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Identification of Avian Influenza Virus A/H5 Clade 2.3.2.1 in Asymptomatic-Ducks (Anas species) at a Live-Poultry Market in East Java, Indonesia

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Abstract—A total of 120 cloacal swab samples were collected from asymptomatic-ducks traded at a live-bird market in East Java-Indonesia during January to February 2017. After virus isolation using 10-days-old embryonated chicken eggs, hemagglutination activity was tested. TaqMan real-time reverse transcription (RT) polymerase chain reaction (PCR) assay was performed employing primer sets to differentiate HA genes of H5 clade 2.1.3 and clade 2.3.2.1. Our result show that Avian influenza virus (AIV)-A/H5 clade 2.3.2.1 was currently prevalent among ducks in a live-poultry

market (LPM), indicating LPM could be an important place as an entry point of avian viruses to human resulting novel reassortant strain.

Keywords—ducks; live-poultry market; AI-H5 clade 2.3.2.1, carrier, East Java Province-Indonesia



I. INTRODUCTION

The diversity of avian influenza virus (AIV) circulation in waterfowl reinforces the assumption that waterfowls have an important role in the spread of AIV to humans. Wild *Anseriformes*, are the most heterogeneous reservoirs and host of the influenza A virus. The A/H5N1 virus circulates in Indonesia since 2003 is clade 2.1.3 (Indonesian lineage), but by the end of 2012 many of death cases in ducks and waterfowl were found, it was assumed that it was caused by the A/H5N1 virus new clade 2.3.2.1 (Eurasian lineage). The purpose of this study was to determine the avian influenza virus-A/H5 that currently circulates in ducks traded at a live-bird market in East Java, Indonesia.

II. MATERIALS AND METHODS

A total of 120 cloacal swab samples were collected from asymptomatic ducks during January to February 2017 at a live-bird market in East Java, Indonesia. Virus isolation were carried out by inoculating the swab samples into 10-days-old embryonated chicken eggs, and followed by hemagglutination assay. To detect the viral genomes, TaqMan real-time reverse transcription (RT) polymerase chain reaction (PCR) assay was performed employing primer sets that differentially detect A/H5 HA genes of clade 2.1.3 and clade 2.3.2.1. In order to confirm HA clades, hemagglutination inhibition (HI) test was conducted using two different anti-HA sera specific for the A/H5 clade 2.1.3 and clade 2.3.2.1 viruses.

III. RESULTS

The results showed that 29 (24%) samples were positive for hemagglutination activity and 6 (5%) of them were positive for the HA gene of clade 2.3.2.1 by RT-PCR. The HI tests indicated that these 6 isolates were A/H5 clade 2.3.2.1 viruses. Clade 2.1.3 virus was not detected by both RT-PCR and HI tests.

IV. CONCLUSION

The present study revealed that AIV-A/H5 clade 2.3.2.1 was currently prevalent among ducks in a live-poultry market where could be an important place as an entry point of avian viruses to human resulting novel reassortant strain.

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REFERENCES

- [1] Ducatez Mariette, S. Sonnberg, J. C. Crumpton, A. Rubrum, P. Phommachanh, B. Douangngeun, M. Peiris, Y. Guan, R. G. Webster, R. Webby. 2017. Highly pathogenic avian influenza H5N1 clade 2.3.2.1 and clade 2.3.4 viruses do not induce a clade-specific phenotype in mallard ducks. Microbiology Society.
- [2] Dharmayanti N.L.P.I., A. Ratnawati, D.A. Hewajuli and R. Indriani. 2011. Genetic characterization of H5N1 avian influenza viruses isolated from pet bird and chickens from live bird market in Bali and

- Bekasi (Indonesia). Indonesian Research Center for Veterinary Science. Vol. 8(3), pp. 244-251.
- [3] Dharmayanti N.L.P.I., R. Hartawan, Pudjiatmoko, H. Wibawa, Hardiman, A. Balish, R. Donis, C.T. Davis, and G. Samaan. 2014. Genetic Characterization of Clade 2.3.2.1 Avian Influenza A(H5N1) Viruses, Indonesia, 2012. Emerging Infectious Diseases www.cdc.gov/eid. Vol. 20, No. 4, April 2014.
- [4] Damayanti R., A.Wiyono, H. Nuradji and M. I. Cahyono. 2016. The pathogenecity of H5N1 highly pathogenic Avian Influenza (HPAI) virus clade 2.3.2. in Indonesian indigenous chicken by contact tranmission with infected duck. Indonesian Research Center for Veterinary Science, Bogor 16114 – Indonesia. J. Indonesian Trop. Anim. Agric. 42(2):72-80, June 2017.
- [5] Food and Agriculture Organization. 2008. FAO Animal Production and Health Paper: Biosecurity For Highly Pathogenic Avian Influenza, Issues and Options. FAO United Nations, Rome 2008.
- [6] Food and Agriculture Organization. 2015. FAO Animal Production and Health Guideliness: Biosecurity Guide For Live Poultry Markets. FAO United Nations, Rome 2015.
- HartawanRiszadan N.L.P.I. Dharmayanti. 2014. Sirkulasi Virus Avian Influenza Subtipe H5N1 Di PasarTradisional Di JawaTimurTahun 2012. BalaiBesarPenelitianVeteriner. BeritaBiologi 13(1).
- [8] HaryoAlbiruni. 2015. Kasus Blue Eyes Syndrome padaPeternakanItik di DesaModopuro, KecamatanMojosari, KabupatenMojokerto, JawaTimur. JSV 33 (1), Juli 2015
- [9] Irianingsih S.H., H. Wibawa, W.B. Prijono, R.A. Rahayu, Wdwianingsih. 2014. Avian Influenza H5 Clade 2.3.2 PenyebabKasusKematianItik Di AjibarangdanPekuncen, KabupatenBanyumasPadaBulanOktober BalaiBesarVeterinerWates. International Standard Serial Number (ISSN): 0863-7968 Vol. 14 Nomer 4 Tahun 2014.
- [10] Kirunda H., H. Kibuuka, A. Byaruhanga, E. Mworozi, J. Bwogi, L. Luswa, M. Millard, F. Wabwiren-Mangen and D.K. Byarugaba. 2014. Poor biosecurity in live bird markets in Uganda: A potential risk for highly pathogenic avian influenza disease outbreak in poultry and spread to humans. International Journal of Public Health and Epidemiology ISSN 2326-7291 Vol. 3 (9), pp. 067-074, September, 2014.
- [11] Molesti E., A. Milani, C. Terregino, G. Cattoli and N.J. Temperton. 2013. Comparative Serological Assays for the Study of H5 and H7 Avian Influenza Viruses. Influenza Research and Treatment Volume 2013 (2013), Article ID 286158.
- [12] OIE. 2012. OIE Terrestrial Manual. Avian Influenza Chapter 2.3.4.
- [13] Trock, S.C., M. Gaeta, A. Gonzalez, J.C. Pederson, and D.A. Senne. 2008. Evaluation of Routine Depopulation, Cleaning, and Disinfection Procedures in the Live Bird Markets, New York. Avian Diseases Mar 2008: Vol. 52, Issue 1, pg(s) 160-162.
- [14] Webster, R.G. 2004. Wet markets A continuing source of severe acute respiratory syndrome and influenza? Lancet 363 (9404): 234 – 236.
- [15] Webster, R.G., W.J. Bean, O.T. Gorman, T.M. Chambers and Y. Kawaoka. 1992. Evolution and Ecology of Influenza A Viruses. Microbiol. Rev. 56: 152 - 179
- [16] World Health Organization. 2016. Cumulative number of confirmed human cases of avian influenza A/(H5N1) reported to WHO. http://www.who.int/influenza/human_animal_interface/EN_GIP_20 161219CumulativeNumberH5N1cases.pdf?va=1
- [17] Wibawa H., W.B Prijono, N.L.P.I. Dharmayanti, S.H. Irianingsih, Y. Miswati, A. Rohmah, E. Andesyha, Romlah, R.S.D. Daulay, dan K. Safitria. 2012. InvestigasiWabahPenyakitPadaItik Di Jawa Tengah, Yogyakarta, danJawaTimur:IdentifikasiSebuah Clade Baru Virus Avian Influenza Subtipe H5N1 Di Indonesia 2012. BalaiBesarVeterinerWates Jogjakarta. International Standard Serial Number (ISSN): 0863-7968.
- [18] Zhou X., Y. Li, Y. Wang, J. Edwards, F. Guo, A.C.A. Clements, B. Huang and R.J.S. Magalhaes 2015. The role of live poultry movement and live bird market biosecurity in the epidemiology of influenza A (H7N9): A cross-sectional observational study in four eastern China provinces. Journal of Infection (2015) 71, 470-479.