Preliminary Report

The Efficacy of Path-Away Disinfectant Against Swine Influenza Virus (H1N1)

Veterinary Research Institute, Department of Veterinary Services Department of

Veterinary Services

Introduction

Path-Away is a plant-based solution developed for purpose of cleansing, sanitizing and deodorizing room ambient. The product was also reported to have ability of killing a wide range of fungi, spores and bacteria that are harmful to human. This communication reports the preliminary efficacy test result of Path-Away against Swine Influenza Virus (H1N1).

Materials and Methods

Preparation of virus pool

The virus H1N1 strain was propagated in Chicken Embryonated Eggs (CEEs) and the virus titer was determined by Hemaaglunitination test (HA).

Disinfectant

The disinfectant namely Path-AwayTM was used in this experiment. Two different dilutions of disinfectant were used in this study. The concentrated stock was diluted to concentration of 2% while the solution from the small personal size bottle which is formulated at 5% was used directly in the study.

A 'carrier' dilution method

A "carrier" is used according to standard protocols for evaluating the effect of surface disinfectant (Sattar et al., 2001). Two (2) pieces of sterile stainless steel penicylinders (type 304 stainless steel) as a carrier was immersed in 7 ml of H1N1 virus pool suspension for 15 minutes. Then, the carriers (rings) were then blot dried and left in incubator for 30 minutes at 37°C (Mermert, Western Germany). Each ring was immersed in 3 ml of disinfectants for 5 minutes. One control carrier was immersed in 1 ml PBS, pH 7.2. Then, 1 ml of diluent (PBS) was added to each ring to elute the virus. These elutes were further assayed for subsequent qualitative analysis by inoculating it into 5 of CEEs.

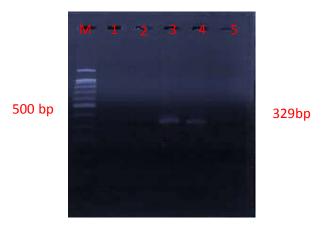
Extraction of virus and Reverse Transcriptase- Polymerase Chain Reaction (RT-PCR)

The viral RNA was extracted from infected allantoic fluid using TRIzol reagent (Invitrogen) as described by the manufacturers. One step RT-PCR AccessQuick RT-PCR Kit (Promega, USA) was employed to amplify the viral RNA using primer NP1529R/ NP1200F (M.-S. Lee et al., 2001).

Results

No amplicon was detected from Path-Away TM treated (2% and 5%) allantoic fluid. However, all the positive control shown amplicon sized at 329 bp. Negative control was also produced no amplicon. The results are illustrated in Figure 1.

Figure 1: RT-PCR product using Primer NP1529R/NP1200F (M.-S. Lee et al., 2001)



Legend: M: Marker, 100bp; 1: Path-AwayTM Treated at concentration of 2%; 2: Path-AwayTM Treated at 5% concentration; 3: Positive control (Not Treated with Path-AwayTM); 4: Positive Control; 5: Negative Control

Discussion

Absence of amplicon indicated that the Swine Influenza Virus H1N1 virus was completely destroyed by the Path-AwayTM. This indicate that Path-AwayTM posed virucidal property. However, according to the Environmental Protection Agency (EPA) and Canada General Standard Board (CGSB) guidelines for the evaluation of disinfectant, there is a requirement to conduct virus infectivity assay (Arshad et al., 2007). As such, further work needs to be conducted.

Conclusion

In conclusion, the Path-AwayTM disinfectant tested in this study can be considered as an effective agent against Swine Influenza Virus (H1N1), as reflected by the absence of amplicon in Path-AwayTM treated allantoic fluid.

References

- 1. Alexander, D.J (2001). Orthomyxoviridae. In: Jordan, F.T.W., Pattison, M. (Eds), Poultry Diseases, 5th ed. W.B. Saunders, London, pp.281-290.
- Canadian General Standard Board (CGSB). Assessments of the efficacy of antimicrobial agents for use on environmental surfaces and medical devices. National Standard of Canada CAN/CGSB-2.161.97. Ottawa: Canada General Standards Board, 1997.

- 3. Chen, J.H.S. (1991). Methods of testing virucides. In: Disinfections, Sterilization and Preservation. Seymour S.Block. 4th ed., ed. Lea & Febiger, Philadelphia. pp. 1076-1093.
- 4. Sattar, S.S., and Springthrope, V.S. (2001). Methods of testing the virucides activity of chemicals. In: Disinfections, Sterilization and Preservation. 5th ed. S.S. Block, ed. Lippinncott Williams & Wilkins, Philadelphia. pp. 1391-1412.
- 5. Sattar, S.A, and Springthrope, V.S. (1988). Test protocols for virucides: need for improvements. Presented at: The 4th International Conference on Progress in Chemical Disinfection. State University of New York, Binghamton, NY: April (1988): 369-386.
- Arshad, S.S, Elmi Salina, A.M, Khairul Anuar, M., Tan, S.W, Amlizawaty, A., Mohd Kamarudin, A.I, Siti Khatijah, M., Omar, A.R (2007). Efficacy of Disinfectants Against Avian Influenza Virus. Presented at: The 19th Veterinary Association Malaysia Congress. 3-5 August 2007: 47-49.
- 7. M.-S Lee et al. Identification and Subtyping of Avian Influenza Viruses by reverse trancription-PCR. Journal of Virological Methods <u>Volume 97, Issues 1-2</u>, September 2001: 13-22.