

**Supplemental Information: Section 1: Detailed ORF Coordinates Data for Individual PCR Loci.**

Table S1: Details of Sequenced African Elephant EEHV Gene Coding Regions:

Virus Name Gene/ORF	HCMV ORF	HSV ORF	Ori- ent	Protein Name	Position No	Gene Size	Protein Size	Genbank Acc#	Status	Kimba Coordinates	Diff %
<u>1. EEHV2 [AfLNg#54]: Sixteen segments, gB, POL, U33, RRA-gO, gH-PAC2, vGPCR1, MCP, TERex3, TERex1,2, EXO-OBP, OBP, POR-HEL, HEL-ORF-M, ORF-M-ORF-O, ORF-K and ORF-L.</u>											
					Total = 29,699		Differences = 8,773-bp		= 29.5%		
U39	UL55	UL29	F	gB	00001-01503 (1503)	(501)	JQ300034	Core	073932-075446	26	
U38	UL54	UL30	F	POL	00001-02313 (2313)	(771)	JQ300035	Core	076511-078856	27	
U33	UL49	Nil	F	Cys-rich	00001-00851 (851)	(284)	JQ300036	β/γ	083537-084432	29	
U28	UL45	UL39	F	RRA	00001-01119 (1119)	(370)	JQ300037	Core	098645-103256	32	
U27.5(ORF-H)	Nil	UL40	F	RRB	01138-02043	906	302	α/γ			
U27(ORF-I)	Nil	Nil	F	PPF	02281-03522	1242	413	Core			
U45.7(ORF-J)	Nil	Nil	F	-	03611-04129	518	173	Novel			
U46	UL73	UL49A	F	gN	04107-04388	282	93	Core			
U47(ORF-D)	UL74	Nil	R	gO	04423-04601 (178)	(59)		β			
U48	UL75	UL22	R	gH	00501-00001 (401)	(194)	JQ300038	Core	105411-108186	28	
U48.5(ORF-E)	Nil	UL23	R	TK	01541-00498	1043	348	α/γ			
U49	UL76	UL24	F		01543-02241	699	232	Core			
U50	UL77	UL25	F	PAC2	02060-02837 (777)	(292)		Core			
U51	UL78	Nil	F	vGPCR1	00001-01049 (1049)	(350)	KC854747	β	109202-110242	31	
U57	UL86	UL19	R	MCP	02767-00001 (2767)	(922)	JQ300039	Core	115578-118288	23	
U60ex3	UL89ex2	UL15ex2	R	TERex3	00576-00001 (576)	(192)	JQ300040	Core	123609-124421	25	
U62	UL91	Nil	F		00598-00816 (220)	(73)		β/γ			
-					00001-00061 (61)	-	JQ300041	-	127304-128190	22	
U66ex2	Nil	Nil	R	TERex2	00174-00022 (149)	(50)		-			
U66ex1	UL89ex1	UL15ex1	R	TERex1	00915-00277 (639)	(213)		-			
U70	UL98	UL12	F	EXO	00001-01036 (1036)	(345)	JQ300042	Core	131988-134629	27	
U71	UL99	UL11	F	myrTeg	00973-01281	309	102	Core			
U72	UL100	UL10	R	gM	02318-01389	930	309	Core			
U73(ORF-G)	Nil	UL09	F	OBP	02353-02684 (332)	(111)		α/β2			
U73(ORF-G)	Nil	UL09	F	OBP	00001-00570 (570)	(190)	JQ300043	α/β2	134851-135420	22	
U76	UL104	UL06	R	POR	01290-00001 (1290)	(430)	JQ300044	Core	140006-141866	21	
U77	UL105	UL05	F	HEL	01244-01863 (620)	(207)		Core			
U77	UL105	UL05	F	HEL	00001-00039 (39)	(13)	KC854730	Core	143627-144721	26	
U77.5(ORF-M)	Nil	Nil	F	Nuclear	00356-01082 (726)	(232)		Novel			
U77.5(ORF-M)	Nil	Nil	F	Nuclear	00001-00536 (536)	(178)	KC854731	Novel	144958-147958	36	
U80.5(ORF-N)	Nil	Nil	R	vCXCL?	00964-00641	323	107	Novel			
U81	UL114	UL02	R	UDG	01941-00991	950	316	Core			
U82	UL115	UL01	R	gL	02746-01907	840	280	Core			
U82.5(ORF-O)	Nil	Nil	R	S/TGlyP	02928-02670 (258)	(86)		Novel			
U85.5(ORF-Kex3)	Nil	Nil	R	SplGlyP	01940-00001 (1940)	(647)	JX011086	Novel	152011-154339	38	
U85.5(ORF-Kex2)	Nil	Nil	R	SplGlyP	02066-02028 (39)	(13)		Novel			
U85.5(ORF-Kex1)	Nil	Nil	R	SplGlyP	02181-02134 (48)	(16)		Novel			
-					00001-00012 (12)	-		-			
U86.5(ORF-L)	Nil	Nil	R	IE-like	00748-00013 (735)	(245)	KC854732	Novel	155029-155769	22	

2. EEHV3A [AflNg47]: Six segments, POL, TERex3, EXO-gM, OBP, HEL & UDG.

Total = 3,707-bp Differences = 1,370-bp = 37%

U38	UL54	UL30	F	POL	00001-00996 (996)	(332)	JQ300045	Core	077783-078775	23
U60ex3	UL89ex2	UL13ex2	R	TERex3	00311-00001 (311)	(104)	JQ300046	Core	123750-124060	21
U70	UL98	UL12	F	EXO	00001-00042 (42)	(14)	JQ300047	Core	132976-133485	52
U71	UL99	UL11	F	myrTeg	00001-00296 (296)	(99)		Core		
U72	UL100	UL10	R	gM	00650-00453 (207)	(69)		Core		
U73(ORF-G)	Nil	UL09	F	OBP	00001-00697 (697)	(232)	JQ300048	α/β2	134673-135249	46
U77	UL105	UL05	F	HEL	00001-00576 (576)	(192)	JQ300049	Core	141286-141861	30
U81	UL114	UL02	R	UDG	00377-00001 (377)	(126)	KT832517	Core	146543-145168	32

3. EEHV3A [AflNg54]: Seven segments, POL, TK, TERex3, EXO-gM, OBP, HEL & UDG.

Total = 4,019-bp Differences = 1,513-bp = 38%

U38	UL54	UL30	F	POL	00001-01001 (1001)	(334)	JQ300050	Core	077783-078790	23
U48.5(ORF-E)	Nil	UL23	R	TK	00425-00001 (425)	(142)	KT832514	α/γ	106300-106718	38
U60ex3	UL89ex2	UL15ex2	R	TERex3	00309-00001 (309)	(103)	JQ300051	Core	123751-124057	21
U70	UL98	UL12	F	Exo	00001-00047 (47)	(14)	JQ300052	Core	132971-133573	50
U71	UL99	UL11	F	myrTeg	00001-00301 (301)	(100)		Core		
U72	UL100	UL10	R	gM	00749-00465 (284)	(93)		Core		
U73(ORF-G)	Nil	UL09	F	OBP	00001-00525 (525)	(172)	JQ300053	α/β2	134842-135249	48
U77	UL105	UL05	F	HEL	00001-00560 (560)	(187)	JQ300054	Core	141285-141844	31
U81	UL114	UL02	R	UDG	00450-00001 (450)	(150)	KT832515	Core	146586-146138	32

4. EEHV6 [NAP42, nodule-D\*]: Fifteen segments, gB-POL, U33, RRA, RRB-gO, gH-PAC2, vGPCR, MCP, TERex3, TERex1,2, EXO-OBP, POR-HEL, HEL-ORF-O, ORF-Kex3(N), ORF-Kex3(C) & ORF-L.

Total = 31,532 Differences = 5,267-bp = 16.7%

U39	UL55	UL29	F	gB	00001-02541 2541	836	JF692766	Core	073959-078890	17
U38	UL54	UL30	F	POL	02563-04906 (2344)	(781)		Core		
U33	UL49	Nil	F	Cys-R	00001-00916 (916)	(305)	JQ300055	β/γ	083537-084442	21
U28	UL45	UL39	F	RRA	00001-00880 (880)	(293)	JQ300056	Core	098630-099507	17
U27.5(ORF-H)	Nil	UL40	F	RRB	00001-00398 (398)	(133)	JQ300057	α/γ	101128-103703	17
U27(ORF-I)	Nil	Nil	F	PPF	00640-01848 1209	302		Core		
U45.7(ORF-J)	Nil	Nil	F	-	01921-02418 498	165		Novel		
U46	UL73	UL49A	F	gN	02402-02692 291	96		Core		
U47(ORF-D)	UL74	Nil	R	gO	03342-02714 (628)	(209)		β		
U48	UL75	UL22	R	gH	00544-00001 (544)	(181)	JQ300058	Core	105359-108081	21
U48.5(ORF-E)	Nil	UL23	R	TK	01559-00477 1083	360		α/γ		
U49	UL76	UL24	F	-	01558-02256 699	232		Core		
U50	UL77	UL25	F	PAC2	02075-02684 (610)	(203)		Core		
U51	UL78	Nil	F	vGPCR1	00001-00831 (831)	(277)	JQ300059	β	109380-110219	22
U57	UL86	UL19	R	MCP	02252-00001 (2252)	(751)	JQ300060	Core	115567-117818	17

U60ex3	UL89ex2	UL15ex2	R	TERex3	00558-00001 (558)	(146)	JQ300061	Core	123626-124493	14
U62	UL91	Nil	F	-	00592-00858	277	92	β/γ		
U63	UL92	Nil	F	-	00806-00875	(20)	(7)	β/γ		
-	-	-	-	-	00001-00033	(33)	-	JQ300062	-	127311-128195
U66ex2	Nil	Nil	R	TERex2	00025-00156	120	30	-		
U66ex1	UL89ex1	UL15ex1	R	TERex1	00238-00881	(644)	(215)	-		
U70	UL98	UL12	F	EXO	00001-00267	(264)	(88)	JQ300063	Core	132751-135415
U71	UL99	UL11	F	myrTeg.	00204-00506	303	101	Core		
U72	UL100	UL10	R	gM	01546-00578	969	323	Core		
U73(ORF-G)	Nil	UL09	F	OBP	01545-02662	(1018)	(339)	α/β2		
U76	UL104	UL06	R	POR	01313-00001	(1313)	(438)	JQ300064	Core	139965-141870
U77	UL105	UL05	F	HEL	01312-01888	(576)	(192)	Core		
U77	UL105	UL05	F	HEL	00001-00021	(21)	(6)	KC854734	Core	143646-147773
U77.5(ORF-M)	Nil	Nil	F	Nuclear	00327-01826	1500	499	Novel		
U80.5(ORF-N)	Nil	Nil	R	vCXCL1?	02253-01957	297	98	Novel		
U81	UL114	UL02	R	UDG	03304-02351	954	317	Core		
U82	UL115	UL01	R	gL	04088-03270	(819)	(272)	Core		
U82.5(ORF-O)	Nil	Nil	R	S/TGlyP	04088-04018	(71)	(23)	Novel		
U85.5(ORF-Kex3)	Nil	Nil	R	SplGlyP(C)	00719-00001	(719)	(240)	KC854736	Novel	152584-153346
U85.5(ORF-Kex3)	Nil	Nil	R	SplGlyP(N)	00656-00001	(656)	(219)	KC854737	Novel	153520-154205
U86.5(ORF-L)	Nil	Nil	R	IE-like	03262-00001	(3262)	(1087)	JX011087	Novel	155435-158615

**5a. EEHV3Av1 [NAP42, nodule B]: Three segments, POL, TERex3, EXO-U71-gM and HEL.**

Total = 2,902-bp Differences = 1,076-bp = 37%

U38	UL54	UL30	F	POL	00001-01284	(1284)	(428)	JQ300065	Core	077760-079041	34
U60ex3	UL89ex2	UL15ex2	R	TERex3	00287-00001	(287)	(76)	JQ300066	Core	123759-124046	21
U70	UL98	UL12	F	EXO	00001-00023	(23)	(8)	JQ300067	Core	132976-133580	51
U71	UL99	UL11	F	myrTeg.	00001-00277	(277)	(92)	Core			
U72	UL100	UL10	R	gM	00731-00440	(291)	(97)	Core			
U77	UL104	UL05	F	HEL	00001-00598	(598)	(199)	JQ300068	Core	141275-141872	30

**5b. EEHV3Av1 [NAP42, nodule-D]: Four segments POL, EXO-U71-gM, OBP and HEL.**

Total = 3,061-bp Differences = 1,224-bp = 40%

U38	UL54	UL30	F	POL	00001-01025	(1025)	(342)	JQ300073	Core	077760-078781	34
U70	UL98	UL12	F	EXO	00001-00023	(23)	(8)	JQ300074	Core	132976-133580	51
U71	UL99	UL11	F	myrTeg.	00001-00277	(278)	(92)	Core			
U72	UL100	UL10	R	gM	00731-00440	(291)	(97)	Core			
U73(ORF-G)	Nil	UL09	F	OBP	00001-00707	(707)	(236)	JQ300076	α/β2	134852-135440	44
U77	UL104	UL05	F	HEL	00001-00598	(598)	(199)	JQ300078	Core	141275-141872	30

**5c. EEHV3Av1 [NAP42, nodule-A]: One segment, TERex3. Total = 287-bp**

U60ex3	UL89ex2	UL15ex2	F	TERex3	00001-00287	(287)	(96)	JQ300079	Core	123759-124046	21
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**5d. EEHV3Av1 [NAP42, lung sample-E]: One segment, POL. Total = 233-bp**

U38	UL54	UL30	F	POL	00001-00233	(233)	(78)	JQ300080	Core	078536-078768	33
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5e. EEHV3Av3 [NAP42, lung sample-H]: One segment, POL. Total = 234-bp

U38	UL54	UL30	F	POL	00001-00234 (234)	(78)	JQ300081	Core	078549-078782	34
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6a. EEHV3Av2 [NAP42, nodule-C]: Three segments, POL, TERex3, EXO-U71-gM and HEL.

Total = 1,994-bp    Differences = 768-bp    = 39%

U38	UL54	UL30	F	POL	00001-00382 (382)	(127)	JQ300069	Core	078404-078782	37
U60ex3	UL89ex2	UL15ex2	R	TERex3	00287-00001 (287)	(76)	JQ300070	Core	123759-124026	21
U70	UL98	UL12	F	EXO	00001-00023 (23)	(8)	JQ300071	Core	132976-133577	52
U71	UL99	UL11	F	myrTeg.	00001-00277 (277)	(92)		Core		
U72	UL100	UL10	R	gM	00727-00439 (286)	(95)		Core		
U77	UL104	UL05	F	HEL	00001-00598 (598)	(213)	JQ300072	Core	141275-141872	30

6b. EEHV3Av2 [NAP42, nodule-D]: Four segments POL, EXO-U71-gM, OBP and HEL.

Total = 2,572-bp    Differences = 983-bp    = 38%

U38	UL54	UL30	F	POL	00001-01025 (1025)	(342)	JQ300073	Core	077760-078781	34
U70	UL98	UL12	F	EXO	00001-00024 (23)	(8)	JQ300075	Core	132976-133577	51
U71	UL99	UL11	F	myrTeg.	00001-00277 (277)	(92)		Core		
U72	UL100	UL10	R	gM	00727-00439 (287)	(95)		Core		
U73(ORF-G)	Nil	UL09	F	OBP	00001-00222 (222)	(74)	JQ300077	α/β2	134853-135076	31
U77	UL104	UL05	F	HEL	00001-00598 (598)	(199)	JQ300078	Core	141275-141872	30

7a. EEHV7 [NAP42, nodule-D]: Seven segments, POL, TK, TERex3, U71-gM, OBP, POR-HEL and UDG.

Total = 4,008-bp    Differences = 1,495-bp    = 37%

U38	UL54	UL30	F	POL	00001-00660 (660)	(220)	JQ300083	Core	078384-079040	34
U48.5(ORF-E)	Nil	UL23	R	TK	00420-00001 (420)	(140)	KT832516	α/δ	106271-106687	38
U60ex3	UL89ex2	UL15ex2	F	TERex3	00330-00001 (330)	(110)	KC854738	Core	123728-124058	22
U71	UL99	UL11	F	MyrTeg	00001-00270 (270)	(90)	JX011088	Core	132826-133524	54
U72	UL100	UL10	R	gM	00698-00463 (235)	(78)		Core		
U73(ORF-G)	Nil	UL09	F	OBP	00001-00623 (623)	(207)	KC854739	α/β2	134672-135230	42
U76	UL104	UL06	R	POR	00341-00001 (341)	(113)	JQ300084	Core	140955-141878	29
U77	UL105	UL05	F	HEL	00292-00924 (632)	(211)		Core		
U81	UL114	UL02	R	UDG	00353-00001 (353)	(118)	KU321582	Core	146123-146476	35

7b. EEHV7 [NAP42, nodule-B]: One segment, POL. Total = 340-bp

U38	UL54	UL30	F	POL	00001-00342 (342)	(114)	JQ300082	Core	078413-078751	35
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8a. EEHV2v1 [NAP42, nodule-B]: One segment, POL. Total = 449-bp

U38	UL54	UL30	F	POL	00001-00449 (449)	(150)	JQ300085	Core	078393-078841	31
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8b. EEHV2v1 [NAP42, nodule-D]: One segment, POL. Total = 464-bp

U38	UL54	UL30	F	POL	00001-00476 (476)	(159)	JQ300086	Core	078383-078858	31
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8c. EEHV2v1 [NAP42, lung tissue sample-H]: One segment, POL. Total = 419-bp

U38	UL54	UL30	F	POL	00001-00419 (419)	(140)	JQ300087	Core	078414-078832	32
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8d. EEHV2v2 [NAP42, lung tissue sample-H]: One segment, POL. Total = 419-bp

U38	UL54	UL30	F	POL	00001-00419 (419)	(140)	JQ300088	Core	078414-078832	32
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9. EEHV2 [SAM7, lung tissue #2]: One segment, OBP. Total = 581-bp

U73(ORF-G)	Nil	UL09	F	OBP	00001-00581 (581)	(197)	KT832496	α/β2	134844-135424	22
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10a. EEHV3A [SAM6, lung tissue #1]: Seven segments, POL, TK, TERex3, U71/gM, OBP, HEL & UDG.

Total = 3,965-bp Differences = 1416-bp 36%

U38	UL54	UL30	F	POL	00001-00389 (389)	(130)	KT832497	Core	078391-078784	36
U48.5	Nil	UL23	R	TK	00406-00001 (406)	(135)	KU147226	αγ	106300-106759	41
U60ex3	UL89ex2	UL15ex2	R	TERex3	00301-00001 (301)	(100)	KT832498	Core	123758-124059	21
U71	UL99	UL11	F	MyrTeg	00001-00238 (238)	(78)	KT832499	Core	132902-133562	53
U72	UL100	UL10	R	gM	00666-00405 (249)	(82)		Core		
U73(ORF-G)	Nil	UL09	F	OBP	00001-00891 (891)	(297)	KT832500	α/β2	134658-135440	42
U76	UL104	UL06	R	POR	00377-00001 (377)	(125)	KT832501	Core	140919-141870	30
U77	UL105	UL05	F	HEL	00376-00952 (576)	(192)		Core		
U81	UL114	UL02	F	UDG	00001-00360 (360)	(120)	KU147227	Core	145115-146475	34

10b. EEHV3A [SAM7, lung tissue #2]: Four segments, POL, TERex3, U71/gM & HEL.

Total = 1,657-bp Differences = 616-bp = 37%

U38	UL54	UL30	F	POL	00001-00389 (389)	(130)	KT832502	Core	078400-078785	35
U60ex3	UL89ex2	UL15ex2	R	TERex2	00315-00001 (315)	(105)	KT832503	Core	123744-124059	21
U71	UL99	UL11	F	MyrTeg	00001-00167 (167)	(55)	KT832504	Core	132940-133475	55
U72	UL100	UL10	R	gM	00512-00324 (188)	(62)		Core		
U77	UL105	UL05	F	HEL	00441-00001 (441)	(147)	KT832505	Core	141312-141756	30

11a. EEHV6 [SAM6, lung tissue #1]: Four segments, POL, TERex3, OBP & HEL.

Total = 2,227-bp Differences = 323-bp = 14.5%

U38	UL54	UL30	F	POL	00001-00468 (468)	(156)	KT832506	Core	078389-078856	20
U60ex3	UL89ex2	UL15ex2	R	TER	00347-00001 (347)	(116)	KT832507	Core	123679-124024	12
U73(ORF-G)	Nil	UL09	F	OBP	00001-00469 (469)	(156)	KT832508	α/β2	134868-135335	16
U76	UL104	UL06	R	POR	00377-00001 (377)	(125)	KT832509	Core	140919-141861	12
U77	UL105	UL05	F	HEL	00376-00943 (567)	(189)		Core		

11b. EEHV6 [SAM7, lung tissue #2]: Three segments, TERex3, OBP, HEL.

Total = 1,186-bp Differences = 163-bp = 13.7%

U60ex3	UL89ex2	UL15ex2	R	TERex3	00237-00001 (237)	(79)	KT832510	Core	123738-123973	11
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U73	Nil	UL09	F	OBP	00001-00458 (458)	(153)	KT832511	$\alpha/\beta 2$	134871-135328	16
U77	UL105	UL06	F	HEL	00491-00001 (491)	(164)	KT832512	Core	141318-141808	13
<u>12. EEHV7 [SAM7, lung tissue #2]: One segment, HEL.</u>									<u>Total = 575-bp.</u>	
U77	UL105	UL05	F	HEL	00001-00581 (581)	(194)	KT832513	Core	141300-141880	28
<u>13. EEHV2 [EP26, spleen tissue]: Two segments, TK &amp; POL.</u>									<u>Total = 1,398-bp. Differences = 373-bp = 27%</u>	
U38	UL54	UL30	F	POL	00001-00489 (489)	(163)	KM282198	Core	078384-078872	28
U48.5/ORF-E	Nil	UL23	R	TK	00738-00001 (738)	(246)	KM282199	$\alpha/\gamma$	106186-107079	26
U49	UL76	UL24	F		00740-00909 (169)	(56)		Core		
<u>14. EEHV3A [EP26, spleen tissue]: Three segments, POL, TERex3, OBP.</u>									<u>Total = 1,121-bp. Differences = 426-bp = 38%</u>	
U38	UL54	UL30	F	POL	00001-00408 (408)	(136)	KM282200	Core	078390-078794	37
U60ex3	UL89ex2	UL15ex2	R	TERex3	00319-00001 (319)	(106)	KM282201	Core	123738-124056	22
U73(ORF-G)	Nil	UL09	F	OBP	00001-00394 (394)	(131)	KM282202	$\alpha/\beta 2$	134994-135361	52
<u>15. EEHV6 [EP26, spleen tissue]: One segment, HEL. Total = 574-bp</u>										
U77	UL105	UL05	F	HEL	00001-00574 (574)	(191)	KM282203	Core	141307-141880	13
<u>16. EEHV3A [NAP27, Hansa, colon tissue]: One segment, UDG Total = 366-bp. Differences = 148-bp</u>										
U81	UL	UL02	R	UDG	00366-00001 (366)	(122)	KT832474	Core	146108-146567	32
<u>17a. EEHV3B [NAP62, whole blood]. Seven segments, POL, TK, TERex3, U71-gM, OBP, HEL &amp; UDG.</u>									<u>Total = 3,088-bp. Differences = 1192-bp = 39%</u>	
U38	UL54	UL30	F	POL	00001-00365 (365)	(122)	KT832467	Core	078390-078749	36
U48.5(ORF-E)	Nil	UL23	R	TK	00315-00001 (315)	(105)	KT832468	$\alpha/\gamma$	106389-106702	55
U60ex3	UL89ex2	UL15ex2	R	TERex3	00323-00001 (323)	(108)	KT832469	Core	123737-124059	23
U71	UL99	UL11	F	MyrTeg	00001-00272 (272)	(91)	KT832470	Core	132857-133516	52
U72	UL100	UL10	R	gM	00674-00454 (221)	(77)		Core		
U73(ORF-G)	Nil	UL09	F	OBP	00001-00472 (472)	(154)	KT832471	$\alpha/\beta 2$	134656-135121	35
U77	UL104	UL05	F	HEL	00001-00572 (572)	(191)	KT832472	Core	141265-141836	30
U81	UL114	UL02	R	UDG	00367-00001 (367)	(122)	KT832473	Core	146108-146474	35
<u>17b. EEHV3B [NAP62, saliva]. Two segments, U71-gM &amp; OBP.</u>									<u>Total = 962-bp. Differences = 450-bp = 47%</u>	
U71	UL99	UL11	F	MyrTeg	00001-00167 (167)	(55)	KT832475	Core	132850-133577	53
U72	UL100	UL10	R	gM	00631-00349 (282)	(93)		Core		
U73(ORF-G)	Nil	UL09	F	OBP	00001-00331 (331)	(110)	KT832476	$\alpha/\beta 2$	134650-134980	35

18. EEHV3B [SAM5, skin nodules]. Five segments, POL, TERex3, U71-gM, OBP & HEL

Total = 2,835-bp Differences = 1078-bp = 38%

U38	UL54	UL30	F	POL	00001-00405 (405)	(135)	KT832491	Core	078392-078793	36
U60ex3	UL89ex2	UL15ex2	R	TERex3	00280-00001 (280)	(93)	KT832492	Core	123758-124037	20
U71	UL99	UL11	F	MyrTeg	00001-00106 (106)	(35)	KT832493	Core	132863-133523	54
U72	UL100	UL10	R	gM	00567-00339 (228)	(76)		Core		
U73(ORF-G)	Nil	UL09	F	OBP	00001-00727 (727)	(242)	KT832494	$\alpha/\beta$	134627-135240	43
U76	UL103	UL06	R	POR	00376-00001 (376)	(125)	KT832495	Core	140919-141779	30
U77	UL104	UL05	F	HEL	00377-00856 (479)	(156)		Core		

(\*) Identical EEHV6 sequence results were obtained at many of these same genomic loci from nodules -B and -C as well as at several loci from nodule-A (data not shown).

( ) = Incomplete ORF or exon only.

The status column refers to whether the gene is common to all mammalian herpesvirus sub-families = Core or just to the designated subset of other herpesvirus sub-families;  $\alpha/\beta$  = Alpha & Beta only;  $\beta/\gamma$  = Beta & Gamma only;  $\beta$  = Beta only;  $\alpha/\beta 2$  = Alpha & *Roseolovirus* genus only; or unique to the *Proboscivirus* genus = Novel.

The %Diff column shows nucleotide divergence values relative to the matching EEHV1(Kimba)locus.



## **Supplemental Information: S2. Listing of PCR Primers Used at Selected PCR Loci:**

Diagnostic two-round redundant PCR primers for the PAN EEHV U38(POL) Codehops region (LGH6710/6711/6712) as well as more sensitive three-round primer sets specific for the EEHV1, EEHV2, EEHV3, EEHV4, EEHV5 and EEHV6 versions of the 520-bp POL and 340-bp TER loci have all been described elsewhere(1-3). Similarly, standard basic consensus primer sets for the EEHV1, EEHV2, EEHV5 and EEHV6 versions of the 679-bp U71-72(gM) and 986-bp U76(POR)-U77(HEL) loci, as well as EEHV1 specific sets for the 910-bp U51(vGPCR1) and 1080-bp U74.5(TK)-U48(gH) loci, were all listed in Stanton et al (4) or in Zachariah et al (5). A second generation high efficiency U60(TERex3) primer set specific for detection of EEHV3 (LGH6707-6708-6727-6728) was also described previously (2). An additional 24 sets of consensus or specific primers (not listed here) for several selected EEHV1A, EEHV1B, EEHV2, EEHV5 and EEHV6 loci were also given in the previous two papers (6, 7). Eleven more of the most relevant selected PCR amplification primer sets used for this paper are listed below.

### 1. EEHV6 Specific U39(gB) (2300-bp):

- R1 LGH3830 5'-CCCTGGTTTTAAAGACTGATAA-3' [1A, 1B, 2 com]
- R2 LGH3828 5'-TGGCCTTATCGTATATGTTCTGG-3' [1A, 1B, 2, 5, 6 com]
- R3 LGH7931 5'-GGACTACCAGAGGCTACAGGAAC-3' [6 only]
- L4 LGH7905 5'-GTTCTGTAGCCTCTGGTAGTC-3' [6 only]
- R4 LGH7930 5'-CCATGTCAATACATCCTATGGAAAGC-3' [6 only]
- L3 LGH7904 5'-GATGCTTTCCATAGGATGTATTGAC-3' [6 only]
- R5 LGH7933 5'-GGTGACTTAAACGTGGCACAGGTTTCAG-3' [6 only]
- R6 LGH7932 5'-GAGTATAGCGGATGCGTGGTGTTCG-3' [6 only]
- L2 LGH7932B 5'-CGACACCACGCATCCGCTATACTC-3' [6 only]

L1 LGH7998B 5'-GGACTTCCACGAGATTGATTCATTC-3' [1B, 6 com]

Set A: First round 1A R1-L3 = 980-bp; second round 2A1 R2-L3 = 790-bp; second round 2A2 = 850-bp; R1-L4; third round 3AB R2-L4 = 650-bp. Set B: First round 1B R3-L2 = 640-bp; second round 2B R4-L2 = 420-bp. Set C: First round 1C; R5-L1 = 930-bp; second round 2C R6-L1 = 860-bp.

2. EEHV6 Specific U38(POL) Codehops plus N-terminal Extended Primers [1,350-bp];

R1 LGH7804B 5'-GCATTTGATATAGAATGTCT[A/T]TCGGG-3' [6 only]

R2 LGH3818B 5'-ACGCA[T/G]CACTTGTTTACATTAGG-3' [1/6 com]

R3 LGH7805B 5'-GGGCATGGAGTCGTTTTTTAACAGATGTC-3' [6 only]

L2 LGH7821B 5'-GAGACCTTATCCATGCACACACGGTAC-3' [6 only]

R4 LGH7828B 5'-TTGACGGACAACARAGCAGAAT-3' [1/6 Com]

R5 LGH7831 5'-GCATTATACAGGCATACAACCTGTG-3' [6 only]

L1 LGH7584B 5'-CATCGATTTTGAAGTTCTCATGGTC-3' [6 only]

First round PCR 1A R3-L1 = 990-bp; second round 2A R4/L1 = 660-bp; third round 3A R5-L1 = 475-bp; and extended first round PCR 1B R1-L1 = 1,350-bp, second round 2B R1-L2 = 450-bp; third round 3B R2-L2 = 340-bp;.

3. Initial Consensus EEHV3, EEHV4, EEHV7 U38(POL) Internal Codehops Effective Primers:

R1 LGH6719 5'-CGTTGAAGGTGTCGCAGAT-3'

L2 LGH6720 5'-ATCCTGGCGCAGCTGCTGAC-3'

L1 LGH7400 5'-CAGCATCATCCAGGCCTACAAC-3'

First round PCR R1-L1 = 330-bp, second round R1-L2 = 220-bp.

4. EEHV3 versus EEHV7 Specific U38(POL) Primers (for sequencing of mixtures only):

R1' LGH6719B3 5'-GCAGATGTAGCTCTTGGTCCTG-3'

R1'' LGH6719B7 5'-GCAGATGTAGCTCTTGGTCCTA-3'

L2'' LGH6720B3 5'-TGGCTGGCCGAGAGGAAGGCCGTC-3

L2' LGH6720B7 5'-TGGCTGGCCGAGAGGAAGGCCGTT-3'

5. Effective Primers for EEHV2 and EEHV7 U38(POL) N-terminal Codehops Extension.

R1 LGH7832 5'-CAAYTCTATNCCYTTTCATNCT-3' [2 and 7]

R2 LGH7833 5'-CCCACGTANCGYTTYTTRCA-3' [2, 6, 7]

L2 LGH7437 5'-GTATCATCAAGCTTATAACC-3' [2 and 7]

L1 LGH7440 5'-GACTTCGCCAGCTTGTATCC-3' [2 and 7]

First round 1A PCR R1-L1 = 670-bp; second round 2A R1-L2 = 640-bp; second round 2B R1-L2 = 630-bp.

6. EEHV7-Specific and GC-rich EEHV Common U38(POL) Primers:

R1 LGH7906B 5'-GCTTCACGAAGACCTTGTCTACC-3' [EEHV7 > 3]

R2 LGH7907 5'-CGAAGACCTTGTCYACCTCCAGC-3' [EEHV7 = 3]

R3 LGH7908 5'-ACGAARACGCTGTCCGTGTCCCCGTA-3' [EEHV7 > 2, 3]

R4 LGH7887B 5'-CGACGTCTACCTGGAAGTTGTCCT-3' [EEHV7 specific]

L2 LGH7925 5'-GCCGTTTCGGGAGAACTCAAGGT-3' [EEHV7 specific]

L1 LGH7924 5'-CTTCGTCAAGGCCACGTACGCACC-3' [EEHV7 > 4]

First round 1A PCR R1-L1 = 520-bp; second round 2A R2-L1 = 510-bp; second round 2B R1-L2 = 450-bp; third round 3A R3-L1 = 470-bp; third round 3B R2-L2 = 440-bp; alternative third round 3C R4-L1 = 320-bp; alternative third round 3D R3-L2 = 300-bp.

7. EEHV7-Specific and GC-rich EEHV Common U48.5(TK) Primers:

R1 LGH9985 5'-CTG TTC CTC TGT CTC ACG CGC CTC AGC GT-3' [EEHV3,4,7 com]

R2 LGH9986 5'-AGC ATG GAG TAC AAC CCG ATG TAG CTG AA-3' [EEHV3,4,7 com]

L7-alt2 LGH11129 5'-GCG ACA CCG TTC CTG GCC AGG GAG GCC-3' [EEHV7-specific]

L7-alt1 LGH11128 5'-CGC TAT ACG ACG AGC CCA TGC TTC-3' [EEHV7-specific]

L2 LGH9987 5'-GTA TAG GTG TAG GTA AGA CGT CAC TGT TC-3' [EEHV3,4,7 com]

L1 LGH9988 5'-CTC ACC GTT TAC CTA GAA GGT TGT ATA GGT G-3' [EEHV3,4,7 com]

First round R1-R4 = 485-bp, second round 2A R1-L2 = 460-bp, second round 2B R2-L1 = 395-bp, third round 3AB R2-L2 = 365-bp. Alternative first round 1A R1-L2 = 460-bp, second round 2 R1-L7-alt1 = 400-bp, third round 3 R1-L7-alt2 = 270-bp.

8. EEHV1 and EEHV6 Specific plus EEHV1,2,5,6 Consensus U57(MCP) Primers:

R1 LGH7980 5'-CARGGCTTTTCTACCATATCTGACG-3' [Common]

R2 LGH8406 5'-GGGAACACCTTRAA[A/C]AGGTCYTGTAACG-3' [Common]

L6 LGH7978 5'-AGCACRGTGGACATGGGCATGGG-3' [Common]

R3 LGH7979 5'-GCGCTGTAACCCATGCCCATGTCCAC-3' [Common]

- R4 LGH8526 5'-CYCTGATGTA[A/C]GGRGCGCATGCYTG-3' [Common]
- R5 LGH8525 5'-CYGGCATGCAGTGTCTCTGAGCTTG-3' [EEHV2, 5]
- L5 LGH7977 5'-GAGAACGCCAGAGT[G/T]CTAGAGATCAC-3' [Common]
- R6 LGH8524 5'-G[C/G]TCGAGCAGCC[G/T]TCTYAACATTAC'3' [EEHV1, 6]
- L4 LGH7976 5'-GAT[A/C]TAYTACTAYTGCATGCTYCC-3' [Common]
- R7 LGH8522 5'-TCGAGRTTTGATACGTCATARTTCT-3' [EEHV1, 2]
- L3 LGH8523 5'-CGGAATTAAAGAACCCGTCTGG-3' [EEHV1, 6]
- L2 LGH8518 5'-CAYCC[G/T]ATGTACGAYTTCAGTGACGTA-3' [EEHV1, 2, 5]
- L1 LGH8517 5'-CAGGACAGATCCAC[C/G]AT[A/C]TCCGTGCA-3' [EEHV1, 2]

Appropriate pairs of primers were chosen from those listed above in order from right to left across this overall 2.7-kb locus located between EEHV1A(Kimba) coordinates 115564 to 118391.

9. EEHV7-Specific U60(TERex3) Primers:

- R1 LGH9380 5'-CGTGCAACACGAGCACGCAAAGTACGTC-3' [EEHV7 only]
- R2 LGH9379 B 5'-CGCCRACCCAGATCCTGAATGTCATCTC-3' [EEHV3, 4, 7 com]
- R3 LGH9378 5'-CGTGTGCGAGGAGCACTTATATAGCTTCGG-3' [EEHV3, 4, 7 >2, 5, 6 com]
- L2 LGH9377 5'-CGAACTCCGTGAYGCTGGCGTCCGTCAGGA-3' [EEHV3, 4, 7 com]
- L1 LGH9376 5'-GAGTTGACGGTGCTGTAGCGGATCATGTGCG-3' [EEHV3, 7 com]

First round PCR R1-L1 = 330-bp; second round A R1-L2 = 300-bp; second round B R2-L1 = 280-bp; third round A R3-L1 = 250-bp; alternative third round B R2-L2 = 250-bp.

10. EEHV7-Specific plus GC-rich EEHV common U71(MyrTeg)-U72(gM) Primers [750-bp block]:

- R1 LGH6793B 5'-CATCGGGA[G/T]CCGGAAAAACGCC-3' [EEHV2, 4-specific]  
R2 LGH9213 5'-GCGTGGGAGGCGCGAGGAGGCCGTGCTG-3' [EEHV7 only]  
R3 LGH9237 5'-CGGGATTGAAAGGCAAGGACGGCAACCC-3' [EEHV7 only]  
L4 LGH9212B 5'-TCRCCATCGTGCGTTGCCTC[A/C]GGATGTTA-3' [EEHV7, 3 > 4]  
L3 LGH9211 5'-GGCAACATCACCGTAATGTACGTGGTTTGG-3' [EEHV7 only]  
L2 LGH9210 5'-CCAGTACGATAAGATCTACCTGGACGA-3' [EEHV3, 4 com]  
L1 LGH6792B 5'-GCATCTTCTTCACCCTCAAGC-3' [EEHV4-specific]

First round PCR R1-L3 = 700-bp; second round R1-L4 = 670-bp; third round R2-L4 = 610-bp. Alternative first round R2-L1 = 680-bp; second round R2-L2 = 650-bp; third round R3-L2 = 600-bp.

11. EEHV7-Specific U73(OBP) Primers [625-bp block]:

- R1 LGH9280 5'-GAC[C/G]GCGGCYATGATCTCGTGGCTCA-3' [EEHV3, 4, 7 com]  
R2 LGH9374 5'-TCTCATCGTGCAGATCGAGAGTCTCTACC-3' [EEHV7 only]  
R3 LGH9375 5'-CCGACTCGAGGGTTCCTACGACGTGTC-3' [EEHV7 only]  
L3 LGH9283 5'-GGCACGAGGCGACGCACGCCAGGAACTTG-3' [EEHV3, 4 com]  
R4 LGH9394 5'-CCTGCTGCT[C/G]ATGGACGCCACCRCTAAC-3' [EEHV7 > 3]  
L2 LGH9285 5'-GTTGTCCGGGAACCGTTCCTGATCTCCTCC-3' [EEHV4 specific]  
L1 LGH9286 5'-GGTTCCTGATCTC[C/G]TCCACGA[C/T]GAAGCAC-3' [EEHV3, 4 com]

LHS segment: First round PCR R2-L1 = 440-bp; second round A R3-L1 = 420-bp; second round B R3-L2 = 400-bp; third round A R4-L1 = 270-bp; third round B R4-L2 = 240-bp. RHS segment: First round PCR R1-L1 = 880-bp; second round R1-L2 = 840-bp; third round R1-L3 = 390-bp.

12. EEHV7-Specific and GC-rich EEHV common U76(POR)-U77(HEL) Primers [850-bp block]:

R1-7 LGH9267 5'-CAGTACTTGATGATGTGCGTCATATCCG-3' [EEHV3, 4, 7 common]

R2-7 LGH7920R 5'-CATG TTCAGGTAGTGCACGGAGAGC-3' [EEHV7 > 4 = 3]

R3-7 LGH7921 5'-CTGAACATGTGTTCCGGCCAGCAAGG-3' [common for all EEHVs]

L4-7 LGH7922B 5'-CAGCAGCATRGAGACGTGGCGAC-3' [EEHV3, 4, 7 common]

L3-7 LGH9246 5'-ACGATGATGTTGTGGCGGCAGATGGGCA-3' [EEHV3, 4, 7 common]

L2-7 LGH8749B 5'-CACGGCGTG[C/G]AGCATGTGCTCCAGG-3' [EEHV3, 4, 7 common]

L1-7 LGH3198 5'-CACAG[A/C]GCGTTGTAGAACC-3' [common for all EEHVs]

First round PCR 1A R1-L2 = 910-bp, second round 2A R1-L3 = 860-bp, third round 3A R1-L4 = 710-bp. Alternative first round PCR 1B R2-L1 = 870-bp; second round 2B R2-L3 = 800-bp; third round 3B R3-L3 = 520-bp. Alternative third round (both sets) 3C R3-L4 = 360-bp.

13. EEHV7-Specific and GC-rich EEHV Common U81(UDG) Primers:

R1 LGH9989 5'-AGG TAT TGG TTG GCT TTC ACG AAG TG-3' [EEHV3,4,7 com]

R2 LGH9990 5'-CTG GCG GCC GCC AGG GGG GAA GGG T GA GC-3' [EEHV3,4,7 com]

L2 LGH9991 5'-CCA CGG CCA CCT CGA CAG AGC TGG-GC3' [EEHV3,4,7 com]

L1 LGH9992 5'-GCC GAC GGC CTG GCC TTC TCC ACG GGT GAC GG-3' [EEHV3,4,7 com]

First round PCR 1A R1-L1 = 430-bp, second round 2A R1-L2 = 335-bp, second round 2B R2-L1 = 380-bp and third round 3AB R2-L2 = 265-bp.

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