer (detectable and protective amounts) of antibody (Table 1).

Table 1 Titer of serum antibody of Holstein cows vaccinated by *Killed AI H5N1 Vaccine*

Cow Code	Titer of Antibody Against AI H5N1 (log 2)			
	Before Vaccination	Two Weeks After 1 <sup>st</sup> Vaccination	Two Weeks After 2 <sup>nd</sup> Vaccination	Two weeks After 3 <sup>rd</sup> Vaccination
<b>Control</b>				
118	0	0	0	0
1719	0	0	0	0
1767	0	0	0	0
<b>Vaccinate d</b>				
888	0	0	0	5
1862	0	0	0	0
1587	0	0	0	3
30	0	3	3	5
220	0	2	3	4
713	0	4	4	5
776	0	3	4	5

Although the majority (85%) of vaccinated cows have shown the ability to produce antibodies, the variation among individuals were observed on the onset of antibody detected. Four out of seven vaccinated cows responded very well, showing high antibody titer (cows code 30, 220, 713, 776). Specific antibody against H5N1 were detected in their blood serum using HI test two weeks following the 1<sup>st</sup> vaccination, with the titer as high as 2<sup>2</sup>-2<sup>4</sup>. After the second administration of vaccine (*booster*) the titer of antibody continued to increase until two weeks after the 3<sup>rd</sup> vaccination, with the titer of antibody between 2<sup>4</sup>-2<sup>5</sup>. The serum antibody anti H5N1 AI was detected later (two weeks after the 3<sup>rd</sup> vaccination) with the titer between 2<sup>3</sup>-2<sup>5</sup> in two remaining cows. One more cow (cow code 1862) was failed to show the antibody titer in her perifer blood (titer 2<sup>0</sup>) even after 2 weeks following the 3<sup>rd</sup> vaccination.

A various factors involve on the formation of antibody such as age, molecule size of antigen, the complexity of chemical structure of antigen, genetic, route of immunization, antigen dose and, the timing and frequency of immunization [3]. It has been reported that the body condition of the animal play an important role on the antibody formation. The body condition interfered by the feed, qualitatively and quantitatively. This has involve at the physiological condition and the immune response. On the other hand, stress, barn condition and the time of sampling may affect antibody concentration [5].

Vaccination is the administration of antigen collected from contageous agent. When exposed to other protein, the body will respond to the cellular and humoral immunity. In the immune system, the immunity mediated by cells (T-

lymphocytes), and provide cellular immunity by activating T-lymphocytes and lymphokines. On the other side, cells generated from humoral system (B-lymphocytes) provides a response on the antigenic challenge by providing the specific antibody. The antibody will be released in the blood and other body fluid [5].

## CONCLUSION

Dry Holstein cows vaccinated by commercial killed Avian Influenza H5N1 vaccine subcutaneously, double doses, three times every two weeks were able to produce specific antibody against antigen virus AI H5N1 in their blood. Dry Holstein cows were very responsive to AI H5N1 antigen injected. Dry cows could be immunized by commercial killed AI H5N1 vaccine and produce the antibody against AI H5N1 in their blood in detectable and protective amounts.

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