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The Genome of Fowlpox Virus

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Here we present the genomic sequence, with analysis, of a pathogenic fowlpox virus (FPV). The 288-kbp FPV genome consists of a central coding region bounded by identical 9.5-kbp inverted terminal repeats and contains 260 open reading frames, of which 101 exhibit similarity to genes of known function. Comparison of the FPV genome with those of other chordopoxviruses (ChPVs) revealed 65 conserved gene homologues, encoding proteins involved in transcription and mRNA biogenesis, nucleotide metabolism, DNA replication and repair, protein processing, and virion structure. Comparison of the FPV genome with those of other ChPVs revealed extensive genome colinearity which is interrupted in FPV by a translocation and a major inversion, the presence of multiple and in some cases large gene families, and novel cellular homologues. Large numbers of cellular homologues together with 10 multigene families largely account for the marked size difference between the FPV genome (260 to 309 kbp) and other known ChPV genomes (178 to 191 kbp). Predicted proteins with putative functions involving immune evasion included eight natural killer cell receptors, four CC chemokines, three G-protein-coupled receptors, two β nerve growth factors, transforming growth factor β, interleukin-18-binding protein, semaphorin, and five serine proteinase inhibitors (serpins). Other potential FPV host range proteins included homologues of those involved in apoptosis (e.g., Bcl-2 protein), cell growth (e.g., epidermal growth factor domain protein), tissue tropism (e.g., ankyrin repeat-containing gene family, N1R/p28 gene family, and a T10 homologue), and avian host range (e.g., a protein present in both fowl adenovirus and Marek's disease virus). The presence of homologues of genes encoding proteins involved in steroid biogenesis (e.g., hydroxysteroid dehydrogenase), antioxidant functions (e.g., glutathione peroxidase), vesicle trafficking (e.g., two α-type soluble NSF attachment proteins), and other, unknown conserved cellular processes (e.g., Hal3 domain protein and *GSN1*/*SUR4*) suggests that significant modification of host cell function occurs upon viral infection. The presence of a cyclobutane pyrimidine dimer photolyase homologue in FPV suggests the presence of a photoreactivation DNA repair pathway. This diverse complement of genes with likely host range functions in FPV suggests significant viral adaptation to the avian host.

Within the *Chordopoxvirinae* subfamily (poxviruses of vertebrates) of the family *Poxviridae*, only members of the *Avipoxvirus* genus infect nonmammalian hosts (118). Avipoxviruses are a large family of cytoplasmic DNA viruses which infect more than 60 species of wild birds representing 20 families (169). Variability in restriction enzyme profiles of viral DNA suggests significant genomic differences among family members (169). Cross-infection studies also suggest genetic differences among viruses, which are reflected as a wide range of pathogenic effects (absence of clinical disease, local pox lesions, local and generalized infection, and generalized infection with death) and a lack of cross protection, depending on the specific virus-host combination (46, 169).

Fowlpox virus (FPV), the prototypical member of the *Avipoxvirus* genus, infects chickens and turkeys. Poxvirus diseases of poultry and other domestic birds (canaries and pigeons) have significant economic impact worldwide, with losses resulting from a drop in egg production in layers, reduced growth rates in broilers, blindness, and in some cases death (46, 170). Two forms of disease are associated with different routes of infection. The most common, the cutaneous form, occurs following infection by biting arthropods that serve as mechanical vectors for viral transmission. The disease is characterized by an inflammatory process with hyperplasia of the epidermis and feather follicles, scab formation, and desquamation of the degenerated epithelium, and it predisposes the host to secondary bacterial infections. The second, or diphtheric, form involves droplet infection of the mucous membranes of the mouth, the pharynx, the larynx, and sometimes the trachea. The prognosis with this form of the disease is poor because lesions often cause death by asphyxiation (169–171).

Vaccination with live-attenuated viruses (FPV and canarypox virus [CaPV]) and nonattenuated viruses (pigeonpox virus) is used to control this disease (59, 77, 136, 182). Fowlpox and pigeonpox vaccines are applied by comb scarification, by the wing-web stick method, or by feather follicle immunization. Vaccination confers protective immunity 10 to 14 days after infection. Problems related to safety and efficacy of commercial FPV vaccines remain (9, 24, 29, 65).

Multivalent recombinant FPV vaccines as well as FPV vaccines which incorporate immune response modifiers have been constructed (28, 96). Recombinant FPV vaccines expressing foreign antigens have been used to immunize animals against other avian and mammalian diseases (26, 83, 112, 121, 124, 125, 187). Because FPV and CaPV undergo abortive replication in mammalian cells, their use as host range-restricted mammalian expression vectors has been suggested (164, 165).

The FPV genome, containing 260 to 309 kbp of double-stranded DNA, is larger than other described chordopoxvirus (ChPV) genomes (45, 115, 120). Past work on FPV genomics, much of which used highly tissue culture-passaged FPV strains, has provided genetic information on approximately one-third of the viral genome, including some viral genes with putative immune evasion and host range functions (16, 18, 20, 57, 93, 127, 150, 163, 166, 191). The rational design of safer and more

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effective FPV vaccines and FPV-based expression vectors will require complete information on viral genes associated with viral virulence and host range and a more complete understanding of how these genes function in viral pathogenesis, immune evasion, and avian host range. Here we report the genomic sequence and analysis of a highly pathogenic strain of FPV.

MATERIALS AND METHODS

FPV DNA isolation, cloning, and sequencing. FPV genomic DNA was extracted from primary chicken embryo fibroblasts infected with a pathogenic FPV strain (fowlpox challenge virus; Animal Health Inspection Service Center for Veterinary Biologics, Ames, Iowa). Random DNA fragments were obtained by incomplete enzymatic digestion with *Tsp*509I endonuclease (New England Biolabs, Beverly, Mass.). DNA fragments of 1.5 to 2.5 kbp were isolated after separation on agarose gels, cloned into the dephosphorylated *Eco*RI site of pUC19 plasmids, and grown in *Escherichia coli* DH10B cells (Gibco BRL, Gaithersburg, Md.). Double-stranded pUC19 plasmids were purified by the alkaline lysis method in accordance with the manufacturer's instruction (5′→3′, Inc., Boulder, Colo.). DNA templates were sequenced from both ends with M13 forward and reverse primers, using dideoxy chain terminator sequencing chemistries (135) and an Applied Biosystems (ABI) PRISM 377 automated DNA sequencer (Perkin-Elmer, Foster City, Calif.). ABI sequence software (version 3.3) was used for lane tracking and trace extraction. Chromatogram traces were base called with Phred (64), which also produced a quality file containing a predicted probability of error at each base position. The sequences were assembled with Phrap (63), with the quality files and default settings being used to produce a consensus sequence, with some subsequent manual editing being performed by the Consed sequence editor (72). An identical sequence was assembled with the TIGR assembler, using quality files and clone length constraints (160). Gap closure was achieved by primer walking of gap-spanning clones and sequencing of PCR products. The final DNA consensus sequence represented on average sixfold redundancy at each base position.

DNA sequence analysis. Genome DNA composition, structure, repeats, and restriction enzyme patterns were analyzed as previously described (1). Open reading frames (ORFs) longer than 30 amino acids with a methionine start codon (155, 156) were evaluated for coding potential by the use of the Hexamer (ftp.sanger.ac.uk/pub/rd) and Glimmer (134) computer programs. Minor ORFs were excluded. Gene families were analyzed and annotated as previously described (1). Early-promoter sequences were predicted as follows. Fifteen-base DNA motifs with similarity to the vaccinia virus (VV) early-promoter consensus sequence (51, 118) were selected from regions located upstream of initiation codons of 30 FPV homologues of VV virus early genes. These motifs were used to generate a scoring matrix (PROFILEMAKE) (55), and this matrix was used to search 100 bases upstream of all FPV ORFs (MOTIFSEARCH) (55). Positive ORFs found by MOTIFSEARCH ($P = 0.001$) were further verified by visual inspection, and those that had substitutions at the most-conserved residues were excluded (14 genes).

Virus abbreviations. Virus names are abbreviated in this article as follows: African swine fever virus, ASFV; *Amsacta moorei* entomopoxvirus, AmEPV; canarypox virus, CaPV; chordopoxvirus, ChPV; cowpox virus, CPV; ectromelia virus, ECT; entomopoxvirus, EPV; fowlpox virus, FPV; *Heliothis armigera* entomopoxvirus, HaEPV; lumpy skin disease virus, LSV; *Lymantria dispar* nuclear polyhedrosis virus, LdNPV; molluscum contagiosum virus, MCV; myxoma virus, MYX; orf virus, OV; *Paramecium bursaria* chlorella virus, PBCV; rabbit fibroma virus, RFV; rabbitpox virus, RPV; reticuloendotheliosis virus, REV; swinepox virus, SPV; tanapoxvirus, TPV; vaccinia virus, VV; and variola virus, VAR.

Nucleotide and protein sequence databases. Accession numbers presented are from the GenBank, SwissProt, or PIR database unless otherwise noted.

Nucleotide sequence accession number. The FPV genome sequence has been deposited in GenBank under accession no. AF198100.

RESULTS AND DISCUSSION

Organization of the FPV genome. The FPV genome was assembled into a contiguous sequence of 288,539 bp, which is slightly smaller in size than previous estimates of 299 to 309 kbp for low-passage-number FPV field strains (45, 115). Because the hairpin loops were not sequenced, the left-most nucleotide of the assembled sequence was arbitrarily designated base 1. The nucleotide composition is 69% A+T and is uniformly distributed over the entire length of the FPV genome. Six small regions (102 to 315 bp in length) with higher C+G content (50%) are located in the terminal genomic regions (nucleotides 3219 to 5618 and 28222 to 285321). The total composition of all FPV ORFs reflects a bias for residues with A- and T-rich codons. Ile, Leu, Lys, Asn, Tyr, and Phe constitute 45% of all encoded amino acids.

FPV encodes 260 putative genes of 60 to 1,949 amino acids in length (Fig. 1; Table 1). Predicted ORFs represent an 85% coding density, with an average ORF length of 943 nucleotides. One hundred and one FPV ORFs have been assigned similarity or putative function based on homologies with other viral or cellular genes. FPV has a genomic organization similar to that of other known ChPVs (71, 108, 141, 142). There is no evidence of introns, both strands are protein encoding, and ORFs frequently occur in head-to-tail tandem arrays (Fig. 1). Fifty-two ORFs partially overlap other ORFs. Within the terminal 50 kbp of the genome, most ORFs (74% of them in these regions) are transcriptionally oriented toward their respective termini. As seen in other poxviruses, the FPV genome contains a central coding region bounded by two identical inverted terminal repeat (ITR) regions of approximately 9.5 kbp each (Fig. 1). The 3′ 148 codons of ORFs FPV010 and FPV251 mark the boundary between the ITR and the central coding region (Fig. 1). The terminal 1,877 nucleotides are noncoding.

The remnant of an integrated avian reticuloendotheliosis virus (REV) genome in the FPV genome is represented by 253 nucleotides (232464 to 232717) that are similar (98% identity) to the long terminal repeat of a chicken B lymphoma-derived REV (accession no. M22223). However, REV *env*, *gag*, and *pol* genes were not found as has been reported for some FPV strains (76). The same long terminal repeat (98% identity over 200 nucleotides) is also found in several strains of Marek's disease virus (MDV), a herpesvirus of chickens. The fragmented remains of a ubiquitin gene are present in the FPV genome from nucleotides 74550 to 74220. Interestingly, the best match to this gene is chicken ubiquitin (accession no. M1110), which exhibits 54% identity over 76 amino acids with one frameshift, two in-frame stops, and two gaps.

ITRs. The FPV genome contains identical ITRs of 9,520 nucleotides at both termini (Fig. 1). Within each ITR, a 1.7-kbp region contains 42 copies of a 31- to 32-bp tandem repeat (70 to 95% identical) between nucleotides 198 and 1835 as well as between nucleotides 286703 to 288340. Sizing of seven cloned fragments spanning this tandem-repeat region produced specific size classes of 1.7, 2.4, 3.3, 5.1, and 5.8 kbp in length, indicative of length polymorphism. Therefore, individual FPV genomes could be at least 8 kbp longer than the genomic sequence assembled here. Each ITR also contains 10 ORFs. These ITR sequence data are consistent with previous descriptions of FPV ITR regions (36, 166).

Gene expression regulatory elements. FPV ORFs contain typical poxvirus promoter sequences upstream of their translation initiation codons. Sequences with similarity to the VV early-promoter consensus sequence (AAAAATGAAAAAA AA) have previously been noted in the 5′ untranslated regions of known and predicted FPV early genes (90, 91, 191). Fifty-six FPV ORFs contain putative early promoters (Table 1). Of these, 22 contain a poxvirus early transcriptional stop sequence (TTTTTXT, where X is any nucleotide) near the translational stop codon (50 bases upstream to 100 bases downstream) and lack the early stop sequence elsewhere in the ORF (189). As seen in other poxviruses, many genes with potential early promoters are members of gene families and/or putative host range genes (Table 1). Three of five homologues of VV intermediate genes (FPV088, FPV126, and FPV165) contain the VV intermediate-promoter sequence ($AAAXAAX_{11-13}TA$ AA) (10, 11, 118), and one (FPV049) contains a single-base substitution (AAAXAG). A total of 55 putative late FPV ORFs, including many of the conserved virion-associated poxvirus genes (Table 1), contain the VV late-promoter sequence

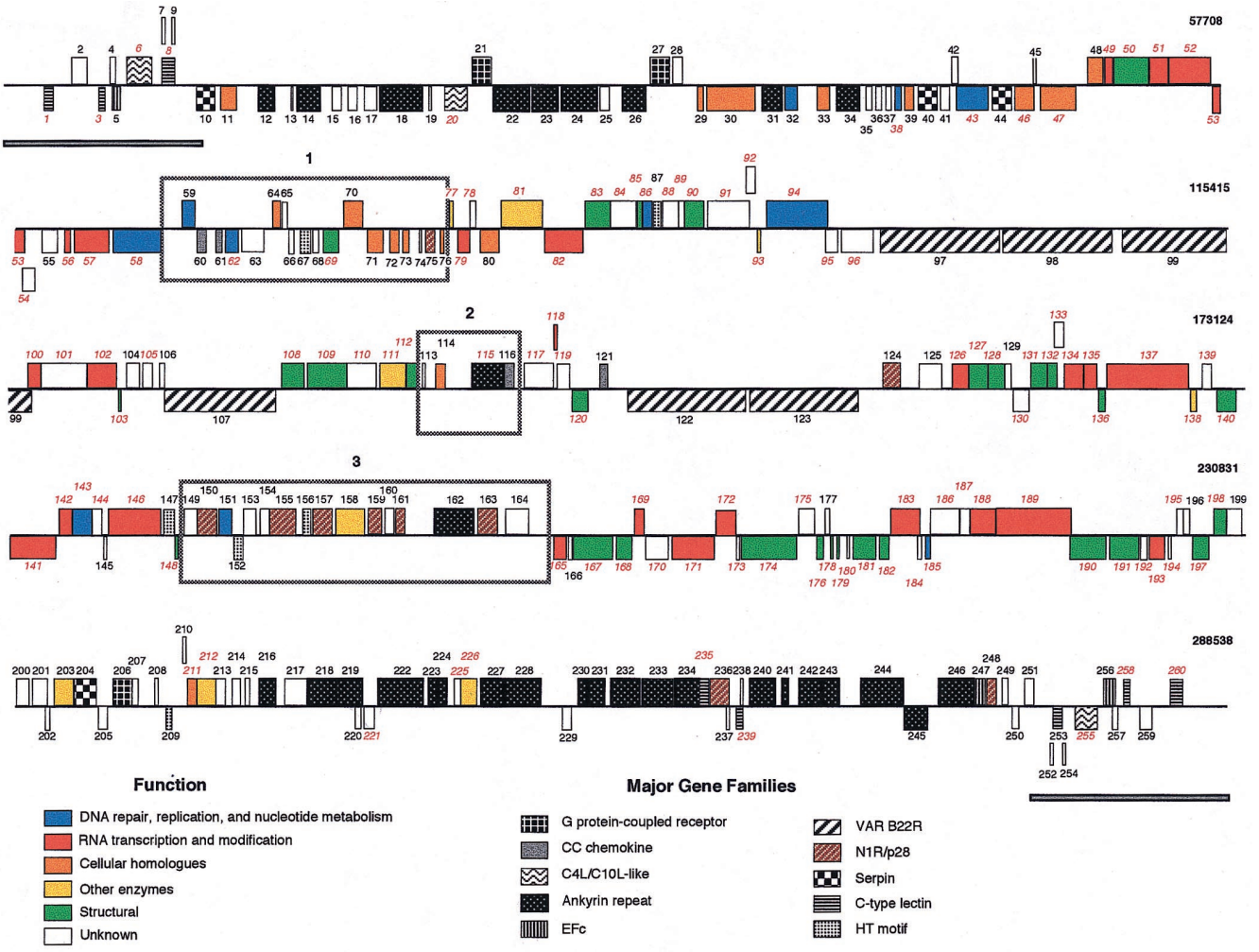

FIG. 1. Linear map of the FPV genome. ORFs are numbered from left to right based on the position of the methionine initiation codon. ORFs transcribed to the right are located above horizontal lines; ORFs transcribed to the left are below. VV homologues are indicated with red italicized numbers. Genes with similar functions and members of gene families are colored according to the figure key. ITRs are represented as gray bars below the ORF map. Boxed regions 1 to 3 indicate novel coding regions at junction sites of major genome rearrangements, and they correspond to similarly numbered regions shown in Fig. 4.

(TAAATG) at the ATG codon (131). The TAAATG late promoter has been previously described to be located upstream of FPV late genes (17, 91, 163, 191), and it is known that early-late and late promoters can be exchanged between FPV and VV with no loss of temporal specificity (27).

Transcription and mRNA biogenesis. FPV contains 26 genes involved in poxvirus transcriptional processes (Table 1). These include RNA polymerase subunits; mRNA transcription initiation, elongation, and termination factors; and the enzymes that direct posttranscriptional processing of viral mRNA (118). FPV RNA polymerase subunits include homologues of VV RPO147 (FPV137), RPO132 (FPV189), RAP94 (FPV141), RPO35 (FPV193), RPO30 (FPV100), RPO22 (FPV135), RPO19 (FPV169), RPO18 (FPV056), and RPO7 (FPV118). Homologues of all previously described early (E), intermediate (I), and late (L) poxvirus transcription factors (TFs) are found in FPV, including the following: $VETF_S$ (FPV057), $VETF_L$ (FPV171), VITF-3 (FPV172 and FPV188), VLTF-1 (FPV126), VLTF-2 (FPV049), VLTF-3 (FPV165), and VLTF-4 (FPV142) (87, 191). FPV079 and FPV183 encode elongation factors for late transcription (VV G2R and A18R) (22, 44, 186). Both transcriptional terminator NPH-1 (FPV052) and the RNA helicase NPH-II (FPV082) are present. FPV146 and FPV051 encode both subunits of the mRNA capping enzyme, and FPV102 and FPV134 encode both subunits of the poly(A) polymerase. FPV053 and FPV054 contain MutT-like motifs and are similar to VV D10R and D9R (85). D10R has recently been shown to be a negative regulator of viral transcription (149).

Nucleotide metabolism. FPV contains homologues of thymidine kinase (FPV086), dUTP pyrophosphatase (FPV038), glutaredoxin (FPV077), two deoxycytidine kinases (dCKs; FPV059 and FPV151), and a putative DNase II (FPV032) (Table 1). Genes encoding dCK and DNase II are unique to FPV and have been previously described (86, 93). Interestingly, sequencing of the complete genome has revealed a second dCK gene (FPV151). These two FPV dCK genes are 42% identical to each other and exhibit 32% amino acid identity to cellular dCK (Table 1). The DNase II homologue, FPV032, is truncated compared to the previously described FPV gene, FPCEL-1 (93). FPV032 represents the largest subunit ($\alpha 2$) of cellular DNase II and includes the conserved histidine at the potential active site (99, 174). The function of this gene in the viral replication cycle is unknown; however, FPCEL-1 is not

TABLE 1. FPV ORFs

ORF	Position (length, aa)[a]	Best match					Predicted structure and/or function[c]	Promoter type[d]	FPV accession no.[b]	Corresponding ORF		Reference(s)
		BlastP score	% Identity	Length, aa[a]	Accession no.[b]	Species[e]				VV	MCV	
FPV001	2491–1877 (205)	121	29	134	AF021350	*Rattus norvegicus*	C-type lectin family; TM			A40R		36
FPV002	3367–4032 (222)								A06621			36
FPV003	4871–4503 (123)	89	30	122	Q07108	*Homo sapiens*	C-type lectin family	E	C31685	A40R		36
FPV004	5125–5427 (101)								A06621			36
FPV005	5589–5224 (122)						EFc family	E	E31685			36
FPV006	5931–7184 (418)	250	27	298	P03296	VV	C4L/C10L-like family		P14361	C10L		36
FPV007	7599–7814 (72)								D00295			
FPV008	7681–8181 (167)	197	29	127	AB015628	*Gallus gallus*	C-type lectin family; SP		P14370	A40R		
FPV009	8048–8245 (66)								D00295			
FPV010	10190–9126 (355)	354	30	357	AB006423	*H. sapiens*	Serpin family		P14369			
FPV011	11112–10279 (278)	596	43	275	S32367	*Bos taurus*	α-SNAP	E				
FPV012	13033–12041 (331)	212	37	127	Q01485	*H. sapiens*	Ankyrin repeat family					
FPV013	13890–13711 (60)											
FPV014	15222–13912 (437)	293	28	334	U13616	*H. sapiens*	Ankyrin repeat family					
FPV015	16176–15646 (177)						TM					
FPV016	17068–16355 (238)											
FPV017	17913–17179 (245)						V-type Ig domain					
FPV018	20091–17992 (700)	288	24	509	X69063	*Mus musculus*	Ankyrin repeat family; TM	L				
FPV019	20487–20176 (104)											
FPV020	22302–21025 (426)	210	24	283	L22579	VAR	C4L/C10L-like family			C10L		
FPV021	22393–23352 (320)	560	36	288	AF100204	*Macaca mulatta*	G-protein-coupled receptor family; TM	E				
FPV022	25111–23378 (578)	289	27	395	L35601	*Drosophila melanogaster*	Ankyrin repeat family					
FPV023	26496–25195 (434)	303	36	206	AB002377	*H. sapiens*	Ankyrin repeat family					
FPV024	28352–26565 (596)	394	26	456	L35601	*D. melanogaster*	Ankyrin repeat family					
FPV025	29066–28458 (203)											
FPV026	30754–29447 (436)	248	33	214	U21734	*Caenorhabditis elegans*	Ankyrin repeat family					
FPV027	30796–31803 (336)	541	36	296	AF100204	*Macaca mulatta*	G-protein-coupled receptor family; TM	E				
FPV028	31857–32396 (180)											
FPV029	33396–33025 (124)	201	36	112	AF151905	*H. sapiens*	Conserved hypothetical protein	E				
FPV030	35950–33500 (817)	1,334	40	716	P22413	*H. sapiens*	Alkaline phosphodiesterase; TM		AJ006408			93
FPV031	37100–36078 (341)	218	31	185	L35601	*D. melanogaster*	Ankyrin repeat family		AJ006408			93
FPV032	37858–37163 (232)	374	40	211	Z46266	*C. elegans*	DNase II		AJ006408			93
FPV033	39535–38675 (287)	600	43	284	U39412	*H. sapiens*	α-SNAP		AJ006408			93
FPV034	40823–39579 (415)	305	30	271	AF102552	*R. norvegicus*	Ankyrin repeat family	E	AJ006408			93
FPV035	41414–41010 (135)											
FPV036	41913–41455 (153)											
FPV037	42406–41921 (162)						TM					
FPV038	42840–42406 (145)	439	61	136	M89913	*H. sapiens*	dUTP pyrophosphatase	L		F2L		
FPV039	43418–42894 (175)	133	27	143	AF115380	*R. norvegicus*	Bcl-2; TM	E				
FPV040	44476–43466 (337)	150	19	290	L40377	*H. sapiens*	Serpin family; TM	E				
FPV041	45178–44561 (206)						TM					
FPV042	45222–45476 (85)											
FPV043	46957–45266 (564)	1,553	52	570	X84740	*H. sapiens*	DNA ligase		Z29716	A50R		150
FPV044	48067–46994 (358)	259	26	374	L28101	*H. sapiens*	Serpin family		Z29716			150
FPV045	49173–49391 (73)											150
FPV046	49228–48119 (370)	717	42	360	AB000199	*R. norvegicus*	Hydroxysteroid dehydrogenase	E	Z29716	A44L	MC152R	150
FPV047	51118–49283 (612)	784	35	542	AF071542	*H. sapiens*	Semaphorin; TM, SP			A39R		150
FPV048	51568–52350 (261)	441	44	196	AL034374	*H. sapiens*	GNS1/SUR4; TM					18
FPV049	52429–52890 (154)	390	50	148	P33814	VAR	Late transcription factor VLTF-2	I	S42254	A1L	MC103L	18
FPV050	52914–54569 (552)	1,667	57	550	Q08517	SPV	Rifampicin resistance protein		S42253	D13L	MC102L	18
FPV051	54604–55470 (289)	937	55	285	U60315	MCV	mRNA capping enzyme, small subunit		S42252	D12L	MC101L	18
FPV052	55548–57458 (637)	2,093	61	633	U60315	MCV	NPH-I, transcription termination factor	L	S42251	D11L	MC100R	18
FPV053	58142–57468 (225)	400	39	212	U60315	MCV	*mutT* motif; gene expression regulator		P32817	D10R	MC099R	18
FPV054	58821–58129 (231)	472	47	207	P04311	VV	*mutT* motif	E		D9R	MC098R	18
FPV055	59898–59074 (275)	97	24	183	P13596	*R. norvegicus*	V-type Ig domain		P21975			18, 163
FPV056	60547–60065 (161)	515	56	161	U60315	MCV	RNA polymerase subunit RPO18	L	P21967	D7R	MC097R	18, 163
FPV057	62435–60537 (633)	2,626	78	631	U60315	MCV	Early transcription factor $VETF_S$	L	P21966	D6R	MC095R	18, 163

Continued on facing page

TABLE 1—*Continued*

ORF	Position (length, aa)[a]	Best match					Predicted structure and/or function[c]	Promoter type[d]	FPV accession no.[b]	Corresponding ORF		Reference(s)
		BlastP score	% Identity	Length, aa[a]	Accession no.[b]	Species[e]				VV	MCV	
FPV058	64791–62419 (791)	2,364	57	785	S47250	RFV	NTPase; DNA replication	L	P21969	D5R	MC094R	163
FPV059	65732–66388 (219)	314	33	239	P27707	*H. sapiens*	Deoxycytidine kinase		P21974			163
FPV060	67002–66439 (188)	82	24	124	U48722	*H. sapiens*	CC chemokine family; TM, SP		P21973			163
FPV061	67765–67379 (129)	65	32	43	U74585	Herpesvirus	CC chemokine family; TM	E	P21972			163
FPV062	68517–67864 (218)	705	58	216	P32941	RFV	Uracil DNA glycosylase; TM	E	P21968	D4R	MC093R	163
FPV063	69767–68568 (400)							E	P21971			163
FPV064	69939–70538 (200)	365	44	163	X71973	*H. sapiens*	Glutathione peroxidase				MC066L	
FPV065	70545–70877 (111)							L				
FPV066	71223–70858 (122)											
FPV067	71611–71342 (90)						HT motif family					
FPV068	72382–71984 (133)	119	38	86	U17055	PBCV-1		E				
FPV069	73268–72459 (270)	172	29	251	P25952	RFV	Virion protein			D3R	MC092R	
FPV070	73394–74212 (273)	621	43	276	X74504	*Mus musculus*	*T10* gene product					
FPV071	75489–74623 (289)	504	38	291	AF132150	*D. melanogaster*	Conserved hypothetical protein					
FPV072	76236–75679 (186)	290	41	146	P19093	*Cavia porcellus*	β-NGF					
FPV073	76782–76261 (174)	71	26	110	AF110798	*H. sapiens*	IL-18 binding protein; SP					
FPV074	77351–77040 (104)						TM					
FPV075	77954–77358 (199)	129	29	131	AF017791	HaEPV	N1R/p28 family; TM					
FPV076	78457–78026 (144)	133	27	118	P34128	*Bungarus multicinctus*	β-NGF					
FPV077	78569–78943 (125)	290	42	120	U60315	MCV	Glutaredoxin	L		G4L	MC059L	
FPV078	79590–79898 (103)	119	26	109	P21024	VV	TM	L		G3L	MC057L	
FPV079	79596–78922 (225)	339	31	240	U60315	MCV	Putative elongation factor			G2R	MC058R	
FPV080	81019–79931 (363)	178	34	134	AJ007836	*Oncorhynchus mykiss*	TGF-β; TM					
FPV081	81091–82968 (626)	1,393	43	623	U60315	MCV	Metalloprotease	L	H48563	G1L	MC056L	16
FPV082	85003–82958 (682)	1,544	46	682	U60315	MCV	RNA helicase/NPH-II		G48563	I8R	MC050R	16
FPV083	85036–86298 (421)	1,713	74	421	D86731	CaPV	Virion core protein	L	F48563	I7L	MC049L	16
FPV084	86304–87473 (390)	1,529	69	391	D86731	CaPV			E48563	I6L	MC048L	16
FPV085	87477–87719 (81)	315	72	81	D86731	CaPV	TM, SP	L	P18521	I5L	MC047L	16
FPV086	87732–88280 (183)	612	65	180	D78347	CaPV	Thymidine kinase	E	P10052	J2R		16
FPV087	88360–88632 (91)	239	55	79	D86731	CaPV	HT motif family		B48563			16
FPV088	88668–89537 (290)	817	55	287	D86731	CaPV	DNA-binding phosphoprotein	EI	AJ223385	I3L	MC046L	16, 127
FPV089	89541–89735 (65)	204	63	68	D86731	CaPV	TM	L	AJ223385	I2L	MC045L	127
FPV090	89745–90677 (311)	1,077	65	301	U60315	MCV	Virion protein	L	AJ223385	I1L	MC044L	127
FPV091	90845–92812 (656)	386	23	453	U60315	MCV	TM		AJ223385	O1L	MC042L	127
FPV092	92757–93149 (131)	215	29	131	U60315	MCV		L	AJ223385	E11L	MC041L	127
FPV093	93433–93152 (94)	283	49	95	U60315	MCV	Potential redox protein *ERV1*		AJ223385	E10R	MC040R	127
FPV094	93460–96423 (988)	2,674	51	1,004	U60315	MCV	DNA polymerase	E	P21402	E9L	MC039L	127, 19
FPV095	97236–96421 (272)	673	50	264	U94848	VV	TM	E		E8R	MC038R	
FPV096	98944–97232 (571)	1,594	50	566	U60315	MCV				E6R	MC037R	
FPV097	104808–99073 (1912)	2,544	35	1,896	L22579	VAR	VAR B22R family; TM	E			MC035R	
FPV098	110282–104877 (1802)	2,556	36	1,806	L22579	VAR	VAR B22R family; TM	E			MC035R	
FPV099	116372–110526 (1949)	4,316	48	1,916	Y15035	CPV	VAR B22R family				MC035R	
FPV100	116439–116984 (182)	639	63	180	U60315	MCV	RNA polymerase subunit RPO30			E4L	MC034L	
FPV101	117040–119190 (717)	696	29	704	AF035773	VAR	TM			E2L	MC032L	
FPV102	119180–120595 (472)	1,222	49	465	U60315	MCV	Poly(A) polymerase PAP_L			E1L	MC031L	
FPV103	120936–120595 (114)	210	46	97	P07396	VV	DNA-binding virion core phosphoprotein	L		F17R	MC030R	
FPV104	121013–121642 (210)	97	31	130	U80056	AmEPV		L				
FPV105	121770–122213 (148)	464	53	147	U60315	MCV				F15L	MC025L	
FPV106	122506–122718 (71)											
FPV107	128102–122772 (1777)	2,483	34	1,817	U18339	VAR	VAR B22R family; TM	E			MC035R	
FPV108	128284–129414 (377)	873	44	373	P25392	MCV	Virion envelope protein; membrane		P36316	F13L	MC021L	35
FPV109	129455–131344 (630)	433	28	604	P21053	VV	Virion release		P36317	F12L	MC019L	35
FPV110	131387–132739 (451)	158	23	234	U60315	MCV		E	P36700	F11L	MC018L	35
FPV111	132820–134151 (444)	1,299	55	421	U52849	MCV	Ser/Thr protein kinase; virus assembly	L		F10L	MC017L	
FPV112	134129–134767 (213)	405	39	213	P33869	VAR	Putative membrane protein; TM			F9L	MC016L	
FPV113	134861–135058 (66)							E				
FPV114	135430–135978 (183)	518	58	174	U80192	*Arabidopsis thaliana*	HAL3 domain					

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TABLE 1—*Continued*

ORF	Position (length, aa)[a]	Best match					Predicted structure and/or function[c]	Promoter type[d]	FPV accession no.[b]	Corresponding ORF		Reference(s)
		BlastP score	% Identity	Length, aa[a]	Accession no.[b]	Species[e]				VV	MCV	
FPV115	137148–138773 (542)	340	28	412	L40632	*Mus musculus*	Ankyrin repeat family	E		M1L		
FPV116	138798–139157 (120)	95	31	57	P14844	*R. norvegicus*	CC chemokine family; SP					
FPV117	139706–141025 (440)	588	31	438	U60315	MCV	TM			G5R	MC060R	
FPV118	141030–141218 (63)	235	69	63	U60315	MCV	RNA polymerase subunit RP07			G5.5R	MC061R	
FPV119	141221–141784 (188)	379	39	176	U60315	MCV				G6R	MC062R	
FPV120	142783–141755 (343)	604	37	395	U60315	MCV	Virion core protein	L		G7L	MC065L	
FPV121	143187–143549 (121)	69	25	82	P10147	*H. sapiens*	CC chemokine family; SP					
FPV122	150066–144457 (1870)	2,506	34	1,862	L22579	VAR	VAR B22R family; TM	E			MC035R	
FPV123	155397–150100 (1766)	2,329	34	1,834	AF012825	ECT	VAR B22R family; TM	E	U17141		MC035R	
FPV124	156366–157232 (289)	173	39	129	AF017791	HaEPV	N1R/p28 family		U17141			
FPV125	158143–159177 (345)	96	33	115	P31836	*Bos taurus*	V-type Ig domain; SP	E				
FPV126	159602–160381 (260)	1,098	78	260	U60315	MCV	VLTF-1; TM	E, I, L	P15908	G8R	MC067R	20
FPV127	160397–161404 (336)	572	38	340	P32998	VAR	Myristylated protein; TM	L	P15909	G9R	MC068R	20
FPV128	161408–162136 (243)	919	69	243	U60315	MCV	Myristylated membrane protein; TM	L	P15910	L1R	MC069R	20
FPV129	162174–162461 (96)						TM	L	P15911		MC070R	20
FPV130	163359–162457 (301)	739	50	281	U60315	MCV		L	P15912	L3L	MC072L	20
FPV131	163385–164143 (253)	494	40	253	U60315	MCV	DNA-binding virion core protein VP8; TM	L	P15913	L4R	MC073R	20
FPV132	164147–164533 (129)	191	41	113	P07615	VV	Putative membrane protein; TM	L	P15914	L5R		20
FPV133	164487–164930 (148)	290	37	145	U60315	MCV		L	P15915	J1R	MC075R	20
FPV134	164966–165889 (308)	865	55	294	U60315	MCV	Poly(A) polymerase PAP_S		M17418	J3R	MC076R	57
FPV135	165889–166446 (186)	560	55	181	P07391	VV	RNA polymerase subunit RPO22		M17418	J4R	MC077R	57
FPV136	166852–166442 (137)	356	48	132	U60315	MCV	Membrane protein		M17418	J5L	MC078L	57
FPV137	166893–170753 (1287)	5,074	72	1,289	U60315	MCV	RNA polymerase subunit RPO147			J6R	MC079R	
FPV138	171265–170768 (166)	463	51	169	U60315	MCV	Protein-tyrosine phosphatase	L		H1L	MC082L	
FPV139	171281–171850 (190)	541	49	191	U60315	MCV	TM			H2R	MC083R	
FPV140	173017–172037 (327)	356	31	316	AF124516	LSV	Virion envelope protein p35; TM	L		H3L	MC084L	
FPV141	175414–173021 (798)	2,331	56	798	U60315	MCV	RNA polymerase-associated protein RAP94	L	L46396	H4L	MC085L	191
FPV142	175558–176079 (174)	80	26	149	S62819	ORF virus	VLTF-4	E, I	L46396	H5R	MC086R	191
FPV143	176083–177030 (316)	936	57	303	L22579	VAR	DNA topoisomerase	L	L46396	H6R	MC087R	191
FPV144	177038–177493 (152)	223	40	145	U94848	VV	Putative 17-kDa protein	E, L		H7R	MC088R	
FPV145	177770–177462 (103)	89	28	114	U60315	MCV	SP	L			MC089L	
FPV146	177778–180330 (851)	2,425	54	814	U60315	MCV	mRNA capping enzyme, large subunit	E, L		D1R	MC090R	
FPV147	180392–180703 (104)						HT motif family					
FPV148	181122–180706 (139)	141	31	133	U60315	MCV	Virion protein			D2L	MC091L	
FPV149	181390–181947 (186)											
FPV150	182017–182844 (276)	302	37	159	U41315	*H. sapiens*	N1R/p28 family					
FPV151	182887–183591 (235)	396	38	239	X77731	*Mus musculus*	dCK					
FPV152	183987–183607 (127)						HT motif family					
FPV153	184086–184709 (208)											
FPV154	184937–185386 (150)											
FPV155	185418–186641 (408)						N1R/p28 family					
FPV156	186900–187295 (132)						HT motif family					
FPV157	187344–188276 (311)	383	41	167	U41315	*H. sapiens*	N1R/p28 family	E				
FPV158	188369–189760 (464)	1,432	56	462	D31902	*Monodelphis domestica*	Photolyase					
FPV159	189902–190624 (241)	160	34	130	AF017791	HaEPV	N1R/p28 family					
FPV160	190708–191175 (156)											
FPV161	191226–191696 (157)	94	24	127	AF017791	HaEPV	N1R/p28 family; TM					
FPV162	193077–194885 (603)	499	31	464	X69063	*Mus musculus*	Ankyrin repeat family	E				
FPV163	195121–195909 (263)	171	32	137	AF017791	HaEPV	N1R/p28 family	E				
FPV164	196404–197552 (383)											
FPV165	199299–198625 (225)	956	77	225	U60315	MCV	Late transcription factor VLTF-3	I		A2L	MC104L	
FPV166	199514–199299 (72)	154	42	69	U60315	MCV		L			MC105L	
FPV167	201502–199532 (657)	2,074	61	654	U60315	MCV	Virion core protein P4b	L	P17355	A3L	MC106L	17
FPV168	202431–201568 (288)						Immunodominant virion protein	L	AJ005164	A4L		18
FPV169	202470–202970 (167)	472	57	166	U60315	MCV	RNA polymerase subunit RPO19	E, L	A08272	A5R	MC108R	

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TABLE 1—*Continued*

ORF	Position (length, aa)[a]	Best match					Predicted structure and/or function[c]	Promoter type[d]	FPV accession no.[b]	Corresponding ORF		Reference(s)
		BlastP score	% Identity	Length, aa[a]	Accession no.[b]	Species[e]				VV	MCV	
FPV170	204092–202971 (374)	781	38	373	P20985	VV			A08272	A6L	MC109L	
FPV171	206225–204099 (709)	2,526	65	709	U60315	MCV	Early transcription factor $VETF_L$	L		A7L	MC110L	
FPV172	206291–207193 (301)	721	45	298	U60315	MCV	Intermediate transcription factor VITF-3			A8R	MC111R	
FPV173	207388–207161 (76)	189	47	69	U60315	MCV	TM	L		A9L	MC112L	
FPV174	210064–207392 (891)	1,948	43	891	U60315	MCV	Virion core protein P4a	L	A20158	A10L	MC113L	
FPV175	210082–210903 (274)	654	47	304	U60315	MCV		L	A20158	A11R	MC114R	
FPV176	211422–210910 (171)	243	40	169	P33837	VAR	Virion protein	L		A12L	MC115L	
FPV177	211437–211640 (68)						SP					
FPV178	211848–211636 (71)	100	35	59	U60315	MCV	Virion protein; SP	L		A13L	MC117L	
FPV179	212190–211918 (91)	167	43	87	U60315	MCV	Virion envelope protein; TM	L		A14L	MC118L	
FPV180	212676–212386 (97)	79	40	35	P33840	VAR		L		A15L	MC120L	
FPV181	213769–212663 (369)	844	44	369	U60315	MCV	Putative myristylated membrane protein; TM	L		A16L	MC121L	
FPV182	214381–213788 (198)	294	36	179	P16711	VV	Phosphorylated virion membrane protein; TM	L		A17L	MC122L	
FPV183	214396–215781 (462)	1,269	53	458	U60315	MCV	DNA helicase; transcriptional elongation			A18R	MC123R	
FPV184	216018–215755 (88)	159	52	73	P33842	VAR		L		A19L	MC124L	
FPV185	216366–217664 (433)	489	27	429	U60315	MCV	Processivity factor	E		A20R	MC126R	
FPV186	216367–216029 (113)	255	45	113	U60315	MCV	TM			A21L	MC125L	
FPV187	217667–218134 (156)	318	43	143	P33845	VAR				A22R	MC127R	
FPV188	218148–219296 (383)	1,034	53	381	P20998	VV	Intermediate transcription factor VITF-3			A23R	MC128R	
FPV189	219326–222808 (1161)	4,855	76	1,156	U60315	MCV	RNA polymerase subunit RPO132	E		A24R	MC129R	
FPV190	224610–222751 (620)	281	25	348	U60315	MCV	A-type inclusion protein	L		A25L	MC130L	
FPV191	226070–224649 (474)	471	29	475	U60315	MCV	A-type inclusion protein	L		A26L	MC133L	
FPV192	226496–226074 (141)	322	43	141	U60315	MCV	TM			A28L	MC134L	
FPV193	227419–226514 (302)	693	45	300	P33812	VAR	RNA polymerase subunit RPO35			A29L	MC135L	
FPV194	227618–227397 (74)	118	37	58	P21088	VV				A30L	MC136L	
FPV195	227797–228135 (113)	160	30	107	U60315	MCV				A31R	MC138R	
FPV196	228139–228498 (120)							L				
FPV197	229395–228493 (301)	725	56	246	U60315	MCV	Virion assembly protein	L		A32L	MC140L	
FPV198	229584–230102 (173)	121	25	173	S61094	VV	C-type lectin like; TM			A34R	MC143R	
FPV199	230160–230816 (219)	113	25	108	U18697	*Ginglymostoma cirratum*	V-type Ig domain; TM					
FPV200	230794–231588 (265)	92	28	128	U18697	*G. cirratum*	V-type Ig domain	L				
FPV201	231612–232460 (283)	169	22	286	U60315	MCV		E	AF006064		MC144R	76
FPV202	232649–232350 (100)											
FPV203	232786–233640 (285)	245	26	247	JQ1743	RFV	Tyrosine PK; TM	E	AF006064			76
FPV204	233679–234704 (342)	243	22	347	AB006423	*H. sapiens*	Serpin family		AF006064			76
FPV205	235366–234713 (218)						TM					
FPV206	235481–236404 (308)	513	35	299	P32249	*H. sapiens*	G-protein-coupled receptor family	E				
FPV207	236417–236716 (100)											
FPV208	237605–237805 (67)											
FPV209	238466–238077 (130)						HT motif family	E				
FPV210	238970–239167 (66)											
FPV211	239109–239483 (125)	125	36	61	D30783	*H. sapiens*	EGF-like protein; TM			C11R		
FPV212	239489–240397 (303)	731	45	302	AB000450	*H. sapiens*	Ser/Thr PK			B1R		
FPV213	240451–240936 (162)						TM					
FPV214	241278–241649 (124)	91	29	124	U94848	VV	Putative 13.7-kDa protein					
FPV215	241765–241986 (74)						TM					
FPV216	242384–243271 (296)	124	24	275	L35601	*D. melanogaster*	Ankyrin repeat family					
FPV217	243703–244686 (328)	234	27	230	AF081810	LdNPV						
FPV218	244729–246111 (461)	298	26	349	L35601	*D. melanogaster*	Ankyrin repeat family	E				
FPV219	246144–247445 (434)	295	31	275	L35601	*D. melanogaster*	Ankyrin repeat family	E				
FPV220	247453–247103 (117)						TM					
FPV221	248001–247453 (183)	79	30	180	P21067	VV		L		A47L		
FPV222	248089–250329 (747)	433	25	641	X69063	*Mus musculus*	Ankyrin repeat family	E				
FPV223	250544–250966 (141)	181	33	141	AF153912	TPV	Ankyrin repeat family					
FPV224	250974–251411 (146)	131	31	143	X62907	*A. thaliana*	Ankyrin repeat family					
FPV225	251791–252102 (104)	118	32	101	Y11842	CPV				B20R		
FPV226	252108–252983 (292)	477	34	291	P16913	VV	Ser/Thr PK			B1R		
FPV227	253039–254121 (361)	189	32	153	U42580	PBCV-1	Ankyrin repeat family					
FPV228	254214–255788 (525)	316	29	395	X16609	*H. sapiens*	Ankyrin repeat family; TM	E				
FPV229	257507–256968 (180)											
FPV230	257639–258202 (188)	200	33	157	U50071	*C. elegans*	Ankyrin repeat family	E				166

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TABLE 1—*Continued*

ORF	Position (length, aa)[a]	Best match					Predicted structure and/or function[c]	Promoter type[d]	FPV accession no.[b]	Corresponding ORF		Reference(s)
		BlastP score	% Identity	Length, aa[a]	Accession no.[b]	Species				VV	MCV	
FPV231	258169–258936 (256)	209	31	193	U21734	*C. elegans*	Ankyrin repeat family					
FPV232	259206–260651 (482)	274	29	294	L35601	*D. melanogaster*	Ankyrin repeat family					
FPV233	260697–262232 (512)	243	26	318	L35601	*D. melanogaster*	Ankyrin repeat family	E				
FPV234	262266–263549 (428)	259	29	330	L35601	*D. melanogaster*	Ankyrin repeat family		P14367			
FPV235	263571–263999 (143)	108	26	56	X54868	*H. sapiens*	C-type lectin family; TM		P14372	A40R		166
FPV236	264005–264844 (280)	170	29	159	L26342	RFV	N1R/p28 family		P14365			166
FPV237	265059–264859 (67)								P14366			166
FPV238	265612–265794 (61)								D00295			166
FPV239	265666–265178 (163)	262	31	152	M88072	*G. gallus*	C-type lectin family; SP	E	P14371	A40R		166
FPV240	265785–267014 (410)	256	29	234	Q01485	*H. sapiens*	Ankyrin repeat family	E	P14360			166
FPV241	267416–267733 (106)	106	33	72	X69065	*Mus musculus*	Ankyrin repeat family					
FPV242	268121–269194 (358)	229	26	335	Q01485	*H. sapiens*	Ankyrin repeat family; TM					
FPV243	269266–270051 (262)	115	24	238	Y15035	CPV	Ankyrin repeat family					
FPV244	271027–273030 (668)	368	32	348	U43965	*H. sapiens*	Ankyrin repeat family					
FPV245	274352–273045 (436)	371	30	340	L35601	*D. melanogaster*	Ankyrin repeat family					
FPV246	274734–276509 (592)	330	33	274	X16609	*H. sapiens*	Ankyrin repeat family	E				
FPV247	276565–276936 (124)						EFc family[e]	E				
FPV248	277051–277503 (151)	130	30	139	AF017791	HaEPV	N1R/p28 family	E	P14364			166
FPV249	277887–278201 (105)								P14363			166
FPV250	278855–278436 (140)	210	42	95	L22174	MDV	US ORF2		P14362			166
FPV251	278971–279414 (148)	187	35	150	U60474	MYX	Serpin family		P14369			166
FPV252	280492–280295 (66)								D00295			166
FPV253	280859–280359 (167)	179	24	166	M88072	*G. gallus*	C-type lectin family; SP		P14370			166
FPV254	280941–280726 (72)								D00295			166
FPV255	282609–281356 (418)	252	27	296	U18337	VAR	C4L/C10L-like family		P14361	C10L		166
FPV256	282951–283316 (122)						EFc family	E	E31685			
FPV257	283415–283113 (101)								A06621			
FPV258	283669–284037 (123)	89	30	122	Q07108	*H. sapiens*	C-type lectin family	E	C31685	A40R		
FPV259	285173–284508 (222)								A06621			
FPV260	286049–286663 (205)	121	29	134	AF021350	*R. norvegicus*	C-type lectin family; TM			A40R		

[a] aa, amino acids.
[b] Accession numbers are from the GenBank or SwissProt database.
[c] Function was deduced either from the degree of amino acid similarity to known genes or by the presence of Prosite signatures. TM, a *Z* score of >1.96 was used for the prediction of transmembrane (TM) domains with the Memsat computer programs; SP, N-terminal signal peptide (*Z* score of >2.5 using Sigcleave).
[d] Putative promoter. E, early; I, intermediate; L, late.
[e] Similar to EFc ORF described in reference 36.

essential for viral growth in vitro (93). Cellular DNase II is thought to function in DNA catabolism during apoptosis (89, 168). FPV lacks other known poxvirus genes thought to be involved in nucleotide metabolism, including thymidylate kinase, thymidylate synthase, ribonucleotide reductase, guanylate kinase, and thioredoxin. This specific complement of nucleotide metabolism genes in FPV suggests that they have significance for cell and/or tissue tropism.

DNA replication and repair. FPV contains homologues of ChPV genes involved in DNA replication and repair (118) (Table 1). These include a DNA ligase (FPV043), ATP-GTP binding protein (FPV058), uracil DNA glycosylase (FPV062), DNA polymerase (FPV094) (19), DNA topoisomerase (FPV143), processivity factor (FPV185), and replication-essential protein kinase (FPV212).

Interestingly, FPV158 is a homologue of class II cyclobutane pyrimidine dimer (CPD) photolyases. Although the gene has been previously described in the *Entomopoxvirinae* (1), this is the first description of a photolyase in a ChPV. FPV158 is most similar to marsupial photolyase (56% identity over 462 amino acids) (188) and is slightly less similar to *Melanoplus sanguinipes* entomopoxvirus (EPV) photolyase (54% identity over 448 amino acids) (1). Both class II photolyase Prosite signatures (PS01083 and PS01084) are present with a single conservative substitution at residue 302. Although the function of this FPV gene is unknown, CPD photolyase is a photoreactive enzyme that efficiently repairs UV-induced CPD lesions in DNA, using visible light as an energy source (75). Since EPVs have insect hosts and FPV is mechanically vectored by insects (48), the presence of a photolyase gene in both viral genomes is suggestive of a relationship between a viral phase in insects and/or the environment and the need for this type of virus-encoded DNA repair.

Protein modification. FPV contains at least six genes with putative protein modification functions (Table 1). The homologues encoded include three serine/threonine protein kinases (PKs) (FPV111, FPV212, and FPV226), one tyrosine PK (FPV203), a tyrosine/serine protein phosphatase (FPV138), and a metalloprotease (FPV081). FPV212 and FPV226 are similar to the serine/threonine PKs B1R and B12R of VV. FPV111 is similar to VV F10L, a serine/threonine PK essential for phosphorylation of virus proteins during virion assembly (14, 54). FPV203 shows similarity to the product of a tyrosine PK-like ORF found in rabbit fibroma virus (RFV) (109); however, neither of these poxvirus proteins contains the critical Asp residue at the predicted active site (Prosite PS00109). FPV138 is a homologue of the VV H1L tyrosine/serine protein phosphatase, which is involved in VV assembly (54). FPV081 is a homologue of the VV protease G1L. This protein contains the characteristic amino-terminal His-XX-Glu-His inverted metalloprotease motif, and it may function in viral protein processing and virion morphogenesis (178).

Structural proteins. FPV encodes homologues of at least 31 known VV structural proteins, and the majority of them are associated with the intracellular mature virus particle (IMV) (Table 1). FPV homologues of VV core proteins include FPV069 (D3R), FPV083 (I7L), FPV090 (I1L), FPV103 (F17R), FPV120 (G7L), FPV131 (L4R), FPV148 (D2L), FPV167 (A3L), FPV168 (A4L), FPV174 (A10L), and FPV176 (A12L) (15, 25). FPV homologues of VV IMV membrane-associated proteins include FPV050 (D13L), FPV085 (I5L), FPV128 (L1R), FPV140 (H3L), FPV178 (A13L), FPV179 (A14L), and FPV182 (A17L). FPV lacks homologues of VV IMV membrane proteins A27L, which is required for extracellular enveloped virion (EEV) envelopment and egress and for heparan sulfate binding (41, 130), and D8L, a cell surface binding protein (103). FPV structural proteins FPV120, FPV131, FPV167, FPV174, FPV176, and FPV182, like their VV homologues, contain the conserved AG proteolytic cleavage sites, which suggests that aspects of structural protein processing are conserved in FPV (173). FPV197 is the homologue of VV ATP-GTP binding protein A32L, which likely functions in virion assembly and DNA packaging (38).

FPV contains three genes that encode proteins potentially associated with EEVs (118, 123). FPV108, FPV109, and FPV198 are similar to VV F13L, F12L, and A34R, respectively (35). Missing from FPV are obvious homologues of VV EEV genes B5R, A33R, A36R, and A56R. EEV membrane proteins are involved with EEV formation, release, and infectivity (23, 111, 181). Since these functions may be associated with aspects of host range, the lack of well-conserved homologues of these genes in FPV is not surprising.

Homologues of five genes representing two conserved poxvirus gene families with putative structural functions are present in FPV. The genes encoding FPV112 and FPV128, homologues of VV F9L and L1R, respectively, comprise one gene family (142). The genes encoding FPV127, FPV136, and FPV181, homologues of VV G9R, J5L, and A16L, respectively, comprise a second, small gene family. G9R and A16L proteins are myristylated and potentially soluble (105), and *J5L* is thought to be an essential gene (190). Invariant cysteine residues and putative transmembrane domains unique to each family are conserved in these FPV ORFs (142).

FPV190 and FPV191 are homologues of poxvirus A-type inclusion (ATI) proteins (Table 1), insoluble proteins that constitute the protein matrix of ATIs. Cytoplasmic ATIs are thought to protect mature virions from environmental insults, and they may be of significance for FPV transmission in nature (40, 82, 128, 133).

Host-related functions. FPV contains a significant number of putative host range genes that exhibit similarity to cellular genes and to other known poxvirus genes. This diverse complement of host range genes, some of which are novel, is suggestive of significant adaptation to the avian host. These genes may function in host immune evasion, immune modulation, and aspects of cell and/or tissue tropism or perform other cellular functions. Most of these genes are found in terminal regions of the FPV genome, although several groupings of them are more centrally located.

Immune evasion functions. FPV080 is a homologue of the eukaryotic transforming growth factor β (TGF-β) (Table 1; Fig. 2A). To our knowledge, this is the first TGF-β gene found in a virus genome. Similarities to eukaryotic TGF-β include the 112-amino-acid peptide region of the active protein, Prosite signature PS00250 (with one mismatch), and cysteines necessary for intra- and interchain disulfide bond formation. TGF-β is a multifunctional peptide that both stimulates connective tissue cell growth and differentiation, particularly during neovascularization and wound healing, and suppresses proliferation of most other cell types (58). TGF-β also exhibits a range of immunomodulatory effects, including suppression of cellular and humoral immune mechanisms, specifically generation and/or activity of cytotoxic T lymphocytes, natural killer (NK) cells, and lymphokine-activated killer cells, generation and/or activity of lymphokines (interleukin-1 [IL-1], IL-6, tumor necrosis factor, and IL-2); and production of polyclonal antibodies (58). Chemoattractant and proinflammatory properties have also been associated with TGF-β (58). A role for FPV080 in suppression of the host immune response and/or cell growth and differentiation is likely.

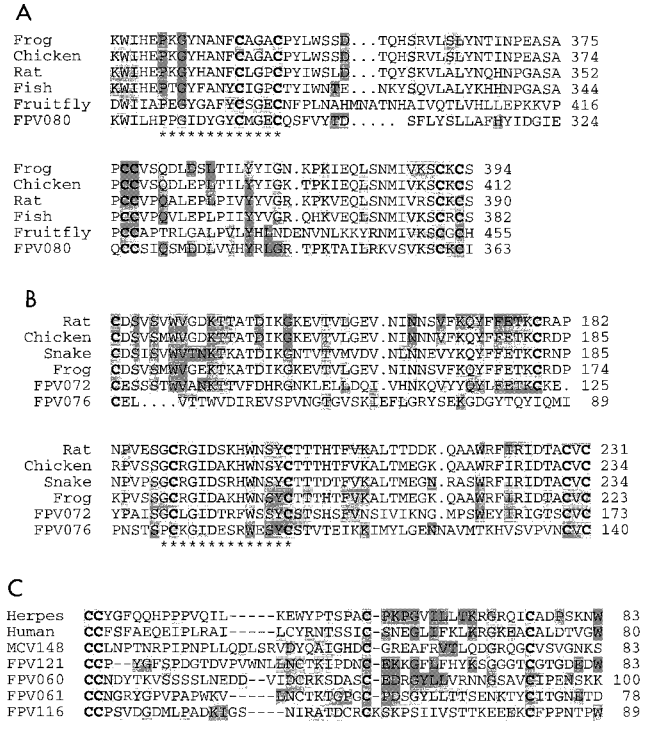

FIG. 2. Multiple amino acid sequence alignments of proteins encoded by putative FPV immune evasion genes. Boldfaced letters represent conserved cysteine residues, asterisks mark Prosite signatures, and shaded residues indicate identity to amino acids of FPV proteins. Amino acid positions are indicated on the right. (A) Alignment of FPV080 with TGF-β genes. The Prosite signature is PS00250. Frog, *Xenopus laevis*, accession no. P17247; chicken, *Gallus gallus*, accession no. P30371; rat, *Rattus norvegicus*, accession no. P17246; fish, *Oncorhynchus mykiss*, accession no. AJ007836; fruit fly, *Drosophila melanogaster*, accession no. M77012. (B) Alignment of FPV072 and FPV076 sequences with those of NGFs. The Prosite signature is PS00248. Rat, *Rattus norvegicus*, accession no. P25247; chicken, *Gallus gallus*, accession no. P05200; snake, *Bungarus multicinctus*, accession no. P34128; frog, *Xenopus laevis*, accession no. P211617. (C) Alignment of FPV060, FPV061, FPV116, and FPV121 sequences with those of viral and cellular CC chemokines. Herpesvirus, Kaposi's sarcoma-associated herpes-like virus, accession no. U74585; human, *Homo sapiens*, accession no. P22362; MCV148, molluscum contagiosum virus, accession no. U60315.

FPV072 and FPV076 are similar to cellular β nerve growth factor (β-NGF) (Table 1; Fig. 2B). This is the first example of a virus encoding β-NGF-like genes. Both FPV proteins contain the six cysteine residues involved in intrachain disulfide bonding and the Prosite β-NGF family signature (PS00248) (Fig. 2B). β-NGF, a member of the neurotrophin protein family, stimulates neuronal survival, division, and differentiation and promotes survival of memory B lymphocytes and mast cells (30, 97, 167). Recently, β-NGF has been shown to be an autocrine survival factor for human immunodeficiency virus type 1-infected macrophages (68). An FPV-encoded β-NGF may

be involved in promoting infected-cell survival. In addition, β-NGF has proinflammatory and immunomodulatory effects (5). β-NGF, which is produced by fibroblasts and keratinocytes in response to injury, induces differentiation, activation, and degranulation of mast cells and modifies expression of mast cell-derived immunoregulatory mediators and cytokines (34, 104, 138, 176, 177, 183). Conceivably, a virus-encoded β-NGF antagonist could have a role in inhibiting antiviral immune responses in FPV-infected skin and respiratory tract. Given that mast cells are initiators and amplifiers of innate immune responses, the presence of β-NGF homologues in FPV suggests that interference with early innate immune responses may be important for viral infection.

FPV060, FPV061, FPV116, and FPV121 exhibit similarity to the CC class of small soluble chemokines found in vertebrates (Table 1). The FPV genes contain the conserved pattern of four cysteines which are necessary for disulfide bond formation (Prosite PS00472), as well as other conserved residues (Fig. 2C). The FPV genes are similar in size (120 to 181 amino acids) to other known CC chemokines. Three of the products contain potential signal sequences at the N terminus, indicating that they may be secreted proteins. In general, CC chemokines attract T lymphocytes and NK cells to sites of infection (113). Other ChPVs modulate CC chemokine activity by secreting novel proteins that specifically bind CC chemokines and inhibit their effects in vitro and in vivo. These inhibitors are widespread among mammalian poxviruses, including VV, variola virus (VAR), cowpox virus (CPV), RFV, myxoma virus (MYX), and rabbitpox virus but is notably absent from FPV (4, 73, 94, 151). In molluscum contagiosum virus (MCV), a CC chemokine-like protein, MC148R, functions as a broad-spectrum CC and CXC chemokine antagonist (49). FPV's large repertoire of CC chemokine homologues functioning as antagonists could result in broad-range inhibition of normal CC chemokine function during host antiviral immune responses. Alternatively, as is the case for the viral macrophage-inhibitory protein 1 chemokine encoded by human herpesvirus 8, FPV chemokine homologues may function as agonists to modify normal host immune responses (47, 61).

FPV contains three genes encoding proteins with homology to G-protein-coupled receptors (Table 1). FPV021 and FPV027 are most similar to a monkey chemokine receptor protein (GPR1), while FPV206 is most closely related to the human Epstein-Barr virus-induced G-protein-coupled receptor (21). The highest level of amino acid similarity to cellular genes occurs at the seven transmembrane domains, the first cytoplasmic domain, and the second extracellular domain. The conserved acidic amino acid-Arg-aromatic amino acid triplets in the amino-terminal portion of the second aromatic loop, which have been implicated in the interaction with G proteins, are conserved in FPV021 and conservatively substituted in FPV027 and FPV206 (8). As with other G-protein-coupled receptors, the FPV proteins contain potential glycosylation sites at their carboxyl termini. G-protein-coupled receptors are integral membrane proteins that transduce extracellular signals to the intracellular environment through activation of the phosphatidylinositol-calcium second-messenger system (139). These receptors have been identified in the capripoxviruses and in swinepox virus, where their function is not known (37, 107). However, G-protein-coupled receptors encoded by several herpesvirus genomes are able to bind chemokines and invoke signal transduction responses that affect viral replication and pathogenesis in the host (2, 7, 13, 67).

FPV073 exhibits similarity to mammalian and ChPV IL-18-binding protein and contains potential N-glycosylation sites and a signal peptide (Table 1) (142, 184). Cellular and MCV IL-18bp homologues have been found to inhibit IL-18-dependent gamma interferon production (3, 185). IL-18 is a multifunctional proinflammatory cytokine of the IL-1 family that induces gamma interferon production, Th-1 responses, and NK cell activity, and it is important for effective host responses to VV infection in mice (50, 56, 79, 114, 161, 162). An anti-inflammatory function for FPV073 is likely.

FPV047 most closely resembles mammalian K/L-type semaphorins and the alcelaphine herpesvirus semaphorin homologue (accession no. U18243) (33% identity over 597 amino acids) (Table 1). Like the K/L-type semaphorin and alcelaphine herpesvirus semaphorin, FPV064 contains a potential amino-terminal signal sequence, a large semaphorin K/L domain, an immunoglobulin (Ig) domain, and a hydrophobic carboxyl terminus (62, 95). VV also encodes a K/L-like semaphorin homologue (A39R); however, the semaphorin domain is truncated and the Ig domain is absent (84). Semaphorins are a large family of secreted and membrane-associated proteins that act as axon guidance molecules during embryonic development and may affect organogenesis, vascularization, and angiogenesis (154). In addition, the CD100 semaphorin protein found on the surface of T lymphocytes functions in cell activation (52). The secreted VV A39R protein binds a plexin-like receptor found on lymphocytes and induces cytokine production and ICAM up-regulation in monocytes (43). The FPV semaphorin homologue may have a similar immunomodulatory function.

FPV contains eight ORFs (FPV001, FPV003, FPV008, FPV235, FPV239, FPV253, FPV258, and FPV260) with homology to C-type lectins NKG2 and CD94 proteins present on NK cells and CD69 protein present on lymphocytes (Table 1). Similar proteins have been described in poxviruses (VV and CPV) and African swine fever virus (ASFV) (122, 145, 179). Although the functions of these viral proteins are unknown, the VV C-type lectin protein, A40R, localizes to infected cell plasma membranes (179). C-type lectin cellular NK cell receptors bind class I major histocompatibility complex antigens and promote or inhibit immune activity through intracellular signaling pathways (66, 132, 175). It is conceivable that the expression of these proteins in FPV-infected cells interferes with normal immune surveillance or host responses.

FPV encodes five homologues of serine proteinase inhibitors (serpins) (FPV010, FPV040, FPV044, FPV204, and FPV251) (Table 1). All contain the serpin Prosite signature (PS00284) and exhibit 21 to 29% amino acid identity to each other. Serpin genes have been found in most ChPVs (rabbitpox virus, RFV, VV, VAR, CPV, MYX, and ectromelia virus [ECT]), where they perform host range functions involving anti-inflammatory activity and/or regulation of cellular apoptosis in specific cells through inhibition of IL-1β-converting enzyme, the cytotoxic-T-lymphocyte-derived protease granzyme B, and other caspases within the apoptosis-regulatory cascade (172).

Other host range functions. The gene encoding FPV039 is the first reported poxvirus member of the *Bcl-2* gene family. FPV039 resembles MCL1, a protein induced during monocyte/macrophage differentiation in myeloid leukemia cell lines, and BFL1 (29% identity over 134 amino acids), an antiapoptosis protein expressed specifically in the bone marrow, spleen, and thymus (88, 100) (Table 1; Fig. 3A). FPV039 contains one BH1 domain and one modified BH2 domain (Prosites PS01080 and PS01258) but lacks additional BH3 and BH4 domains. As with other viral Bcl-2 homologues, FPV039 may prevent a cellular apoptotic response to viral infection (12).

FPV070 is a homologue of the mouse *T10* gene and a yeast protein of unknown function (Table 1; Fig. 3B). This gene has

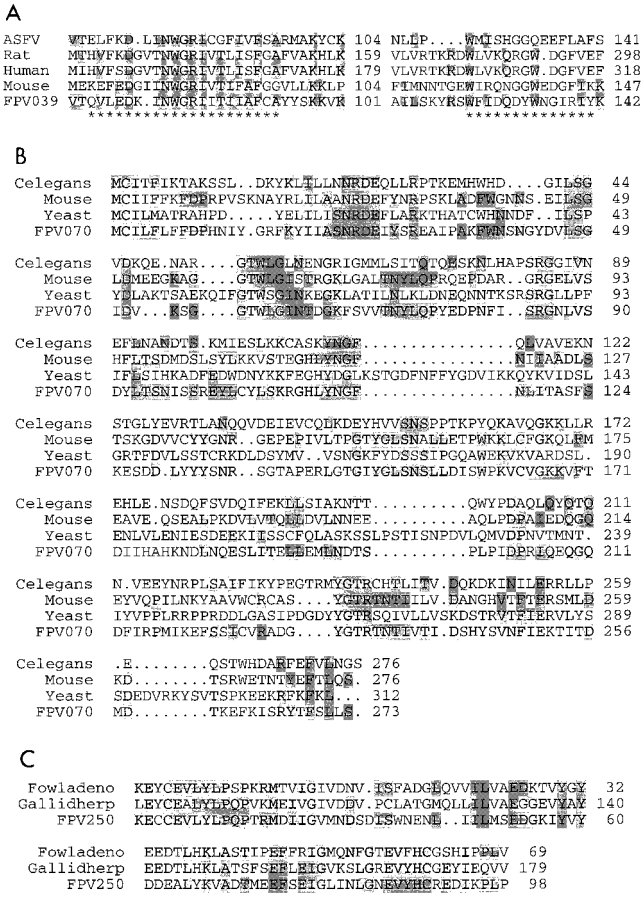

FIG. 3. Multiple amino acid sequence alignments of proteins encoded by putative FPV host range genes. Asterisks mark Prosite signatures; shaded residues exhibit identity to amino acids to FPV proteins. Amino acid positions are indicated. (A) Alignment of FPV039 sequence with those viral and cellular Bcl-2 homologues. ASFV, accession no. Q07819; rat, *Rattus norvegicus*, accession no. AF115380; human, *Homo sapiens*, accession no. Q07820; mouse, *Mus musculus*, accession no. Q07440. (B) Alignment of FPV070 sequence with homologues of the mouse T10 protein. Celegans, *Caenorhabditis elegans*, accession no. Z78016; mouse, *Mus musculus*, accession no. X74504; yeast, *S. cerevisiae*, accession no. P53275. (C) Alignment of FPV250 sequence with those of proteins from avian viruses. Fowladeno, fowl adenovirus, accession no. AF007578; Gallidherp, Marek's disease virus, accession no. L22174.

not been previously found in a viral genome. *T10* encodes a protein which is specifically expressed at high levels in epithelial cells of the trachea, esophagus, lung, and velopharyngeal region during early embryogenesis (74). The diphtheric form of FPV infection in chickens involves viral infection of the mucous membranes of the mouth, pharynx, and larynx and sometimes the trachea (169–171). An FPV T10 homologue may perform a host range function in epithelial cells of the respiratory tract.

FPV217 and FPV250 have similarities to genes of unknown function present in other viruses (Table 1; Fig. 3C). FPV217 is similar to a gene in *Lymantria dispar* nuclear polyhedrosis virus. FP250 is similar to putative proteins encoded by MDV and fowl adenovirus (44% identical over 99 amino acids) (32, 146). The presence of this homologue in three different avian DNA viruses suggests a significant avian host range function.

FPV211 contains an epidermal growth factor (EGF)-like domain which includes the six cysteine residues involved in disulfide bond formation, a potential signal peptide, and a transmembrane domain (Table 1). The similarity of FPV211 to secreted poxvirus EGF-like growth factors is based solely on the presence of the EGF domain. Poxvirus EGF-like growth factors are not essential for virus replication in vitro, influence virulence in vivo, and stimulate cell proliferation at sites of viral replication (110). FPV211 may contribute to the hyperplasia observed in FPV-infected tissue (169).

FPV contains 31 ORFs with ankyrin repeat motifs (Table 1). This large gene family is clustered at both ends of the genome, with two additional ORFs (FPV115 and FPV162) being found in more central locations (Fig. 1). Proteins encoded by FPV ankyrin family genes contain 1 to 12 copies of the ankyrin repeat motif (102), range in size from 104 to 747 amino acids, and are from 20 to 45% identical to each other depending on ORF size and alignment length. This level of amino acid identity is higher than that to ankyrin repeats of proteins found in a wide phylogenetic range of organisms. The ankyrin gene copy number may differ in FPV strains. Sequence from a genomic region in the right end of a highly passaged FPV strain contains nine fewer ankyrin genes than the number found here (166). Poxvirus ankyrin repeat genes have been associated with host range functions in MYX, CPV, and VV, and they may inhibit virus-induced apoptosis (70, 80, 119, 126, 153, 159). Ankyrin repeat motifs are clearly involved in mediating protein-protein interactions (101, 140). In CPV, which has a relatively broad host range, at least 16 ankyrin repeat genes have been identified (145). Loss or disruption of many of these genes in other orthopoxviruses that have a more restricted host range has suggested that loss of ankyrin genes may be associated with the narrowing of host range (6, 145).

FPV contains 10 ORFs (FPV075, FPV124, FPV150, FPV155, FPV157, FPV159, FPV161, FPV163, FPV236, and FPV248) with homology to N1R of RFV, p28 of ECT, and other ChPV and EPV genes (Table 1). Amino acid identity among FPV NR1/p28 family members is 20 to 38% and includes a conserved amino-terminal region with an invariant tryptophan residue. This domain is necessary for localization of RFV N1R to viral factories (31). FPV150 and FPV157, together with RFV N1R, ECT p28, and CPV and VAR homologues, contain a carboxyl-terminal C_3HC_4 RING finger. RING fingers are cysteine-rich zinc-binding motifs that are present in functionally diverse proteins, mediate protein-protein interactions, and help direct protein ubiquitination (81, 137). ECT p28 is a host range factor required for viral replication in mouse macrophages and for viral virulence in mice (143, 144); thus, a role in viral virulence and/or host range is likely for some members of this FPV family.

FPV064 encodes a homologue of cellular and MCV (MC066L) glutathione peroxidase (Table 1). FPV064 contains the glutathione peroxidase signature sequence (Prosites PS00460 and PS00763) including the active site for selenocysteine encoded by the opal codon (UGA). Cellular glutathione peroxidases reduce hydroxyperoxides with glutathione and are believed to provide protection from oxidative stress caused by ingested or endogenously formed hydroxyperoxides (157). MC066L protects human keratinocytes against cytotoxic effects of UV radiation and hydrogen peroxide and may permit efficient viral replication under conditions of environmental stress (148). A similar function for FPV064 is likely.

Cellular functions. FPV114 shares a 180-amino-acid conserved domain with proteins found in plants (accession no. U80192), yeast (accession no. P36024 and X88900), roundworms (accession no. Z81069), and bacteria (accession no. P24285, P30197, Q04810, and D90910). FPV114 is most closely related to the yeast *Hal3* and *SIS2* genes and a putative Hal3 homologue from the plant *Arabidopsis thaliana* (Table 1). These proteins function as inhibitory subunits of cellular pro-

tein phosphatases, and they promote salt tolerance and affect growth (53). FPV114, roundworm, and bacterial homologues lack the amino- and carboxyl-terminal domains found in the yeast protein. Bacterial homologues function in DNA/pantothenate and lantibiotic metabolism (39, 92). The wide phylogenetic distribution of FPV114 homologues suggests that their function is highly conserved.

FPV048 encodes a 261-amino-acid protein that is similar to the members of the GNS1/SUR4 family of integral membrane proteins (Table 1). Similarities to the *GNS1*/*SUR4* gene family include a defined motif (BLOCKs database signature BL01188) and a conserved protein structure consisting of an N-terminal region with two transmembrane domains, a central hydrophilic loop, a C-terminal region with one to three transmembrane domains, and the Prosite family signature (PS01188). The yeast *GNS1* and *SUR4* genes function in glucose metabolism, and they are suspected to have pleiotrophic functions in the cellular response to nutrient availability (60, 69, 129).

FPV011 and FPV033 are similar to the eukaryotic α-type soluble NSF attachment protein (α-SNAP) (Table 1). FPV033 has been previously described, but this is the first report of a second FPV α-SNAP homologue (93). FPV011 and FPV033 are similar in size (278 and 267 amino acids long, respectively) and exhibit 34% amino acid identity to each other over 249 amino acids. α-SNAPs are involved in vesicular trafficking, mediating intracellular membrane fusion by recruiting soluble NSF to membrane receptors (158). α-SNAPs and their yeast homologues (Sec17) are required for vesicular transport through the Golgi complex and for exocytosis (42, 117). The fact that FPV033 is not essential for growth in vitro suggests that it has a host range function (93).

FPV093 is the homologue of VV E10R, a protein that is conserved in many cytoplasmic DNA viruses and eukaryotes and contains a pattern of cysteine residues typical of glutaredoxin and thioredoxin redox-active centers (78). The homologue of this protein in ASFV, 9GL, has recently been shown to be involved in virion maturation and viral growth in swine macrophages (98).

FPV030 exhibits homology to human PC-1, which has alkaline phosphodiesterase and nucleotide pyrophosphatase activities and has been previously found in FPV (33, 93). The function of this conserved but nonessential FPV gene is unknown; however, it has been suggested that it may provide an external source of nucleotides or regulate signal transduction (93).

FPV046 encodes a homologue of 3-β-hydroxysteroid dehydrogenase (3βHSDH), previously described in FPV and other poxviruses (VV and MCV) (Table 1) (116, 142, 150). In VV, 3βHSDH has steroidogenic activity in vitro and is involved in viral virulence in vivo (116). Cellular 3βHSDH catalyzes the oxidative conversion of both δ(5)-ene-3-β-hydroxysteroid and ketosteroids, performing a crucial role in the biosynthesis of all classes of steroid hormones.

Two unrelated FPV ORFs, FPV029 and FPV071, have striking similarity to genes present in a diverse phylogenetic range of organisms (Table 1). FPV029 is similar to proteins of unknown function from yeast (accession no. P34222), bacteria (accession no. U67463 and AE000927), a roundworm (accession no. AF067936), a plant (accession no. AL031804), the fruit fly (accession no. AE0015722), and humans (accession no. AF151905). All of these proteins show several conserved domains, and the bacterial genes and the FPV037 ORF have similar lengths. FPV071 is similar to genes of unknown function from yeast (accession no. P40506), humans (accession no. AI391502), tomato (accession no. AI771876), and fruit fly (accession no. AF132150).

Gene families of unknown function. FPV097, FPV098, FPV099, FPV107, FPV122, and FPV123 are homologues of VAR B22R (Table 1). B22R homologues are also present in CPV, ECT, and MCV but are absent from VV (6, 71). FPV gene family members exhibit 34 to 52% amino acid identity to each other and 32 to 36% identity to the other poxvirus homologues, with the highest level of similarity being in the carboxyl-terminal regions. Several features make these FPV B22R homologues notable. They represent the largest genes in FPV (1,766 to 1,949 amino acids), and they comprise 12% of the viral genome. FPV contains multiple B22R homologues, while other poxviruses either contain a single copy of the gene or lack it (71, 108, 142). FPV B22R homologues are present in a central genomic region, while orthopoxvirus homologues are located in the terminal regions of their respective genomes (Fig. 1) (71, 108). Although no function has yet been assigned to any of these ORFs, it has been suggested that they are type II membrane proteins (108, 145).

FPV017, FPV055, FPV125, FPV199, and FPV200 have similarity to V-type Ig domains of diverse proteins (Table 1). All five proteins contain conserved Ig domain cysteines and surrounding residues. FPV017 and FPV199 are notably similar to each other (25% amino acid identity), as are FPV055 and FPV125 (37% identity over 270 amino acids). Cellular members of the Ig superfamily include secreted and membrane-bound receptors and cell adhesion proteins (180). Ig domain-containing proteins, including hemagglutinin, cytokine receptor, and HLA antigen homologues, are present in other poxviruses (141, 142, 147, 152).

FPV067, FPV087, FPV147, FPV152, FPV156, and FPV209 comprise the His-X-X-Thr motif gene family (HT motif). These genes exhibit 18 to 34% identity over 69 to 102 amino acids and contain the HT motif at residues 19 to 33 or 51 to 65. FPV147, FPV152, and FPV209 also have $HX_{4-5}T$ motifs upstream of the primary HT motif. The HT family ORFs have no significant similarity to other sequences in the database.

FPV006 and FPV255 are 35% identical to FPV020 over 283 amino acids. All three FPV ORFs are similar to VV C10L and C4L and homologues present in CPV and VAR (Table 1). Like their orthopoxvirus homologues, these FPV ORFs are located in the terminal genomic regions. Although their function has not been determined, C10L and C4L are dispensable for virus growth in cell culture (126).

Relationship of FPV to other ChPVs. FPV resembles other ChPVs in overall genome structure and composition (the presence of a central conserved core of genes, ITRs, and large numbers of homologues). However, compared to those of other ChPVs, the genome of FPV exhibits large-scale genomic rearrangement, more extensive gene families, and the presence of novel host range genes. Genomic comparisons of FPV and VV have shown major rearrangement of blocks of genes (115). Analysis of the complete FPV genomic sequence reveals that FPV contains at least two major genomic rearrangements in the conserved colinear core of genes present in VV, VAR, and MCV (71, 108, 142) (Fig. 4). A 12-kbp FPV genomic region containing ORFs FPV049 to FPV058 (comparable to VV A1L to D5R) is inverted and translocated toward the left end of the genome relative to VV. A 56-kbp FPV genomic region containing ORFs FPV077 to FPV112 (comparable to VV G4L to F9L) is inverted relative to VV. At the junction sites of these major rearrangements there are novel coding regions of 5 to 17.5 kbp (see boxed areas 1 to 3 in Fig. 1 and regions 1 to 3 in Fig. 4). Genes within these junction regions are predominantly homologues of cellular genes and/or are members of gene families. This clustering of cellular homologues and gene families in central genomic locations has not been previously ob-

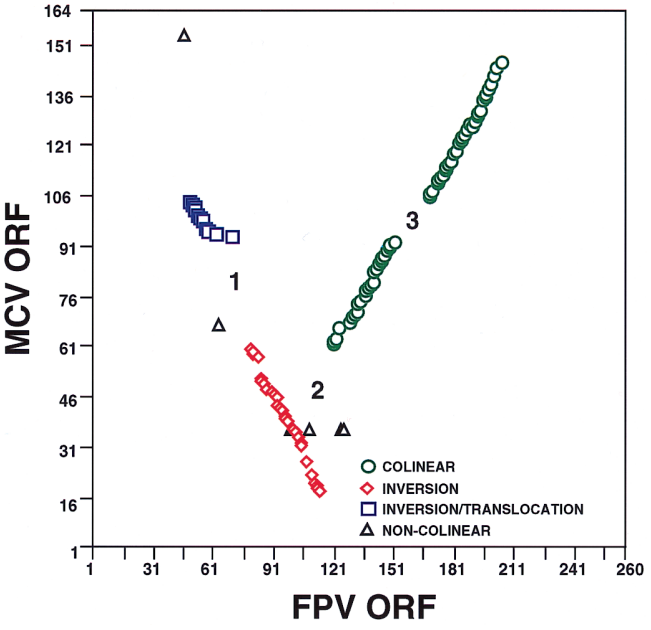

FIG. 4. Comparison of gene orders of FPV and MCV homologues. Symbols represent homologous genes. Green circles, colinear genes; red diamonds, inverted genes; blue squares, inverted and translocated genes; black triangles, noncolinear genes. Noncolinear genes include FPV046 (hydroxysteroid dehydrogenase) and FPV064 (glutathione peroxidase), as well as FPV097, FPV098, FPV099, FPV107, FPV122, and FPV123 (VAR B22R homologues). Areas numbered 1 to 3 indicate novel coding regions at junction sites of major genome rearrangement and correspond to similarly numbered boxes in Fig. 1.

served in the subfamily *Chordopoxvirinae*, in which these types of genes are generally found in terminal variable regions of the genome (71, 106, 141). This observation suggests that blocks of genes may have translocated from terminal variable regions of the genome to central regions during large-scale rearrangements of FPV. FPV genome colinearity with genomes of other ChPVs is also interrupted at multiple sites by insertions or deletions of individual genes and multiple copies of B22R gene family members (Fig. 1 and 4 and Table 1).

The FPV genome (260 to 309 kbp) is larger than other completely sequenced ChPV genomes (178 to 191 kbp). This size difference is due largely to the presence of multiple and, in some cases, large gene families. In other ChPVs, gene families contain fewer members (e.g., genes encoding ankyrin and serpins) or are represented as a single gene (e.g., genes encoding N1R/p28, B22R, CC chemokine, and NKG2-like proteins). Notably, the FPV ankyrin repeat family (31 genes), N1R/p28 family (10 genes), and B22R family (6 genes) comprise 32% of the total genome. In addition, cellular homologues novel to FPV are often found in multiple copies (e.g., β-NGF, α-SNAP, and dCK). It has been suggested that the size of the ankyrin repeat multigene family may affect poxvirus host range (145). The large number of FPV multigene family members, together with the wide avian host range of FPV, provides support for the role of gene families in host range (169).

Conclusions. FPV genome analysis provides basic knowledge of viral functions, including mRNA biogenesis, DNA replication and repair, nucleotide metabolism, protein processing, manipulation of cellular responses, viral virulence, and host range, which underlie FPV interactions with its avian host and the environment. An improved understanding of these interactions will permit the engineering of novel vaccine viruses and expression vectors with enhanced efficacy and greater versatility. Additionally, the identification and characterization of FPV virulence and host range genes will contribute novel concepts to our overall understanding of pathogen-host interactions, information that is likely to have a broad impact on future strategies for controlling avian infectious diseases in general.

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REFERENCES

1. **Afonso, C. L., E. R. Tulman, Z. Lu, E. Oma, G. F. Kutish, and D. L. Rock.** 1999. The genome of *Melanoplus sanguinipes* entomopoxvirus. J. Virol. **73:** 533–552.
2. **Ahuja, S. K., and P. M. Murphy.** 1993. Molecular piracy of mammalian interleukin-8 receptor type B by herpesvirus saimiri. J. Biol. Chem. **268:** 20691–20694.
3. **Aizawa, Y., K. Akita, M. Taniai, K. Torigoe, T. Mori, Y. Nishida, S. Ushio, Y. Nukada, T. Tanimoto, H. Ikegami, M. Ikeda, and M. Kurimoto.** 1999. Cloning and expression of interleukin-18 binding protein. FEBS Lett. **445:** 338–342.
4. **Alcami, A., J. A. Symons, P. D. Collins, T. J. Williams, and G. L. Smith.** 1998. Blockade of chemokine activity by a soluble chemokine binding protein from vaccinia virus. J. Immunol. **160:**624–633.
5. **Aloe, L., M. D. Simone, and F. Properzi.** 1999. Nerve growth factor: a neurotrophin with activity on cells of the immune system. Microsc. Res. Tech. **45:**285–291.
6. **Antoine, G., F. Scheiflinger, F. Dorner, and F. G. Falkner.** 1998. The complete genomic sequence of the modified vaccinia Ankara strain: comparison with other orthopoxviruses. Virology **244:**365–396.
7. **Arvanitakis, L., E. Geras-Raaka, A. Varma, M. C. Gershengorn, and E. Cesarman.** 1997. Human herpesvirus KSHV encodes a constitutively active G-protein-coupled receptor linked to cell proliferation. Nature **385:**347–350.
8. **Attwood, T. K., E. E. Eliopoulos, and J. B. C. Findlay.** 1991. Multiple sequence alignment of protein families showing low sequence homology: a methodological approach using database pattern-matching discriminators for G-protein-linked receptors. Gene **98:**153–159.
9. **Back, A., R. A. Soncini, O. Ruthes, S. Madureira, Jr., and R. Flores.** 1995. An atypical fowl pox outbreak in broilers in southern Brazil. Avian Dis. **39:** 902–906.
10. **Baldick, C. J., Jr., J. G. Keck, and B. Moss.** 1992. Mutational analysis of the core, spacer, and initiator regions of vaccinia virus intermediate-class promoters. J. Virol. **66:**4710–4719.
11. **Baldick, C. J., Jr., and B. Moss.** 1993. Characterization and temporal regulation of mRNAs encoded by vaccinia virus intermediate-stage genes. J. Virol. **67:**3515–3527.
12. **Barry, M., and G. McFadden.** 1998. Apoptosis regulators from DNA viruses. Curr. Opin. Immunol. **10:**422–430.
13. **Beisser, P. S., G. Grauls, C. A. Bruggeman, and C. Vink.** 1999. Deletion of the R78 G protein-coupled receptor gene from rat cytomegalovirus results in an attenuated, syncytium-inducing mutant strain. J. Virol. **73:**7218–7230.
14. **Betakova, T., E. J. Wolffe, and B. Moss.** 1999. Regulation of vaccinia virus morphogenesis: phosphorylation of the A14L and A17L membrane proteins and C-terminal truncation of the A17L protein are dependent on the F10L kinase. J. Virol. **73:**3534–3543.
15. **Binns, M., C. Mason, and M. Boursnell.** 1990. A 39,000 M_r immunodominant protein of fowlpox virus contains multiple copies of a 12 amino acid repeat sequence. J. Gen. Virol. **71:**2883–2888.
16. **Binns, M. M., M. E. G. Boursnell, and M. A. Skinner.** 1992. Gene translocations in poxviruses: the fowlpox virus thymidine kinase gene is flanked by 15 bp direct repeats and occupies the locus which in vaccinia virus is occupied by the ribonucleotide reductase large subunit gene. Virus Res. **24:** 161–172.
17. **Binns, M. M., M. E. G. Boursnell, F. M. Tomley, and J. Campbell.** 1989. Analysis of the fowlpoxvirus gene encoding the 4b core polypeptide and demonstration that it possesses efficient promoter sequences. Virology **170:** 288–291.
18. **Binns, M. M., B. S. Britton, C. Mason, and M. E. Boursnell.** 1990. Analysis of the fowlpox virus genome region corresponding to the vaccinia virus D6 to A1 region: location of, and variation in, non-essential genes in poxviruses. J. Gen. Virol. **71:**2873–2881.
19. **Binns, M. M., L. Stenzler, F. M. Tomley, J. Campbell, and M. E. Boursnell.** 1987. Identification by a random sequencing strategy of the fowlpoxvirus DNA polymerase gene, its nucleotide sequence and comparison with other viral DNA polymerases. Nucleic Acids Res. **15:**6563–6573.

20. **Binns, M. M., F. M. Tomley, J. Campbell, and M. E. G. Boursnell.** 1988. Comparison of a conserved region in fowlpox virus and vaccinia virus genomes and the translocation of the fowlpox virus thymidine kinase gene. J. Gen. Virol. **69:**1275–1283.
21. **Birkenbach, M., K. Josefsen, R. Yalamanchili, G. Lenoir, and E. Kieff.** 1993. Epstein-Barr virus-induced genes: first lymphocyte-specific G protein-coupled peptide receptors. J. Virol. **67:**2209–2220.
22. **Black, E. P., N. Moussatche, and R. C. Condit.** 1998. Characterization of the interactions among vaccinia virus transcription factors G2R, A18R, and H5R. Virology **245:**313–322.
23. **Blasco, R., and B. Moss.** 1991. Extracellular vaccinia virus formation and cell-to-cell virus transmission are prevented by deletion of the gene encoding the 37,000-dalton outer envelope protein. J. Virol. **65:**5910–5920.
24. **Blaxland, J. D.** 1976. Atypical fowl pox in chickens. Vet. Rec. **99:**222.
25. **Boulanger, D., P. Green, T. Smith, C.-P. Czerny, and M. A. Skinner.** 1998. The 131-amino-acid repeat region of the essential 39-kilodalton core protein of fowlpox virus FP9, equivalent to vaccinia virus A4L protein, is nonessential and highly immunogenic. J. Virol. **72:**170–179.
26. **Boyle, D. B.** 1994. Disease and fertility control in wildlife and feral animal populations: options for vaccine delivery using vectors. Reprod. Fertil. Dev. **6:**393–400.
27. **Boyle, D. B.** 1992. Quantitative assessment of poxvirus promoters in fowlpox and vaccinia virus recombinants. Virus Genes **6:**281–290.
28. **Boyle, D. B., and H. G. Heine.** 1993. Recombinant fowlpox virus vaccines for poultry. Immunol. Cell Biol. **71:**391–397.
29. **Boyle, D. B., A. D. Pye, and B. E. H. Coupar.** 1997. Comparison of field and vaccine strains of Australian fowlpox viruses. Arch. Virol. **142:**737–748.
30. **Bradshaw, R. A., T. L. Blundell, R. Lapatto, N. Q. McDonald, and J. Murray-Rust.** 1993. Nerve growth factor revisited. Trends Biochem. Sci. **18:** 48–52.
31. **Brick, D. J., R. D. Burke, L. Schiff, and C. Upton.** 1998. Shope fibroma virus RING finger protein N1R binds DNA and inhibits apoptosis. Virology **249:** 42–51.
32. **Brunovskis, P., and L. F. Velicer.** 1995. The Marek's disease virus (MDV) unique short region: alphaherpesvirus-homologous, fowlpox virus-homologous, and MDV-specific genes. Virology **206:**324–338.
33. **Buckley, M. F., K. A. Loveland, W. J. McKinstry, O. M. Garson, and J. W. Goding.** 1990. Plasma cell membrane glycoprotein PC-1. J. Biol. Chem. **265:** 17506–17511.
34. **Bullock, E. D., and E. M. Johnson, Jr.** 1996. Nerve growth factor induces the expression of certain cytokine genes and *bcl-2* in mast cells. Potential role in survival promotion. J. Biol. Chem. **271:**27500–27508.
35. **Calvert, J. G., R. Ogawa, N. Yanagida, and K. Nazerian.** 1992. Identification and functional analysis of the fowlpox virus homolog of the vaccinia virus p37K major envelope antigen gene. Virology **191:**783–792.
36. **Campbell, J. I. A., M. W. Binns, F. M. Tomley, and M. E. G. Boursnell.** 1989. Tandem repeated sequences within the terminal region of the fowlpox virus genome. J. Gen. Virol. **70:**145–154.
37. **Cao, J. X., P. D. Gershon, and D. N. Black.** 1995. Sequence analysis of *Hin*dIII Q2 fragment of capripoxvirus reveals a putative gene encoding a G-protein-coupled chemokine receptor homologue. Virology **209:**207–212.
38. **Cassetti, M. C., M. Merchlinsky, E. J. Wolffe, A. S. Weisberg, and B. Moss.** 1998. DNA packaging mutant: repression of the vaccinia virus A32 gene results in noninfectious, DNA-deficient, spherical, enveloped particles. J. Virol. **72:**5769–5780.
39. **Chen, N.-Y., S.-Q. Jiang, D. A. Klein, and H. Paulus.** 1993. Organization and nucleotide sequence of the *Bacillus subtilis* diaminopimelate operon, a cluster of genes encoding the first three enzymes of diaminopimelate synthesis and dipicolinate synthase. J. Biol. Chem. **268:**9448–9465.
40. **Cheville, N. F.** 1966. Cytopathologic changes in fowlpox (turkey origin) inclusion body formation. Am. J. Pathol. **49:**723–737.
41. **Chung, C.-S., J.-C. Hsiao, Y.-S. Chang, and W. Chang.** 1998. A27L protein mediates vaccinia virus interaction with cell surface heparan sulfate. J. Virol. **72:**1577–1585.
42. **Clary, D. O., I. C. Griff, and J. E. Rothman.** 1990. SNAPs, a family of NSF attachment proteins involved in intracellular membrane fusion in animals and yeast. Cell **61:**709–721.
43. **Comeau, M. R., R. Johnson, R. F. DuBose, M. Petersen, P. Gearing, T. VandenBos, L. Park, T. Farrah, R. M. Buller, J. I. Cohen, L. D. Strockbine, C. Rauch, and M. K. Spriggs.** 1998. A poxvirus-encoded semaphorin induces cytokine production from monocytes and binds to a novel cellular semaphorin receptor, VESPR. Immunity **8:**473–482.
44. **Condit, R. C., Y. Xiang, and J. I. Lewis.** 1996. Mutation of vaccinia virus gene G2R causes suppression of gene A18R ts mutants: implications for control of transcription. Virology **220:**10–19.
45. **Coupar, B. E. H., T. Teo, and D. B. Boyle.** 1990. Restriction endonuclease mapping of the fowlpox virus genome. Virology **179:**159–167.
46. **Cunningham, C. H.** 1984. Avian pox, p. 597–609. *In* M. S. Hofstad, H. J. Barnes, B. W. Calnek, W. M. Reid, and H. W. Yoder, Jr. (ed.), Diseases of poultry. Iowa State University Press, Ames.
47. **Dairaghi, D. J., R. A. Fan, B. E. McMaster, M. R. Hanley, and T. J. Schall.** 1999. HHV8-encoded vMIP-I selectively engages chemokine receptor CCR8. J. Biol. Chem. **274:**21569–21574.
48. **DaMassa, A. J.** 1965. The role of *Culex tarsalis* in the transmission of fowl pox virus. Avian Dis. **10:**57–66.
49. **Damon, I., P. M. Murphy, and B. Moss.** 1998. Broad spectrum chemokine antagonistic activity of a human poxvirus chemokine homolog. Proc. Natl. Acad. Sci. USA **95:**6403–6407.
50. **Dao, T., K. Ohashi, T. Kayano, M. Kurimoto, and H. Okamura.** 1996. Interferon-gamma-inducing factor, a novel cytokine, enhances Fas ligand-mediated cytotoxicity of murine T helper 1 cells. Cell. Immunol. **173:** 230–235.
51. **Davison, A. J., and B. Moss.** 1989. Structure of vaccinia virus early promoters. J. Mol. Biol. **210:**749–769.
52. **Delaire, S., A. Elhabazi, A. Bensussan, and L. Boumsell.** 1998. CD100 is a leukocyte semaphorin. Cell. Mol. Life Sci. **54:**1265–1276.
53. **De Nadal, E., J. Clotet, F. Posas, R. Serrano, N. Gomez, and J. Arino.** 1998. The yeast halotolerance determinant Hal3p is an inhibitory subunit of the Ppz1p Ser/Thr protein phosphatase. Proc. Natl. Acad. Sci. USA **95:**7357–7362.
54. **Derrien, M., A. Punjabi, M. Khanna, O. Grubisha, and P. Traktman.** 1999. Tyrosine phosphorylation of A17 during vaccinia virus infection: involvement of the H1 phosphatase and the F10 kinase. J. Virol. **73:**7287–7296.
55. **Devereux, J., P. Haeberli, and O. Smithies.** 1984. A comprehensive set of sequence analysis programs for the VAX. Nucleic Acids Res. **12:**387–395.
56. **Dinarello, C. A.** 1999. IL-18: a TH1-inducing, proinflammatory cytokine and new member of the IL-1 family. J. Allergy Clin. Immunol. **103:**11–24.
57. **Drillien, R., D. Spehner, D. Villeval, and J.-P. Lecocq.** 1987. Similar genetic organization between a region of fowlpox virus DNA and the vaccinia virus *Hin*dIII J fragment despite divergent location of the thymidine kinase gene. Virology **160:**203–209.
58. **Durum, S. K., and J. J. Oppenheim.** 1993. Proinflammatory cytokines and immunity, p. 801–835. *In* W. E. Paul (ed.), Fundamental immunology, 3rd ed. Raven Press, New York, N.Y.
59. **Eidson, C. S., P. Villegas, and S. H. Kleven.** 1975. Efficacy of turkey herpesvirus vaccine when administered simultaneously with fowl pox vaccine. Poult. Sci. **54:**1975–1981.
60. **El-Sherbeini, M., and J. A. Clemas.** 1995. Cloning and characterization of *GNS1*: a *Saccharomyces cerevisiae* gene involved in synthesis of 1,3-β-glucan in vitro. J. Bacteriol. **177:**3227–3234.
61. **Endres, M. J., C. G. Garlisi, H. Xiao, L. Shan, and J. A. Hedrick.** 1999. The Kaposi's sarcoma-related herpesvirus (KSHV)-encoded chemokine vMIP-I is a specific agonist for the CC chemokine receptor (CCR)8. J. Exp. Med. **189:**1993–1998.
62. **Ensser, A., and B. Fleckenstein.** 1995. Alcelaphine herpesvirus type 1 has a semaphorin-like gene. J. Gen. Virol. **76:**1063–1067.
63. **Ewing, B., and P. Green.** 1998. Base-calling of automated sequencer traces using *Phred*. II. Error probabilities. Genome Res. **8:**186–194.
64. **Ewing, B., L. Hillier, M. C. Wendl, and P. Green.** 1998. Base-calling of automated sequencer traces using *Phred*. I. Accuracy assessment. Genome Res. **8:**175–185.
65. **Fatunmbi, O. O., and W. M. Reed.** 1996. Evaluation of a commercial quail pox vaccine (Bio-Pox Q) for the control of "variant" fowl poxvirus infections. Avian Dis. **40:**792–797.
66. **Fukuda, M., N. Hiraoka, and J.-C. Yeh.** 1999. C-type lectins and sialyl Lewis X oligosaccharides: versatile roles in cell-cell interaction. J. Cell Biol. **147:** 467–470.
67. **Gao, J.-L., and P. M. Murphy.** 1994. Human cytomegalovirus open reading frame *US28* encodes a functional β chemokine receptor. J. Biol. Chem. **269:** 28539–28542.
68. **Garaci, E., M. C. Caroleo, L. Aloe, S. Aquaro, M. Piacentini, N. Costa, A. Amendola, A. Micera, R. Calio, C. F. Perno, and R. Levi-Montalcini.** 1999. Nerve growth factor is an autocrine factor essential for the survival of macrophages infected with HIV. Proc. Natl. Acad. Sci. USA **96:**14013–14018.
69. **Garcia-Arranz, M., A. M. Maldonado, M. J. Mazon, and F. Portillo.** 1994. Transcriptional control of yeast plasma membrane H^+-ATPase by glucose. J. Biol. Chem. **269:**18076–18082.
70. **Gillard, S., D. Spehner, R. Drillien, and A. Kirn.** 1986. Localization and sequence of a vaccinia virus gene required for multiplication in human cells. Proc. Natl. Acad. Sci. USA **83:**5573–5577.
71. **Goebel, S. J., G. P. Johnson, M. E. Perkus, S. W. Davis, J. P. Winslow, and E. Paoletti.** 1990. The complete DNA sequence of vaccinia virus. Virology **179:**247–266.
72. **Gordon, D., C. Abajian, and P. Green.** 1998. *Consed*: a graphical tool for sequence finishing. Genome Res. **8:**192–202.
73. **Graham, K. A., A. S. Lalani, J. L. Macen, T. L. Ness, M. Barry, L.-Y. Liu, A. Lucas, I. Clark-Lewis, R. W. Moyer, and G. McFadden.** 1997. The T1/35kDa family of poxvirus-secreted proteins bind chemokines and modulate leukocyte influx into virus-infected tissues. Virology **229:**12–24.
74. **Halford, S., D. I. Wilson, S. C. Daw, C. Roberts, R. Wadey, S. Kamath, A. Wickremasinghe, J. Burn, J. Goodship, and M. G. Mattei.** 1993. Isolation of a gene expressed during early embryogenesis from the region of 22q11

commonly deleted in DiGeorge syndrome. Hum. Mol. Genet. **10:**1577–1582.
75. **Hearst, J. E.** 1995. The structure of photolyase: using photon energy for DNA repair. Science **268:**1858–1859.
76. **Hertig, C., B. E. H. Coupar, A. R. Gould, and D. B. Boyle.** 1997. Field and vaccine strains of fowlpox virus carry integrated sequences from the avian retrovirus, reticuloendotheliosis virus. Virology **235:**367–376.
77. **Hitchner, S. B.** 1981. Canary pox vaccination with live embryo-attenuated virus. Avian Dis. **25:**874–881.
78. **Holmgren, A.** 1985. Thioredoxin. Annu. Rev. Biochem. **54:**237–271.
79. **Hyodo, Y., K. Matsui, N. Nayashi, H. Tsutsui, S. I. Kashiwamura, H. Yamauchi, K. Hiroishi, K. Takeda, Y.-I. Tagawa, Y. Iwakura, N. Kayagaki, M. Kurimoto, H. Okamura, T. Hada, H. Yagita, S. Akira, K. Nakanishi, and K. Higashino.** 1999. IL-18 up-regulates perforin-mediated NK activity without increasing perforin messenger RNA expression by binding to constitutively expressed IL-18 receptor. J. Immunol. **162:**1662–1668.
80. **Ink, B. S., C. S. Gilbert, and G. I. Evan.** 1995. Delay of vaccinia virus-induced apoptosis in nonpermissive Chinese hamster ovary cells by the cowpox virus *CHOhr* and adenovirus *E1B 19K* genes. J. Virol. **69:**661–668.
81. **Joazeiro, C. A. P., S. S. Wing, H.-K. Huang, J. D. Leverson, T. Hunter, and Y.-C. Liu.** 1999. The tyrosine kinase negative regulator c-Cbl as a RING-type, E2-dependent ubiquitin-protein ligase. Science **286:**309–312.
82. **Kato, S., and W. Cutting.** 1959. A study of the inclusion bodies of rabbit myxoma and fibroma virus and a consideration of the relationship between all pox virus inclusion bodies. Stanford Med. Bull. **17:**34–45.
83. **Kent, S. J., A. Zhao, S. J. Best, J. D. Chandler, D. B. Boyle, and I. A. Ramshaw.** 1998. Enhanced T-cell immunogenicity and protective efficacy of a human immunodeficiency virus type 1 vaccine regimen consisting of consecutive priming with DNA and boosting with recombinant fowlpox virus. J. Virol. **72:**10180–10188.
84. **Kolodkin, A. L., D. J. Matthes, and C. S. Goodman.** 1993. The *semaphorin* genes encode a family of transmembrane and secreted growth cone guidance molecules. Cell **75:**1389–1399.
85. **Koonin, E. V.** 1993. A highly conserved sequence motif defining the family of MutT-related proteins from eubacteria, eukaryotes and viruses. Nucleic Acids Res. **21:**4847.
86. **Koonin, E. V., and T. G. Senkevich.** 1993. Fowlpox virus encodes a protein related to human deoxycytidine kinase: further evidence for independent acquisition of genes for enzymes of nucleotide metabolism by different viruses. Virus Genes **7:**289–295.
87. **Kovacs, G. R., and B. Moss.** 1996. The vaccinia virus H5R gene encodes late gene transcription factor 4: purification, cloning, and overexpression. J. Virol. **70:**6796–6802.
88. **Kozopas, K. M., T. Yang, H. L. Buchan, P. Zhou, and R. W. Craig.** 1993. *MCL1*, a gene expressed in programmed myeloid cell differentiation, has sequence similarity to *BCL2*. Proc. Natl. Acad. Sci. USA **90:**3516–3520.
89. **Krieser, R. J., and A. Eastman.** 1998. The cloning and expression of human deoxyribonuclease II. J. Biol. Chem. **273:**30909–30914.
90. **Kumar, S., and D. B. Boyle.** 1990. Mapping of a major early/late gene of fowlpox virus. Virus Res. **15:**175–185.
91. **Kumar, S., and D. B. Boyle.** 1990. A poxvirus bidirectional promoter element with early/late and late functions. Virology **179:**151–158.
92. **Kupke, T., S. Stevanovic, H.-G. Sahl, and F. Gotz.** 1992. Purification and characterization of EpiD, a flavoprotein involved in the biosynthesis of the lantibiotic epidermin. J. Bacteriol. **174:**5354–5361.
93. **Laidlaw, S. M., M. A. Anwar, W. Thomas, P. Green, K. Shaw, and M. A. Skinner.** 1998. Fowlpox virus encodes nonessential homologs of cellular alpha-SNAP, PC-1, and an orphan human homolog of a secreted nematode protein. J. Virol. **72:**6742–6751.
94. **Lalani, A. S., T. L. Ness, R. Singh, J. K. Harrison, B. T. Seet, D. J. Kelvin, G. McFadden, and R. W. Moyer.** 1998. Functional comparisons among members of the poxvirus T1/35kDa family of soluble CC-chemokine inhibitor glycoproteins. Virology **250:**173–184.
95. **Lange, C., T. Liehr, M. Goen, E. Gebhart, B. Fleckenstein, and A. Ensser.** 1998. New eukaryotic semaphorins with close homology to semaphorins of DNA viruses. Genomics **51:**340–350.
96. **Leong, K. H., A. J. Ramsay, D. B. Boyle, and I. A. Ramshaw.** 1994. Selective induction of immune responses by cytokines coexpressed in recombinant fowlpox virus. J. Virol. **68:**8125–8130.
97. **Levi-Montalcini, R.** 1987. The nerve growth factor 35 years later. Science **237:**1154–1162.
98. **Lewis, T., L. Zsak, T. G. Burrage, Z. Lu, G. F. Kutish, J. G. Neilan, and D. L. Rock.** 2000. An African swine fever virus *ERV1-ALR* homologue, *9GL*, affects virion maturation and viral growth in macrophages and viral virulence in swine. J. Virol. **74:**1275–1285.
99. **Liao, T.-H.** 1985. The subunit structure and active site sequence of porcine spleen deoxyribonuclease. J. Biol. Chem. **260:**10708–10713.
100. **Lin, E. Y., A. Orlofsky, M. S. Berger, and M. B. Prystowsky.** 1993. Characterization of A1, a novel hemopoietic-specific early-response gene with sequence similarity to *bcl-2*. J. Immunol. **151:**1979–1988.
101. **Lin, J.-H., A. Makris, C. McMahon, S. E. Bear, C. Patriotis, V. R. Prasad, R. Brent, E. A. Golemis, and P. N. Tsichlis.** 1999. The ankyrin repeat-containing adaptor protein Tvl-1 is a novel substrate and regulator of Raf-1. J. Biol. Chem. **274:**14706–14715.
102. **Lux, S. E., K. M. John, and V. Bennett.** 1990. Analysis of cDNA for human erythrocyte ankyrin indicates a repeated structure with homology to tissue-differentiation and cell-cycle control proteins. Nature **34:**36–42.
103. **Maa, J.-S., J. F. Rodriguez, and M. Esteban.** 1990. Structural and functional characterization of a cell surface binding protein of vaccinia virus. J. Biol. Chem. **265:**1569–1577.
104. **Marshall, J. S., K. Gomi, M. G. Blennerhassett, and J. Bienenstock.** 1999. Nerve growth factor modifies the expression of inflammatory cytokines by mast cells via a prostanoid-dependent mechanism. J. Immunol. **162:**4271–4276.
105. **Martin, K. H., D. W. Grosenbach, C. A. Franke, and D. E. Hruby.** 1997. Identification and analysis of three myristylated vaccinia virus late proteins. J. Virol. **71:**5218–5226.
106. **Massung, R. F., J. J. Esposito, L. I. Liu, J. Qi, T. R. Utterback, J. C. Knight, L. Aubin, T. E. Yuran, J. M. Parsons, V. N. Loparev, N. A. Selivanov, K. F. Cavallaro, A. R. Kerlavage, B. W. J. Mahy, and J. C. Venter.** 1993. Potential virulence determinants in terminal regions of variola smallpox virus genome. Nature **366:**748–751.
107. **Massung, R. F., V. Jayarama, and R. W. Moyer.** 1993. DNA sequence analysis of conserved and unique regions of swinepox virus: identification of genetic elements supporting phenotypic observations including a novel G protein-coupled receptor homologue. Virology **197:**511–528.
108. **Massung, R. F., L.-I. Liu, J. Qi, J. C. Knight, T. E. Yuran, A. R. Kerlavage, J. M. Parsons, J. C. Venter, and J. J. Esposito.** 1994. Analysis of the complete genome of smallpox variola major virus strain Bangladesh-1975. Virology **201:**215–240.
109. **Massung, R. F., G. McFadden, and R. W. Moyer.** 1992. Nucleotide sequence analysis of a unique near-terminal region of the tumorigenic poxvirus, Shope fibroma virus. J. Gen. Virol. **73:**2903–2911.
110. **McFadden, G., K. Graham, and M. Barry.** 1996. New strategies of immune modulation by DNA viruses. Transplant. Proc. **28:**2085–2088.
111. **McIntosh, A. A., and G. L. Smith.** 1996. Vaccinia virus glycoprotein A34R is required for infectivity of extracellular enveloped virus. J. Virol. **70:**272–281.
112. **McMillen, J. K., M. D. Cochran, D. E. Junker, D. N. Reddy, and D. M. Valencia.** 1994. The safe and effective use of fowlpox virus as a vector for poultry vaccines. Dev. Biol. Stand. **82:**137–145.
113. **Melchers, F., A. G. Rolink, and C. Schaniel.** 1999. The role of chemokines in regulating cell migration during humoral immune responses. Cell **99:**351–354.
114. **Micallef, M. J., T. Tanimoto, K. Torigoe, Y. Nishida, K. Kohno, H. Ikegami, and M. Kurimoto.** 1999. Simultaneous exposure to interleukin-18 and interleukin-10 in vitro synergistically augments murine spleen natural killer cell activity. Cancer Immunol. Immunother. **48:**109–117.
115. **Mockett, B., M. M. Binns, M. E. G. Boursnell, and M. A. Skinner.** 1992. Comparison of the locations of homologous fowlpox and vaccinia virus genes reveals major genome reorganization. J. Gen. Virol. **73:**2661–2668.
116. **Moore, J. B., and G. L. Smith.** 1992. Steroid hormone synthesis by a vaccinia enzyme: a new type of virus virulence factor. EMBO J. **11:**1973–1980.
117. **Morgan, A., and R. D. Burgoyne.** 1995. A role for soluble NSF attachment proteins (SNAPs) in regulated exocytosis in adrenal chromaffin cells. EMBO J. **14:**232–239.
118. **Moss, B.** 1996. Poxviridae: the viruses and their replication, p. 2637–2671. *In* B. N. Fields, D. M. Knipe, P. M. Howley, et al. (ed.), Fields virology. Lippincott-Raven, Philadelphia, Pa.
119. **Mossman, K., S. F. Lee, M. Barry, L. Boshkov, and G. McFadden.** 1996. Disruption of M-T5, a novel myxoma virus gene member of poxvirus host range superfamily, results in dramatic attenuation of myxomatosis in infected European rabbits. J. Virol. **70:**4394–4410.
120. **Muller, H. K., R. Wittek, W. Schaffner, D. Schumperli, A. Menna, and R. Wyler.** 1978. Comparison of five poxvirus genomes by analysis with restriction endonucleases *Hin*dIII, *Bam*I and *Eco*RI. J. Gen. Virol. **38:**135–147.
121. **Nazerian, K., R. L. Witter, L. F. Lee, and N. Yanagida.** 1996. Protection and synergism by recombinant fowl pox vaccines expressing genes from Marek's disease virus. Avian Dis. **40:**368–376.
122. **Neilan, J. G., M. V. Borca, Z. Lu, G. F. Kutish, S. B. Kleiboeker, C. Carrillo, L. Zsak, and D. L. Rock.** 1999. An African swine fever virus ORF with similarity to C-type lectins is non-essential for growth in swine macrophages *in vitro* and for virus virulence in domestic swine. J. Gen. Virol. **80:**2693–2697.
123. **Ogawa, R., J. G. Calvert, N. Yanagida, and K. Nazerian.** 1993. Insertional inactivation of a fowlpox virus homologue of the vaccinia virus F12L gene inhibits the release of enveloped virions. J. Gen. Virol. **74:**55–64.
124. **Omar, A. R., K. A. Schat, L. F. Lee, and H. D. Hunt.** 1998. Cytotoxic T lymphocyte response in chickens immunized with a recombinant fowlpox virus expressing Marek's disease herpesvirus glycoprotein B. Vet. Immunol. Immunopathol. **62:**73–82.
125. **Paoletti, E.** 1996. Applications of pox virus vectors to vaccination: an update. Proc. Natl. Acad. Sci. USA **93:**11349–11353.

126. **Perkus, M. E., S. J. Goebel, S. W. Davis, G. P. Johnson, K. Limbach, E. K. Norton, and E. Paoletti.** 1990. Vaccinia virus host range genes. Virology **179:**276–286.
127. **Pollitt, E., M. A. Skinner, and S. Heaphy.** 1998. Nucleotide sequence of the 4.3kbp BamHI-N fragment of fowlpox virus FP9. Virus Genes **17:**5–9.
128. **Purcell, D. A., J. K. Clarke, J. B. McFerran, and D. A. Hughes.** 1972. The morphogenesis of pigeonpox virus. J. Gen. Virol. **15:**79–83.
129. **Revardel, E., M. Bonneau, P. Durrens, and M. Aigle.** 1995. Characterization of a new gene family developing pleiotropic phenotypes upon mutation in *Saccharomyces cerevisiae*. Biochim. Biophys. Acta **1263:**261–265.
130. **Rodriguez, J. F., and G. L. Smith.** 1990. IPTG-dependent vaccinia virus: identification of a virus protein enabling virion envelopment by Golgi membrane and egress. Nucleic Acids Res. **18:**5347–5351.
131. **Rosel, J. L., P. L. Earl, J. P. Weir, and B. Moss.** 1986. Conserved TAAATG sequence at the transcriptional and translational initiation sites of vaccinia virus late genes deduced by structural and functional analysis of the *Hin*dIII H genome fragment. J. Virol. **60:**436–449.
132. **Ryan, J. C., and W. E. Seaman.** 1997. Divergent functions of lectin-like receptors on NK cells. Immunol. Rev. **155:**79–89.
133. **Sadasiv, E. C., P. W. Chang, and G. Gulka.** 1985. Morphogenesis of canary poxvirus and its entrance into inclusion bodies. Am. J. Vet. Res. **46:**529–535.
134. **Salzberg, S. L., A. L. Delcher, S. Kasif, and O. White.** 1998. Microbial gene identification using interpolated Markov models. Nucleic Acids Res. **26:**544–548.
135. **Sanger, F., S. Nicklen, and A. R. Coulson.** 1977. DNA sequencing with chain-terminating inhibitors. Proc. Natl. Acad. Sci. USA **74:**5463–5467.
136. **Sarma, D. K., and S. N. Sharma.** 1988. Comparative immunity of fowlpox virus vaccines. J. Vet. Med. Biol. **35:**19–23.
137. **Saurin, A. J., K. L. B. Borden, M. N. Boddy, and P. S. Freemont.** 1996. Does this have a familiar RING? Trends Biochem. Sci. **21:**208–214.
138. **Scholzen, T., C. A. Armstrong, N. W. Bunnett, T. A. Luger, J. E. Olerud, and J. C. Ansel.** 1998. Neuropeptides in the skin: interactions between the neuroendocrine and skin immune systems. Exp. Dermatol. **7:**81–96.
139. **Schoneberg, T., G. Schultz, and T. Gudermann.** 1999. Structural basis of G protein-coupled receptor function. Mol. Cell. Endocrinol. **151:**181–193.
140. **Sedgwick, S. G., and S. J. Smerdon.** 1999. The ankyrin repeat: a diversity of interactions on a common structural framework. Trends Biochem. Sci. **24:**311–316.
141. **Senkevich, T. G., J. J. Bugert, J. R. Sisler, E. V. Koonin, G. Darai, and B. Moss.** 1996. Genome sequence of a human tumorigenic poxvirus: prediction of specific host response-evasion genes. Science **273:**813–816.
142. **Senkevich, T. G., E. V. Koonin, J. J. Bugert, G. Darai, and B. Moss.** 1997. The genome of *Molluscum contagiosum* virus: analysis and comparison with other poxviruses. Virology **233:**19–42.
143. **Senkevich, T. G., E. V. Koonin, and R. M. L. Buller.** 1994. A poxvirus protein with a RING zinc finger motif is of crucial importance for virulence. Virology **198:**118–128.
144. **Senkevich, T. G., E. J. Wolffe, and R. M. L. Buller.** 1995. Ectromelia virus RING finger protein is localized in virus factories and is required for virus replication in macrophages. J. Virol. **69:**4103–4111.
145. **Shchelkunov, S. N., P. F. Safronov, A. V. Totmenin, N. A. Petrov, O. I. Ryazankina, V. V. Gutorov, and G. J. Kotwal.** 1998. The genome sequence analysis of the left and right species-specific terminal region of a cowpox virus strain reveals unique sequences and a cluster of intact ORFs for immunomodulatory and host range proteins. Virology **243:**432–460.
146. **Sheppard, M., W. Werner, E. Tsatas, R. McCoy, S. Prowse, and M. Johnson.** 1998. Fowl adenovirus recombinant expressing VP2 of infectious bursal disease virus induces protective immunity against bursal disease. Arch. Virol. **143:**915–930.
147. **Shida, H.** 1986. Nucleotide sequence of the vaccinia virus hemagglutinin gene. Virology **150:**451–462.
148. **Shisler, J. L., T. G. Senkevich, M. L. Berry, and B. Moss.** 1998. Ultraviolet-induced cell death blocked by a selenoprotein from a human dermatotropic poxvirus. Science **279:**102–105.
149. **Shors, T., J. G. Keck, and B. Moss.** 1999. Down regulation of gene expression by the vaccinia virus D10 protein. J. Virol. **73:**791–796.
150. **Skinner, M. A., J. B. Moore, M. M. Binns, G. L. Smith, and M. E. G. Boursnell.** 1994. Deletion of fowlpox virus homologues of vaccinia virus genes between the 3 β-hydroxysteroid dehydrogenase (A44L) and DNA ligase (A50R) genes. J. Gen. Virol. **75:**2495–2498.
151. **Smith, C. A., T. Davis Smith, P. J. Smolak, D. Friend, H. Hagen, M. Gerhart, L. Park, D. J. Pickup, D. Torrance, K. Mohler, K. Schooley, and R. G. Goodwin.** 1997. Poxvirus genomes encode a secreted, soluble protein that preferentially inhibits β chemokine activity yet lacks sequence homology to known chemokine receptors. Virology **236:**316–327.
152. **Smith, G. L., and Y. S. Chan.** 1991. Two vaccinia virus proteins structurally related to the interleukin-1 receptor and the immunoglobulin superfamily. J. Gen. Virol. **72:**511–518.
153. **Spehner, D., S. Gillard, R. Drillien, and A. Kirn.** 1988. A cowpox virus gene required for multiplication in Chinese hamster ovary cells. J. Virol. **62:**1297–1304.
154. **Spriggs, M. K.** 1999. Shared resources between the neural and immune systems: semaphorins join the ranks. Curr. Opin. Immunol. **11:**387–391.
155. **Staden, R., and A. D. McLachlan.** 1982. Codon preference and its use in identifying protein coding regions in long DNA sequences. Nucleic Acids Res. **10:**141–156.
156. **Staden, R.** 1982. An interactive graphics program for comparing and aligning nucleic acid and amino acid sequences. Nucleic Acids Res. **10:**2951–2961.
157. **Stadtman, T. C.** 1996. Selenocysteine. Annu. Rev. Biochem. **65:**83–100.
158. **Stenbeck, G.** 1998. Molecules in focus soluble NSF-attachment proteins. Int. J. Biochem. Cell Biol. **30:**573–577.
159. **Sutter, G., A. Ramsey-Ewing, R. Rosales, and B. Moss.** 1994. Stable expression of the vaccinia virus K1L gene in rabbit cells complements the host range defect of a vaccinia virus mutant. J. Virol. **68:**4109–4116.
160. **Sutton, G. G., O. White, M. D. Adams, and A. R. Kerlavage.** 1995. TIGR assembler: a new tool for assembling large shotgun sequencing projects. Genome Sci. Tech. **1:**9–19.
161. **Takeda, K., H. Tsutsui, T. Yoshimoto, O. Adachi, N. Yoshida, T. Kishimoto, H. Okamura, K. Nakanishi, and S. Akira.** 1998. Defective NK cell activity and Th1 response in IL-18-deficient mice. Immunity **8:**383–390.
162. **Tanaka-Kataoka, M., T. Kunikata, S. Takayama, K. Iwaki, K. Ohashi, M. Ikeda, and M. Kurimoto.** 1999. In vivo antiviral effect of interleukin 18 in a mouse model of vaccinia virus infection. Cytokine **11:**593–599.
163. **Tartaglia, J., J. Winslow, S. Goebel, G. P. Johnson, J. Taylor, and E. Paoletti.** 1990. Nucleotide sequence analysis of a 10.5 kbp *Hin*dIII fragment of fowlpox virus: relatedness to the central portion of the vaccinia virus *Hin*dIII D region. J. Gen. Virol. **71:**1517–1524.
164. **Taylor, J., and E. Paoletti.** 1988. Fowlpox virus as a vector in non-avian species. Vaccine **6:**466–468.
165. **Taylor, J., R. Weinberg, J. Tartaglia, C. Richardson, G. Alkhatib, D. Briedis, M. Appel, E. Norton, and E. Paoletti.** 1992. Nonreplicating viral vectors as potential vaccines: recombinant canarypox virus expressing measles virus fusion (F) and hemagglutinin (HA) glycoproteins. Virology **187:**321–328.
166. **Tomley, F., M. Binns, J. Campbell, and M. Boursnell.** 1988. Sequence analysis of an 11.2 kilobase, near-terminal, *Bam*HI fragment of fowlpox virus. J. Gen. Virol. **69:**1025–1040.
167. **Torcia, M., L. Bracci-Laudiero, M. Lucibello, L. Nencioni, D. Labardi, A. Rubartelli, F. Cozzolino, L. Aloe, and E. Garaci.** 1996. Nerve growth factor is an autocrine survival factor for memory B lymphocytes. Cell **85:**345–356.
168. **Torriglia, A., P. Perani, J. Y. Brossas, E. Chaudun, J. Treton, Y. Courtois, and M. F. Counis.** 1998. L-DNase II, a molecule that links proteases and endonucleases in apoptosis, derives from the ubiquitous serpin leukocyte elastase inhibitor. Mol. Cell. Biol. **18:**3612–3619.
169. **Tripathy, D. N.** 1991. Pox, p. 583–596. *In* B. W. Calnek, H. J. Barnes, C. W. Beard, W. M. Reid, and H. W. Yoder, Jr. (ed.), Diseases of poultry, 9th ed. Iowa State University Press, Ames.
170. **Tripathy, D. N., and C. H. Cunningham.** 1984. Avian pox, p. 524–534. *In* M. S. Hofstad, H. J. Barnes, W. M. Reid, and H. W. J. Yoder (ed.), Diseases of poultry, 8th ed. Iowa State University Press, Ames.
171. **Tripathy, D. N., and L. E. Hanson.** 1978. Pathogenesis of fowlpox in laying hens. Avian Dis. **22:**259–265.
172. **Turner, P. C., and R. W. Moyer.** 1998. Control of apoptosis by poxviruses. Semin. Virol. **8:**453–469.
173. **VanSlyke, J. K., S. S. Whitehead, E. M. Wilson, and D. E. Hruby.** 1991. The multistep proteolytic maturation pathway utilized by vaccinia virus P4a protein: a degenerate conserved cleavage motif within core proteins. Virology **183:**467–478.
174. **Wang, C.-C., S.-C. Lu, H.-L. Chen, and T.-H. Liao.** 1998. Porcine spleen deoxyribonuclease II. J. Biol. Chem. **273:**17192–17198.
175. **Weis, W. I., M. E. Taylor, and K. Drickamer.** 1998. The C-type lectin superfamily in the immune system. Immunol. Rev. **163:**19–34.
176. **Welker, P., J. Grabbe, A. Grutzkau, and B. M. Henz.** 1998. Effects of nerve growth factor (NGF) and other fibroblast-derived growth factors on immature human mast cells (HMC-1). Immunology **94:**310–317.
177. **Weskamp, G., and U. Otten.** 1987. An enzyme-linked immunoassay for nerve growth factor (NGF): a tool for studying regulatory mechanisms involved in NGF production in brain and in peripheral tissues. J. Neurochem. **48:**1779–1786.
178. **Whitehead, S. S., and D. E. Hruby.** 1994. A transcriptionally controlled *trans*-processing assay: putative identification of a vaccinia virus-encoded proteinase which cleaves precursor protein P25K. J. Virol. **68:**7603–7608.
179. **Wilcock, D., S. A. Duncan, P. Traktman, W.-H. Zhang, and G. L. Smith.** 1999. The vaccinia virus A40R gene product is a nonstructural, type II membrane glycoprotein that is expressed at the cell surface. J. Gen. Virol. **80:**2137–2148.
180. **Williams, A. F., and A. N. Barclay.** 1988. The immunoglobulin superfamily—domains for cell surface recognition. Annu. Rev. Immunol. **6:**381–405.
181. **Wolffe, E. J., S. N. Isaacs, and B. Moss.** 1993. Deletion of the vaccinia virus B5R gene encoding a 42-kilodalton membrane glycoprotein inhibits extracellular virus envelope formation and dissemination. J. Virol. **67:**4732–4741.
182. **Woodward, H., and D. C. Tudor.** 1973. The immunizing effect of commercial pigeon pox vaccines on pigeons. Poult. Sci. **52:**1463–1468.

183. **Woolf, C. J., Q. P. Ma, A. Allchorne, and S. Poole.** 1996. Peripheral cell types contributing to the hyperalgesic action of nerve growth factor in inflammation. J. Neurosci. **16:**2716–2723.
184. **Xiang, Y., and B. Moss.** 1999. Identification of human and mouse homologs of the MC51L-53L-54L family of secreted glycoproteins encoded by the *Molluscum contagiosum* poxvirus. Virology **257:**297–302.
185. **Xiang, Y., and B. Moss.** 1999. IL-18 binding and inhibition of interferon γ induction by human poxvirus encoded proteins. Proc. Natl. Acad. Sci. USA **96:**11537–11542.
186. **Xiang, Y., D. A. Simpson, J. Spiegel, A. Zhou, R. H. Silverman, and R. C. Condit.** 1998. The vaccinia virus A18R DNA helicase is a postreplicative negative transcription elongation factor. J. Virol. **72:**7012–7023.
187. **Yanagida, N., R. Ogawa, Y. Li, L. F. Lee, and K. Nazerian.** 1992. Recombinant fowlpox viruses expressing the glycoprotein B homolog and the pp38 gene of Marek's disease virus. J. Virol. **66:**1402–1408.
188. **Yasui, A., A. P. M. Eker, S. Yasuhira, H. Yajima, T. Kobayashi, M. Takao, and A. Oikawa.** 1994. A new class of DNA photolyases present in various organisms including aplacental mammals. EMBO J. **13:**6143–6151.
189. **Yuen, L., and B. Moss.** 1987. Oligonucleotide sequence signaling transcriptional termination of vaccinia virus early genes. Proc. Natl. Acad. Sci. USA **84:**6417–6421.
190. **Zajac, P., D. Spehner, and R. Drillien.** 1995. The vaccinia virus J5L open reading frame encodes a polypeptide expressed late during infection and required for viral multiplication. Virus Res. **37:**163–173.
191. **Zantinge, J. L., P. J. Krell, J. B. Derbyshire, and E. Nagy.** 1996. Partial transcriptional mapping of the fowlpox virus genome and analysis of the *Eco*RI L fragment. J. Gen. Virol. **77:**603–614.