

Supplementary Fig. S1. Map of the provinces of northern Vietnam. The H5N1 influenza viruses analysed in this study were isolated from the provinces shown in grey. The numbers indicate the following provinces: 1, Tuyen Quang; 2, Son La; 3, Phu Tho; 4, Vinh Phuc; 5, Ha Tay; 6, Ha Noi; 7, Bac Ninh; 8, Bac Giang; 9, Hai Duong; 10, Thai Binh; 11, Ha Nam; 12, Nam Dinh; 13, Ninh Binh; 14, Thanh Hoa.

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Supplementary Fig. S2. Phylogenetic relationships of the (a) PB2, (b) PB1, (c) PA, (d) NP, (e) NA, (f) M and (g) NS genes of H5N1 influenza viruses isolated from patients in Vietnam. Numbers above and below the branch nodes indicate neighbour-joining bootstrap values. Analyses were based on nt 1026–2199, 43–1256, 772–2163, 106–924, 63–1367, 74–782, and 83–745 for the PB2, PB1, PA, NP, NA, M and NS genes, respectively. All trees were rooted to A/goose/Guangdong/1/96. Viruses analysed in this study are shown in red (clade 1) and blue (clade 2.3.4). Scale bar, 0.01 nucleotide substitutions per site. Abbreviations: Ck, chicken; Dk, duck; Qa, quail; Tk, turkey; BHG, barheaded goose; Gs, goose; MDk, Muscovy duck; WSw, whooper swan; HC, house crow; TS, tree sparrow.





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Supplementary Fig. S3. Polymerase activity of UT3040 and UT3040II. 293 cells were co-transfected with protein expression plasmids for PB2, PB1, PA and NP from UT3040 and UT3040II and a plasmid for the expression of an influenza viral minigenome encoding firefly luciferase. Twenty-four hours later, the transfected cells were subjected to the dual-luciferase assay. Relative firefly luciferase activity, normalized to he *Renilla* luciferase activity, is shown. Error bars indicate SD of triplicate experiments.

Supplementary Table S1. Amino acid differences between UT3040 and UT3040II viruses

Protein	Amino acid	Amino acid in:		
	position	UT3040	UT3040II	Other 20 strains
PB1	536	Ν	S	Ν
PA	142	Е	K	K*
NP	189	М	Ι	М

*UT3028II and HN31432M possess E at this position.

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