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Highly Pathogenic Avian Influenza A(H5N1) Virus Infection in Cats, South Korea, 2023

Appendix.

Appendix Table 1. Gross lesions in various tissues of four cats diagnosed with highly pathogenic avian influenza, South Korea

Lesions	Y- Cat No.1*	Y-Cat No.2*	Y-Cat No.3*	G-Cat No.1*
Body Condition Score	7	7	7	5
Jaundice	none	mild	moderate	severe
Congestion	mild	mild	mild	mild
Edema, Congestion, hemorrhage	severe	severe	severe	severe
Pericardial effusion, hemorrhage	moderate	moderate	moderate	moderate
Enlarged, hemorrhage	none	severe	moderate	moderate
Necrotic foci	severe	severe	severe	severe
Hemorrhage, bloody contents	none	None	None	severe
	Body Condition Score Jaundice Congestion Edema, Congestion, hemorrhage Pericardial effusion, hemorrhage Enlarged, hemorrhage Necrotic foci Hemorrhage, bloody contents	Body Condition Score 7 Jaundice none Congestion mild Edema, Congestion, hemorrhage severe Pericardial effusion, hemorrhage moderate Enlarged, hemorrhage none Necrotic foci severe Hemorrhage, bloody contents none	Body Condition Score 7 7 Jaundice none mild Congestion mild mild Edema, Congestion, hemorrhage severe severe Pericardial effusion, hemorrhage moderate moderate Enlarged, hemorrhage none severe Necrotic foci severe severe Hemorrhage, bloody contents none None	Body Condition Score 7 7 7 Jaundice none mild moderate Congestion mild mild mild Edema, Congestion, hemorrhage severe severe severe Pericardial effusion, hemorrhage moderate moderate moderate Enlarged, hemorrhage none severe moderate Necrotic foci severe severe severe

*Y-Cat No.1, Cat carcass No.1 in Shelter 1; Y-Cat No.2, Cat carcass No.2 1 in Shelter 1; Y-Cat No.3, Cat carcass No.3 in Shelter 1; G-Cat No.1, Cat carcass No.1 in Shelter 2.

Appendix Table 2. Pathologic lesions and reactivity to anti-influenza viral nucleoprotein in various tissues of four cats diagnosed with high pathogenicity avian influenza, South Korea, 2023*

	Hematoxylin and eosin stain				Immunohistochemical stain						
		Y-Cat Y-Cat G-Cat						G-Cat			
Tissue	Lesions	No.1	No.2	Y-Cat No.3	No.1	Cells	Y-Cat No.1	Y-Cat No.2	Y-Cat No.3	No.1	
Brain	Neuronal Necrosis	none	none	none	Moderate	Neurons	none	none	none	Moderate	
	Meningoencephalitis	none	none	none	Moderate	Ependymal cells	none	none	none	Moderate	
	Gliosis	none	none	none	Moderate	Glial cells	none	none	none	Moderate	
Lung	Interstitial pneumonia	Severe	Severe	Severe	Moderate	Macrophages	Severe	vere Severe	Severe	Moderate	
	Pneumocytic necrosis	Severe	Severe	Severe	Moderate	Bronchial epithelial cells	Severe	Severe	Severe	Moderate	
Heart	Myocytic necrosis	Mild	Mild	Mild	Mild	Myocytes	Mild	Mild	Mild	Mild	
Spleen	Lymphocytic necrosis Lymphocytic depletion	Mild Mild	Moderate Moderate	Mild Mild	Mild Mild	Macrophages & necrotic debris	Mild	Mild	Mild	Mild	
Liver	Hepatic necrosis	Severe	Severe	Severe	Severe	Kupffer cells and necrotic debris	Severe	Severe	Severe	Severe	

Appendix Table 3. Molecular detection of avian influenza in environmental samples from the cat shelters at Shelter 1 and Shelter 2

Workin	g zone	Sampling site	Number of samples	M-positive samples	H5-positive samples	
Shelter 1	Inside	Cage	8	0	0	
			2	1	0	
		Feces of cats	3	0	0	
		Floor, door, wall	40	2	0	
		Refrigerator	2	1	0	
		Vehicle	2	0	0	
		Dried food	11	0	0	
	Outside	Feathers of wild	4	0	0	
		birds				
		Feces of wild birds	2	0	0	
Shelter 2	Inside	Cage	18	18 0		
			10	0	0	
			12	2	0	
			28	0	0	
		Raw meat food	22	0	0	
		(chicken meat)				
	Raw meat fo		4	4	4	
		(duck meat)				
		Floor, door, wall	12	0	0	
		Human shoes or	10	0	0	
		clothes				
		Vacuum cleaner	10	1	0	
		Window	4	0	0	
	Outside	Vehicle	2	0	0	
		Feathers of wild	10	0	0	
		birds				
		Feces of wild birds	4	0	0	
	Total		183	11	4	

Appendix Table 4. Nucleotide identity versus influenza A/environment/Korea/AM305E-13/2023(H5N1) isolated from duck meat cat food

cat 100	a									
No.	Strain	Origin	PB2	PB1	PA	HA	NP	NA	М	NS
1	A/duck/Korea/H537/2022(H5N1)	Korean Poultry	99.77	99.6	99.58	99.71	99.53	99.79	99.9	100
2	A/feline/Korea/M302– 5/2023(H5N1)	Cat carcass in Shelter 1	99.95	99.91	100	99.94	100	100	100	100
3	A/feline/Korea/M302– 6/2023(H5N1)	Cat carcass in Shelter 1	99.91	99.91	100	99.94	100	99.93	100	100
4	A/feline/Korea/M302– 7/2023(H5N1)	Cat carcass in Shelter 1	99.95	99.91	99.95	100	100	99.93	100	100
5	A/feline/South_Korea/SNU- 01/2023(H5N1)	Cat carcass in Shelter 1	99.95	-	-	100	-	99.93	100	100
6	A/feline/South_Korea/SNU- 02/2023(H5N1)	Cat carcass in Shelter 1	99.91	-	-	100	-	99.93	100	100
7	A/feline/Korea/M305– 3/2023(H5N1)	Cat in Shelter 2	99.95	100	99.95	99.88	99.93	100	100	100
8	A/feline/Korea/M305– 4/2023(H5N1)	Cat in Shelter 2	99.95	100	100	100	100	100	100	100
9	A/feline/Korea/M305– 7/2023(H5N1)	Cat from Shelter 2	99.95	100	100	99.94	99.93	100	100	100
10	A/feline/Korea/M305– 11/2023(H5N1)	Cat carcass from Shelter 2	99.95	100	100	99.94	99.93	100	100	100
11	A/environment/Korea/M305E2– 24/2023(H5N1)	Raw duck meat food manufactured on 26th May	100	100	100	100	100	100	100	100
12	A/environment/Korea/M305E2– 25/2023(H5N1)	Raw duck meat food manufactured on 15th June	100	100	100	100	100	100	100	100
13	A/environment/Korea/M305E3– 1/2023(H5N1)	Raw duck meat food manufactured on 27th July	100	100	100	100	100	100	100	100

Appendix Table 5. Additional amino acid differences identified among influenza A (H5N1) virus detected in Korean domestic ducks, cats, and duck meat-based cat food, Korea*

Virus strain		PB2		PB1				PA	HA	HA
Name	Origin	I3M	P104T	K52R	G211R	Q460K	E752G	V669I	D99N	S519G
A/duck/Korea/H537/2022(H5N1)	Korean Poultry	I	Р	K	G	Q	Е	V	D	S
A/feline/Korea/M302-5/2023(H5N1)		М	Р	R	R	Q	Е	V	D	S

Virus strain		F	PB2 PB1				PA	HA	HA	
Name	Origin	I3M	P104T	K52R	G211R	Q460K	E752G	V669I	D99N	S519G
A/feline/Korea/M302-6/2023(H5N1)	Cat carcass in	ı	Т	K	R	Q	G	V	D	S
A/feline/Korea/M302-7/2023(H5N1)	shelter 1	М	Р	K	R	K	Е	V	D	S
A/feline/South_Korea/SNU- 01/2023(H5N1)		N/A	Р	K	G	Q	E	N.A.	D	S
A/feline/South_Korea/SNU- 02/2023(H5N1)		N/A	Р	K	G	Q	E	N.A.	D	S
A/feline/Korea/M305-3/2023(H5N1)	Cats in shelter	ı	Р	K	G	Q	Е	- 1	N	G
A/feline/Korea/M305-4/2023(H5N1)	2	I	Р	K	G	Q	E	V	D	S
A/feline/Korea/M305-7/2023(H5N1)		I	Р	K	G	Q	E	V	N	S
A/feline/Korea/M305–11/2023(H5N1	Cat carcass from shelter 2	I	Р	K	G	Q	E	V	N	S
A/environment/Korea/M305E2– 24/2023(H5N1)	Raw duck meat food manufactured on 26th May	I	Ρ	К	G	Q	E	V	D	S
A/environment/Korea/M305E2– 25/2023(H5N1)	Raw duck meat food manufactured on 15th June	I	Р	K	G	Q	E	V	D	S
A/environment/Korea/M305E- 13/2023(H5N1)	Raw duck meat food manufactured on 6th July	I	Р	K	G	Q	E	V	D	S
A/environment/Korea/M305E3– 1/2023(H5N1) *N/A, not available	Raw duck meat food manufactured on 27th July	I	Р	К	G	Q	E	V	D	S