

## Lack of Antibody Formation Against Inactivated Avian Influenza Virus in Ducks and Chickens After Intranasally Immunization

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### INTRODUCTION

Vaccination is one of control strategies implemented in endemic countries such as Egypt and Indonesia [1,2]. Most commercial AI vaccines available in Indonesia are adjuvanted inactivated AI vaccines applied through intramuscular routes. Vaccine application by subcutaneous or intramuscular injection can cause pain and stress in poultry, the route of vaccine through the nasal drip (intranasal) is a more convenient and painless.

However, respiratory applied inactivated influenza is poorly immunogenic. Therefore prior to developing inactivated intranasal vaccine, it is necessary to study antibody response to inactivated AI virus which exposed through intranasal route. The aim of our research was to determined antibody response of ducks and chickens against avian influenza virus (AIV) subtype H5N1 after intranasally immunization.

### MATERIALS AND METHODS

A total of 22 AIV antibody negative ducks and 50 specified pathogen free (SPF) chickens were used in this research. Each animal groups were divided into two groups: control group and immunization group. Birds in control group were inoculated using 0,1 ml presterilized phosphate buffer saline (PBS), whilst the other groups were inoculated using 0,1 ml inactivated AIV subtype H5N1 clade 2.3.2 (A/Ck/SR1/15) containing 128 hemagglutinin unit (HAU)/25µl intranasally. Serum samples were collected at day 1, 4, 7 and 10 post inoculation. Antibody against AIV were determined using Enzyme Linked Immunosorbent Assay (ELISA) technique.

### RESULT AND DISCUSSION

Our results showed that 3 of 13 (23%) ducks inoculated with inactivated AI virus had specific antibodies to AI, while others did not form

specific antibodies against AI. Two of the three ducks with specific antibodies were positive only on day 1 after inoculation, and only one duck had positive AI antibodies on day 1,4,7, and 10 after inoculation. Whilst, in chickens, only 2 of 25 (8%) chickens inoculated with inactivated AI virus had antibodies specific to AI, antibodies detected on day 4 after inoculation. All birds among control group were not develop apecific antibody against AIV.

Our result were in accordance to earlier study by De Geus *et al.* [3] showed that adjuvanted inactivated AI H9N2 with chitosan or aluminum OH applied through intranasal route were not able to induce spesific antibody formation. However other study by Worall *et al.* [4] were in contrary with our result, their study showed that inactivated AI H5N1 vaccine with chitosan and sialidase applied through intanasal route were able to stimulated the formation of IgA specific to AI. In this study, we only evaluated the presence of antibodies in serum IgG, and did not evaluate the presence of specific IgA from the mucosa. Provision of antigens via intranasal route should stimulate local immune formation better.

### CONCLUSION

Lack of antibody formation might be due to low concentration of virus or most likely due to absence of adjuvantia, showing the need of improving immunogenicity of inactivated AIV if we want to develop intranasally vaccine.

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### REFERENCES

[1] Swayne DE. 2006. Principles for vaccine

protection in chickens and domestic waterfowl against avian influenza: Emphasis on Asian H5N1 high pathogenecity avian influenza. *Annals of the New York Academy of Science*. 1081: 174-181.

- [2] Siregar SW, Darminto, Weaver J, Bouma A. 2007. The vaccination programme in Indonesia. *Development in Biologicals (Basel)*. 130: 149-156.
- [3] De Geus ED, Van Harleem DA, Poetri ON, De Wit JJ, Vervelde L. 2011. A lack of antibody formation against inactivated influenza virus after aerosol vaccination in presence or absence of adjuvantia. *Veterinary Immunology & Immunopathology*. 143 : 143-147.
- [4] Worrall EE, Sudarisman, Priadi A. 2009. Sialivac: An intranasal homologous inactivated split virus vaccine containing bacterial sialidase for the control of avian influenza in poultry. *Vaccine*. 27: 4161-4168.