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(54) Title: AFRICAN SWINE FEVER VACCINE

FIGURE 4

Table 1

Experiment #	AR000737		AR000742		AR000750		AR000858				
	1	2		3		4					
Experiment Pool	Pool A	Pool A	Pool B	Pool A	Pool B	Pool 1	Pool 2	Pool 3	Pool 4	Pool 5	
	A151R	B602L	B602L	B602L		B602L	B602L	B602L	B602L		
	B646L	B646L	I329L	B646L	B646L	B646L		B646L		B646L	
	CP204L	CP204L	MGF505-4R	CP204L	CP204L	CP204L		CP204L		CP204L	
	CP530R, C129R	E183L	MGF360-11L	E183L		E183L	E183L	E183L	E183L	E183L	
	M448R, L8L	E199L	EP364R	E199L	E199L	E199L	E199L			E199L	
	I73R, I215L	EP153R	EP153R	EP153R		EP153R	EP153R	EP153R	EP153R		
	E146L, MGF110-5L	F317L	F317L	F317L	F317L	F317L	F317L			F317L	
	MGF110-4L	MGF505-5R	MGF505-5R	MGF505-5R	MGF505-5R	MGF505-5R	MGF505-5R			MGF505-5R	
Vectors used (Prime-Boost)	Ad-MVA	Ad-MVA	Ad-MVA	Ad-MVA	Ad-MVA	Ad-MVA	Ad-Ad	Ad-Ad	Ad-MVA	Ad-Ad	
Protection	0/6	2/6	0/6	6/6	0/6	2/5	4/5	3/5	3/5	0/5	

(57) Abstract: The present invention provides an African swine fever virus (ASFV) subunit vaccine which comprises: (i) one or more recombinant polynucleotides which encode polypeptides shown as SEQ ID NO: 1, 2 and 3 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 1, 2 or 3; wherein the total number of different ASFV polypeptides encoded by the one or more recombinant polynucleotides is 10 or fewer; or (ii) recombinant polypeptides shown as SEQ ID NO: 1, 2 and 3 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 1, 2 and 3; wherein vaccine comprises 10 or fewer different ASFV polypeptides.

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FIELD OF THE INVENTION

The present invention relates to a vaccine for the treatment and/or prevention of African swine fever.

5 BACKGROUND TO THE INVENTION

African swine fever (ASF) is a devastating haemorrhagic disease of domestic pigs caused by a double-stranded DNA virus, African swine fever virus (ASFV). ASFV is the only member of the *Asfarviridae* family which replicates predominantly in the cytoplasm of cells. Virulent strains of ASFV can kill domestic pigs within about 5-14 days of infection with a mortality rate approaching
10 100%.

There is currently no treatment for ASF. Prevention in countries outside Africa has been attempted on a national basis by restrictions on incoming pigs and pork products, compulsory boiling of waste animal products under licence before feeding to pigs and the application of a slaughter policy when the disease is diagnosed. Prevention in Africa of spread from wildlife is
15 based on measures to keep warthogs and materials contaminated by warthogs away from the herd. Spread in domestic pig populations is based on good biosecurity measures and implementation of quarantine and slaughter of pigs on affected farms.

To date, no effective attenuated or inactivated virus vaccines have been developed (see <http://www.thepigsite.com/pighealth/article/441/african-swine-fever-asf>).

20 Experimental protection against lethal challenge is possible by using a low virulent ASFV strain OURT88/3 that was recovered from a soft tick. OURT88/3 is a non-pathogenic isolate of ASFV from Portugal. Previous infection with ASFV OURT88/3 has been shown to confer protection against challenge with related virulent viruses (Boinas et al (2004) J Gen Virol 85:2177-2187; Oura et al (2005) J. Gen. Virol. 86:2445-2450). In addition some gene-deleted viruses are also
25 effective in inducing protection against challenge (eg Benin Δ MGF and Benin Δ DP148R).

Although these viruses have been shown to induce a protective immune response in certain animals, this effect does not appear to be universal. Immunisation appears to be ineffective in protecting some pigs from subsequent challenge. It can also be associated with the induction of adverse clinical responses, such as fever or joint swelling, in some pigs.

30 The ability of subunit compositions comprising ASFV polypeptides to induce protection against ASFV has also been investigated. However, these studies have determined that the ability of an ASFV polypeptide to induce an immunogenic response does not necessarily predict an

ability to induce protection to ASFV challenge (see Neilan *et al.*; Virology; 319(2); 337-342; 2004). Further, ASFV polypeptides may only induce protection when used in specific combinations. For example, Neilan *et al.* (as above) reported that a combination of p30, p54, p72 and p22 was not sufficient to induce protection. In contrast, Argilaguet *et al.* (Antiviral Res; 98(1); 61-65; 2013) reported that a combination of p30, p54 and EP402R gave partial protection against ASFV in DNA vaccination experiments.

There is thus a need for alternative measures to control ASFV infection and prevent spread of the disease.

10 SUMMARY OF ASPECTS OF THE INVENTION

The present inventors have determined a minimal combination of three ASFV polypeptides that are capable of inducing repeatable protection against ASFV when administered prior to ASFV challenge. The inventors have determined that protection is not induced when any one of the three ASFV polypeptides is absent. Further, the inventors have demonstrated that this minimal combination of three ASFV polypeptides may be combined with additional ASFV polypeptides to provide improved vaccine compositions for inducing reliable protection against ASFV challenge and treatment and/or prevention of ASF.

Thus, in a first aspect, the present invention provides an ASF virus subunit vaccine which comprises: (i) one or more recombinant polynucleotides which encode polypeptides shown as SEQ ID NO: 1, 2 and 3 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 1, 2 or 3 or an immunogenic fragment thereof; wherein the total number of different ASF virus (ASFV) polypeptides encoded by the one or more recombinant polynucleotides is 10 or fewer; or (ii) recombinant polypeptides shown as SEQ ID NO: 1, 2 and 3 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 1, 2 and 3 or an immunogenic fragment thereof; wherein vaccine comprises 10 or fewer different ASFV polypeptides.

The term subunit vaccine is used herein to refer to a vaccine which comprises individual polypeptides or polynucleotides encoding said polypeptides; in contrast to vaccines which comprise whole virus particles (such a live, attenuated virus and dead virus).

The number of different ASFV polypeptides encoded by the one or more recombinant polynucleotides may be 9 or fewer, 8 or fewer, 7 or fewer, or 6 or fewer polypeptides ASFV polypeptides; or the vaccine may comprise 9 or fewer, 8 or fewer, 7 or fewer, or 6 or fewer.

The vaccine may comprise: (i) one or more further polynucleotides encoding an ASFV polypeptide selected from SEQ ID NO: 4-8 or an immunogenic fragment thereof, or a variant

with at least 70% sequence identity to one of SEQ ID NO: 4-8 or an immunogenic fragment thereof; or (ii) one or more further polypeptides selected from SEQ ID NO: 4-8 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 4-8 or an immunogenic fragment thereof.

- 5 The vaccine may comprise: a) recombinant polynucleotides which encode polypeptides comprising SEQ ID NO: 1-8 or an immunogenic fragment thereof or a variant with at least 70% sequence identity to SEQ ID NO: 1-8 or immunogenic fragments thereof; or recombinant polypeptides comprising SEQ ID NO: 1-8 or an immunogenic fragment thereof or variants with at least 70% sequence identity to SEQ ID NO: 1-8 or immunogenic fragments thereof; (b)
- 10 recombinant polynucleotides which encode polypeptides comprising SEQ ID NO: 1-3 and 6-8 or an immunogenic fragment thereof or variants with at least 70% sequence identity to SEQ ID NO: 1-3 and 6-8 or immunogenic fragments thereof; or recombinant polypeptides comprising SEQ ID NO: 1-3 and 6-8 or an immunogenic fragment thereof or variants with at least 70% sequence identity to SEQ ID NO: 1-3 and 6-8 or immunogenic fragments thereof; or (c)
- 15 recombinant polynucleotides which encode polypeptides comprising SEQ ID NO: 1-5 or an immunogenic fragment thereof or variants with at least 70% sequence identity to SEQ ID NO: 1-5 or immunogenic fragments thereof; or recombinant polypeptides comprising SEQ ID NO: 1-5 or an immunogenic fragment thereof or variants with at least 70% sequence identity to SEQ ID NO: 1-5 or immunogenic fragments thereof.
- 20 The recombinant polynucleotides may comprise SEQ ID NO: 9, 10 and 11 or a variant thereof with at least 70% sequence identity.

The further polynucleotides may comprise one or more of SEQ ID NO: 12-16 or a variant thereof with at least 70% sequence identity.

- 25 The polynucleotide may be present in a vector. The vector may be selected from an adenovirus, a modified vaccinia Ankara vector and a pseudorabies virus vector.

Each of the recombinant polynucleotides may be provided in the same vector.

In a further aspect the present invention provides a vaccine according to the first aspect of the invention for use in treating and/or preventing African swine fever in a subject.

- 30 The present invention further relates to a method for treating and/or preventing African swine fever in a subject which comprises administering a therapeutically effective amount of a vaccine according to the invention to the subject.

In another aspect the present invention relates to the use of a recombinant polynucleotide, vector or recombinant polypeptide as defined in the first aspect of the invention in the manufacture of a medicament for the treatment and/or prevention of African swine fever in a subject.

- 5 The subject may be a swine subject. Suitably, the subject may be a domestic pig.

The vaccine may be administered according to a prime-boost procedure. For example, the priming composition may comprise one or more adenovirus vectors and the boosting composition may comprise one or more modified vaccinia Ankara vectors.

- 10 The vaccine may be administered by oral, intravenous, intramuscular, subcutaneous, intranasal or intradermal administration.

DESCRIPTION OF THE FIGURES

Figure 1 – Scheme of Adenovirus-MVA prime/boost protocol

Figure 2 – Representative amino acid sequences for ASFV polypeptides for use in the present invention (SEQ ID NO: 1-8 and 17)

- 15 **Figure 3** – Representative nucleic acid sequences encoding ASFV polypeptides for use in the present invention (SEQ ID NO: 9-16)

Figure 4 – Table 1

DETAILED DESCRIPTION OF THE INVENTION

AFRICAN SWINE FEVER VIRUS (ASFV)

- 20 The present inventors have determined a minimal combination of three ASFV polypeptides that are capable of inducing repeatable protection against ASFV.

- African swine fever virus (ASFV) is the causative agent of African swine fever (ASF). The virus causes a haemorrhagic fever with high mortality rates in pigs, but persistently infects its natural hosts, warthogs, bushpigs with no disease signs. It also infects soft ticks of the *Ornithodoros* genus, which are thought to be used as a vector.
- 25

ASFV replicates in the cytoplasm of infected cells, and is the only member of the *Asfarviridae* family to do so. ASFV is endemic to sub-Saharan Africa and exists in the wild through a cycle of infection between ticks and wild pigs, bushpigs and warthogs. ASFV was first described after

European settlers brought pigs into areas endemic with ASFV and, as such, is an example of an 'emerging infection'.

ASFV is a large, icosahedral, double-stranded DNA virus with a linear genome containing at least 150 genes. The number of genes differs slightly between different isolates of the virus.

- 5 ASFV has similarities to the other large DNA viruses, e.g., poxvirus, iridovirus and mimivirus. In common with other viral haemorrhagic fevers, the main target cells for replication are those of monocyte, macrophage lineage.

- 10 Based on sequence variation in the C-terminal region of the B646L gene encoding the major capsid protein p72, 24 ASFV genotypes (I-XXIX) have been identified. All ASFV p72 genotypes have been circulating in eastern and southern Africa. Genotype I is been circulating in Sardinia and western Africa. Genotype II is circulating in Europe, China, Vietnam and Cambodia. Genotype VIII is confined to four East African countries.

Examples of strains from some of the genotypes are given below. Strains shown in bold were used in the identity analysis shown in Table 2.

- 15 Genotype I : **OURT88/3**; Brazil/79; Lisbon/60; BA715; **ASFV-Pret**; **Benin 97/1**; IC/1/96; IC/576; CAM/82; Madrid/62; Malta/78; ZAR85; Katange63; Togo; Dakar59; OURT88/1; BEN/1/97; Dom_Rep; VAL/76; IC/2/96; Awoshie/99; NIG/1/99; NIG/1/98; ANG/70; BEL/85; SPEC120; Lisbon/57; **ASFV-Warm**; GHA/1/00; GAM/1/00; Ghana; HOL/86; NAM/1/80; NUR/90/1; CAM/4/85; **ASFV-Teng**; Tengani; **ASFV-E75**.

- 20 Genotype II: **Georgia 2007/1**, Belgium 2018/1, China/2018/AnhuiXCGQ

Genotype III: BOT 1/99

Genotype IV: **ASFV-Warthog**; RSA/1/99/W

Genotype VI: MOZ 94/1

Genotype VII: VICT/90/1; **ASFV-Mku**; RSA/1/98

- 25 Genotype VIII: NDA/1/90; KAL88/1; ZAM/2/84; JON89/13; KAV89/1; DEZda; **Malawi LIL 20/1**

Genotype IX: UGA/1/95; **Ken06.Bus**

Genotype X: BUR/1/84; BUR/2/84; BUR/90/1; UGA/3/95; TAN/Kwh12; Hindell; **ASFV-Ken**; Virulent Uganda 65; **Ken05/Tk1**.

The present inventors have determined that the composition comprising a minimal combination of ASFV polypeptides SEQ ID NO: 1-3 are capable of inducing protection against ASFV challenge.

Table 2

Gene Name	Alternative Gene Names	Amino acid SEQ ID NO:	cDNA SEQ ID NO:
B602L	ASFV chaperone	1	9
E183L	p54/j13L	2	10
EP153R	ASFV lectin	3	11
B646L	p72	4	12
CP204L	CP196L / p30	5	13
E199L		6	14
F317L		7	15
MGF505-5R		8	16

5 Representative amino acid sequences of polypeptides in Table 2 are shown in Figure 2 (SEQ ID NO: 1-8 and 17).

Representative nucleic acid sequences encoding the polypeptides in Table 2 are shown as SEQ ID NO: 9-16 in Figure 3.

10 POLYPEPTIDE

The term “polypeptide” is used in the normal sense to mean a series of residues, typically L-amino acids, connected one to the other typically by peptide bonds between the α-amino and carboxyl groups of adjacent amino acids. The term is synonymous with "protein".

15 The term “recombinant polypeptide” is used to mean that the polypeptide is isolated from its natural environment, for example, extracted from or produced outside of an ASFV. In particular, the recombinant polypeptide is not provided in the context of an ASFV and is expressed from a recombinant nucleic acid, i.e. DNA or RNA that is created artificially.

The vaccine of the present invention may comprise a combination of ASFV polypeptides, as defined herein.

20 The present vaccine may comprise recombinant polypeptides shown as SEQ ID NO: 1, 2 and 3 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 1, 2 and 3 or an immunogenic fragment thereof.

In any embodiment of the present invention, the polypeptide of SEQ ID NO: 3 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ

ID NO: 3 or an immunogenic fragment thereof may be substituted with SEQ ID NO: 17 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 17 or an immunogenic fragment thereof.

SEQ ID NO: 17 is a representative EP153R sequence from a Genotype II strain.

5 Suitably, the variant may comprise an amino acid sequence which is at least 70, 80, 85, 90, 95, 98 or 99% identical to one of SEQ ID NO: 1, 2 or 3; or an immunogenic fragment thereof.

Suitably, the variant may comprise an amino acid sequence which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 1 or an immunogenic fragment thereof.

10 Suitably, the variant may comprise an amino acid sequence which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 2 or an immunogenic fragment thereof.

Suitably, the variant may comprise an amino acid sequence which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 3 or an immunogenic fragment thereof.

15 The present vaccine may comprise recombinant polypeptides shown as SEQ ID NO: 1, 2 and 3; or a variant with at least 70% sequence identity to one of SEQ ID NO: 1, 2 and/or 3. In one embodiment, the vaccine may consist of recombinant polypeptides shown as SEQ ID NO: 1, 2 and 3; or a variant with at least 70% sequence identity to one of SEQ ID NO: 1, 2 and/or 3.

In one embodiment, the recombinant polypeptides may consist of amino acid sequences shown as SEQ ID NO: 1, 2 and 3 or a variant thereof which shares at least 70% sequence identity

20 The vaccine may further comprise one or more polypeptides selected from SEQ ID NO: 4-8 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 4-8 or an immunogenic fragment thereof.

Suitably, the variant may comprise an amino acid sequence which is at least 70, 80, 85, 90, 95, 98 or 99% identical to one of SEQ ID NO: 4-8; or an immunogenic fragment thereof.

25 Suitably, the variant may comprise an amino acid sequence which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 4 or an immunogenic fragment thereof.

Suitably, the variant may comprise an amino acid sequence which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 5 or an immunogenic fragment thereof.

Suitably, the variant may comprise an amino acid sequence which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 6 or an immunogenic fragment thereof.

Suitably, the variant may comprise an amino acid sequence which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 7 or an immunogenic fragment thereof.

Suitably, the variant may comprise an amino acid sequence which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 8 or an immunogenic fragment thereof.

- 5 The vaccine may further comprise one or more polypeptides selected from SEQ ID NO: 4-8 or a variant with at least 70% sequence identity to one of SEQ ID NO: 4-8.

The vaccine may comprise at least 2, at least 3, at least 4 or at least 5 polypeptides selected from SEQ ID NO: 4-8 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 4-8.

- 10 The vaccine may comprise 2, 3, 4 or 5 polypeptides selected from SEQ ID NO: 4-8 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 4-8 or an immunogenic fragment thereof.

Suitably, the vaccine may comprise recombinant polypeptides comprising SEQ ID NO: 1-8 or an immunogenic fragment thereof; or variants with at least 70% sequence identity to SEQ ID NO:

- 15 1-8 or immunogenic fragments thereof.

Suitably, the vaccine may comprise recombinant polypeptides comprising SEQ ID NO: 1-3 and 6-8 or an immunogenic fragment thereof; or variants with at least 70% sequence identity to SEQ ID NO: 1-3 and 6-8 or immunogenic fragments thereof.

Suitably, the vaccine may comprise recombinant polypeptides comprising SEQ ID NO: 1-5 or an immunogenic fragment thereof; or variants with at least 70% sequence identity to SEQ ID NO:

- 20 1-5 or immunogenic fragments thereof.

VARIANT

Although a variant can be considered in terms of similarity (i.e. amino acid residues having similar chemical properties/functions), in the context of the present invention it is preferred to

- 25 express a variant in terms of sequence identity.

Sequence comparisons can be conducted by eye, or more usually, with the aid of readily available sequence comparison programs. These publicly and commercially available computer programs can calculate sequence identity between two or more sequences.

Sequence identity may be calculated over contiguous sequences, i.e. one sequence is aligned

- 30 with the other sequence and each amino acid in one sequence directly compared with the

corresponding amino acid in the other sequence, one residue at a time. This is called an “ungapped” alignment. Typically, such ungapped alignments are performed only over a relatively short number of residues (for example less than 50 contiguous amino acids).

5 Although this is a very simple and consistent method, it fails to take into consideration that, for example, in an otherwise identical pair of sequences, one insertion or deletion will cause the following amino acid residues to be put out of alignment, thus potentially resulting in a large reduction in % homology when a global alignment is performed. Consequently, most sequence comparison methods are designed to produce optimal alignments that take into consideration possible insertions and deletions without penalising unduly the overall homology score. This is
10 achieved by inserting “gaps” in the sequence alignment to try to maximise local homology.

However, these more complex methods assign “gap penalties” to each gap that occurs in the alignment so that, for the same number of identical amino acids, a sequence alignment with as few gaps as possible - reflecting higher relatedness between the two compared sequences - will achieve a higher score than one with many gaps. “Affine gap costs” are typically used that
15 charge a relatively high cost for the existence of a gap and a smaller penalty for each subsequent residue in the gap. This is the most commonly used gap scoring system. High gap penalties will of course produce optimised alignments with fewer gaps. Most alignment programs allow the gap penalties to be modified. However, it is preferred to use the default values when using such software for sequence comparisons. For example when using the GCG
20 Wisconsin Bestfit package (see below) the default gap penalty for amino acid sequences is -12 for a gap and -4 for each extension.

Calculation of maximum % sequence identity therefore firstly requires the production of an optimal alignment, taking into consideration gap penalties. A suitable computer program for carrying out such an alignment is the GCG Wisconsin Bestfit package (University of Wisconsin,
25 U.S.A; Devereux *et al.*, 1984, Nucleic Acids Research 12:387). Examples of other software than can perform sequence comparisons include, but are not limited to, the BLAST package (see Ausubel *et al.*, 1999 *ibid* – Chapter 18), FASTA (Atschul *et al.*, 1990, J. Mol. Biol., 403-410) and the GENWORKS suite of comparison tools. Both BLAST and FASTA are available for offline and online searching (see Ausubel *et al.*, 1999 *ibid*, pages 7-58 to 7-60). However it is preferred
30 to use the GCG Bestfit program.

In one embodiment, the sequence identity is determined across the entirety of the sequence selected from SEQ ID NO: 1-8 or SEQ ID NO: 9-16. In one embodiment, the sequence identity is determined across the entirety of the candidate sequence being compared to a sequence selected from SEQ ID NO: 1-8 or SEQ ID NO: 9-16.

Although the final sequence identity can be measured in terms of identity, the alignment process itself is typically not based on an all-or-nothing pair comparison. Instead, a scaled similarity score matrix is generally used that assigns scores to each pairwise comparison based on chemical similarity or evolutionary distance. An example of such a matrix commonly used is the BLOSUM62 matrix - the default matrix for the BLAST suite of programs. GCG Wisconsin programs generally use either the public default values or a custom symbol comparison table if supplied (see user manual for further details). It is preferred to use the public default values for the GCG package, or in the case of other software, the default matrix, such as BLOSUM62.

Once the software has produced an optimal alignment, it is possible to calculate % sequence identity. The software typically does this as part of the sequence comparison and generates a numerical result.

The term “variant” according to the present invention includes any substitution of, variation of, modification of, replacement of, deletion of or addition of one (or more) amino acids from or to the sequence providing the resultant amino acid sequence retains substantially the same activity as the unmodified sequence.

Conservative substitutions may be made, for example according to the Table below. Amino acids in the same block in the second column and preferably in the same line in the third column may be substituted for each other:

ALIPHATIC	Non-polar	G A P
		I L V
	Polar - uncharged	C S T M
		N Q
	Polar - charged	D E
		K R
AROMATIC		H F W Y

With respect to function, the variant should be capable of inducing an immune response. By way of example, the induction of an immune response may be determined by the demonstration of a recall response in peripheral immune cells or splenocytes isolated from a subject previously immunised with the polypeptide or immunogenic fragment thereof. For instance, a recall response may be demonstrated by interferon production (e.g. IFN γ) and/or a proliferative response following an *in vitro* challenge with an antigen or polypeptide previous used to immunise a subject. Preferably, the variant should be capable of inducing a protective immune response in a subject, against subsequent challenge with ASFV when administered in

combination with a polypeptide selected from SEQ ID NO: 1-3 or a variant or immunogenic fragment thereof as described herein.

The polypeptide may comprise an immunogenic fragment of an amino acid sequence selected from SEQ ID NO: 1-8 or a variant thereof which shares at least 70% sequence identity.

- 5 By immunogenic fragment, it is meant a portion or a part of SEQ ID NO: 1-8 which is capable of inducing an immune response. Preferably, the immunogenic fragment should be capable of inducing a protective immune response in a subject against subsequent challenge with ASFV when administered in combination with a polypeptide selected from SEQ ID NO: 1-3 or a variant or immunogenic fragment thereof as described herein
- 10 Immunogenic fragments of SEQ ID NO: 1-8, or a variant thereof, may for example be between 8 and 200 amino acids, for example 8 to 150 amino acids, 8 to 100 amino acids, 8 to 75 amino acids, 8 to 50 amino acids, 8 to 25 amino acids, 8 to 20 amino acids, 8 to 15 amino acids or 8 to 12 amino acids. In one embodiment an immunogenic fragment of SEQ ID NO: 1-8, or a variant thereof, may be 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28,
- 15 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49 or 50 amino acids in length.

POLYNUCLEOTIDE

The polynucleotide may be any suitable type of polynucleotide, such as a synthetic RNA/DNA sequence, a cDNA sequence or a partial genomic DNA sequence.

- 20 As used herein, the terms nucleic acid sequence and polynucleotide are intended to be synonymous with each other. "Polynucleotide" generally refers to any polyribonucleotide or polydeoxiribonucleotide, which may be unmodified RNA or DNA or modified RNA or DNA. "Polynucleotides" include, without limitation single- and double-stranded DNA, DNA that is a mixture of single- and double-stranded regions, single- and double-stranded RNA, and RNA
- 25 that is mixture of single- and double-stranded regions, hybrid molecules comprising DNA and RNA that may be single-stranded or, more typically, double-stranded or a mixture of single- and double-stranded regions. In addition, "polynucleotide" refers to triple-stranded regions comprising RNA or DNA or both RNA and DNA. The term polynucleotide also includes DNAs or RNAs containing one or more modified bases and DNAs or RNAs with backbones modified for
- 30 stability or for other reasons. "Modified" bases include, for example, tritylated bases and unusual bases such as inosine. A variety of modifications has been made to DNA and RNA; thus, "polynucleotide" embraces chemically, enzymatically or metabolically modified forms of polynucleotides as typically found in nature, as well as the chemical forms of DNA and RNA

characteristic of viruses and cells. "Polynucleotide" also embraces relatively short polynucleotides, often referred to as oligonucleotides.

5 The term "recombinant polynucleotide" is used to mean that the polynucleotide is isolated from its natural environment, for example, extracted from or produced outside of an ASFV. In particular, the recombinant polynucleotide is not provided in the context of an ASFV and is provided as a polynucleotide, i.e. DNA or RNA that is created artificially.

10 It will be understood by a skilled person that numerous different polynucleotides can encode the same polypeptide as a result of the degeneracy of the genetic code. In addition, it is to be understood that skilled persons may, using routine techniques, make nucleotide substitutions that do not affect the polypeptide sequence encoded by the polynucleotides described here to reflect the codon usage of any particular host organism in which the polypeptides are to be expressed.

15 The term "variant" is used to mean a naturally occurring nucleic acid sequence which differs from a subject sequence. The variant may have at least 70% sequence identity with the subject sequence or up to and including 20 mutations.

The present vaccine may comprise one or more polynucleotides which encode a combination of recombinant polypeptides according to the vaccine of the present invention.

Illustrative polynucleotides encoding each of polypeptides SEQ ID NO: 1-8 are shown in Table 2.

20 The present vaccine may comprise one or more polynucleotides which encode recombinant polypeptides shown as SEQ ID NO: 1, 2 and 3 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 1, 2 and 3 or an immunogenic fragment thereof.

25 The present vaccine may comprise one or more polynucleotides which encode recombinant polypeptides shown as SEQ ID NO: 1, 2 and 3; or a variant with at least 70% sequence identity to one of SEQ ID NO: 1, 2 and/or 3. In one embodiment, the vaccine may consist of one or more polynucleotides which encode recombinant polypeptides shown as SEQ ID NO: 1, 2 and 3; or a variant with at least 70% sequence identity to one of SEQ ID NO: 1, 2 and/or 3.

30 The vaccine may further comprise one or more polynucleotides which encode one or more polypeptides selected from SEQ ID NO: 4-8 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 4-8 or an immunogenic fragment thereof.

The vaccine may further comprise one or more polynucleotides which encode one or more polypeptides selected from SEQ ID NO: 4-8 or a variant with at least 70% sequence identity to one of SEQ ID NO: 4-8.

5 The vaccine may comprise one or more polynucleotides which encode at least 2, at least 3, at least 4 or at least 5 polypeptides selected from SEQ ID NO: 4-8 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 4-8.

The vaccine may comprise one or more polynucleotides which encode 2, 3, 4 or 5 polypeptides selected from SEQ ID NO: 4-8 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 4-8 or an immunogenic fragment thereof.

10 Suitably, the vaccine may comprise one or more polynucleotides which encode recombinant polypeptides comprising SEQ ID NO: 1-8 or an immunogenic fragment thereof; or variants with at least 70% sequence identity to SEQ ID NO: 1-8 or immunogenic fragments thereof.

Suitably, the vaccine may comprise one or more polynucleotides which encode recombinant polypeptides comprising SEQ ID NO: 1-3 and 6-8 or an immunogenic fragment thereof; or
15 variants with at least 70% sequence identity to SEQ ID NO: 1-3 and 6-8 or immunogenic fragments thereof.

Suitably, the vaccine may comprise one or more polynucleotides which encode recombinant polypeptides comprising SEQ ID NO: 1-5 or an immunogenic fragment thereof; or variants with at least 70% sequence identity to SEQ ID NO: 1-5 or immunogenic fragments thereof.

20 A polynucleotide encoding the polypeptide shown as SEQ ID NO: 1-3 or a variant thereof may comprise SEQ ID NO: 9-11, respectively; or a variant thereof which shares at least 70% sequence identity to one of SEQ ID NO: 9-11.

A polynucleotide encoding the polypeptide shown as SEQ ID NO: 1 or a variant thereof may
25 comprise SEQ ID NO: 9; or a variant thereof which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 9.

A polynucleotide encoding the polypeptide shown as SEQ ID NO: 2 or a variant thereof may comprise SEQ ID NO: 10; or a variant thereof which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 10.

A polynucleotide encoding the polypeptide shown as SEQ ID NO: 3 or a variant thereof may
30 comprise SEQ ID NO: 11; or a variant thereof which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 11.

A polynucleotide encoding the polypeptide shown as SEQ ID NO: 4-8 or a variant thereof may comprise SEQ ID NO: 12-16, respectively; or a variant thereof which shares at least 70% sequence identity to one of SEQ ID NO: 12-16.

5 A polynucleotide encoding the polypeptide shown as SEQ ID NO: 4 or a variant thereof may comprise SEQ ID NO: 12; or a variant thereof which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 12.

A polynucleotide encoding the polypeptide shown as SEQ ID NO: 5 or a variant thereof may comprise SEQ ID NO: 13; or a variant thereof which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 13.

10 A polynucleotide encoding the polypeptide shown as SEQ ID NO: 6 or a variant thereof may comprise SEQ ID NO: 14; or a variant thereof which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 14.

15 A polynucleotide encoding the polypeptide shown as SEQ ID NO: 7 or a variant thereof may comprise SEQ ID NO: 15; or a variant thereof which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 15.

A polynucleotide encoding the polypeptide shown as SEQ ID NO: 8 or a variant thereof may comprise SEQ ID NO: 16; or a variant thereof which is at least 70, 80, 85, 90, 95, 98 or 99% identical to SEQ ID NO: 16.

20 The vaccine may further comprise one or more polypeptides selected from SEQ ID NO: 4-8 or a variant with at least 70% sequence identity to one of SEQ ID NO: 4-8.

Suitably, the vaccine may comprise one or more polynucleotides comprising SEQ ID NO: 9-16 or variants with at least 70% sequence identity to SEQ ID NO: 9-16.

Suitably, the vaccine may comprise one or more polynucleotides comprising SEQ ID NO: 9-11 and 14-16; or variants with at least 70% sequence identity to SEQ ID NO: 9-11 and 14-16.

25 Suitably, the vaccine may comprise one or more polynucleotides comprising SEQ ID NO: 9-13; or variants with at least 70% sequence identity to SEQ ID NO: 9-13 or immunogenic fragments thereof.

COMBINATION

The present invention is based, at least in part, on the inventors' determination of a minimal combination of polypeptides, or immunogenic fragments thereof, which is capable of inducing protection against ASFV.

5 It is to be understood that references to polypeptides or combinations of polypeptides provided herein are intended to apply equally to corresponding polynucleotides or combinations of polynucleotides which encode said polypeptides.

The total number of different ASFV polypeptides (or encoded by the one or more recombinant polynucleotides) in the present vaccine is 10 or fewer.

10 Suitably, the number of different ASFV polypeptides is 9 or fewer, 8 or fewer, 7 or fewer, or 6 or fewer.

Suitably, the number of different ASFV polypeptides is 3, 4, 5, 6, 7 or 8.

Suitably, the number of different ASFV polypeptides is 3.

Suitably, the number of different ASFV polypeptides is 5.

Suitably, the number of different ASFV polypeptides is 6.

15 Suitably, the number of different ASFV polypeptides is 8.

VECTOR

As used herein, a "vector" may be any agent capable of delivering or maintaining nucleic acid in a host cell, and includes viral, bacterial and eukaryotic vectors, plasmids, naked nucleic acids, nucleic acids complexed with polypeptide or other molecules and nucleic acids immobilised
20 onto solid phase particles.

Polynucleotides in accordance with the present invention may be delivered by viral or non-viral techniques.

Non-infectious delivery systems include but are not limited to DNA transfection methods. Here, transfection includes a process using a non-infectious vector to deliver nucleic acid of the
25 invention to a target cell.

Typical transfection methods include electroporation, nucleic acid biolistics, lipid-mediated transfection, compacted nucleic acid-mediated transfection, liposomes, immunoliposomes, lipofectin, cationic agent-mediated, cationic facial amphiphiles (CFAs), multivalent cations such

as spermine, cationic lipids or polylysine, 1, 2,-bis (oleoyloxy)-3-(trimethylammonio) propane (DOTAP)-cholesterol complexes and combinations thereof.

Non-viral delivery systems may also include, but are not limited to, bacterial delivery systems. Bacteria have previously been used as anticancer agents and as delivery agents for anticancer
5 drugs.

Cell adhesion molecules are a large group of molecules involved in a variety of cell-to-cell and cell-to-extra-cellular matrix (ECM) interactions and are exploited by a number of pathogenic micro-organisms as receptors for cell entry. These molecules may be used for the targeting and uptake of both gene and drug delivery systems.

10 A gene gun delivery system may also be used for the delivery of DNA.

Viral delivery systems include but are not limited to adenovirus vectors, modified vaccinia Ankara vectors, adeno-associated viral (AAV) vectors, herpes viral vectors such as pseudorabies virus vector, retroviral vectors, lentiviral vectors or baculoviral vectors, venezuelan equine encephalitis virus (VEE), poxviruses such as: canarypox virus, entomopox virus,
15 penguin alphavirus, and alphavirus based DNA vectors.

In one embodiment the vector may be an adenovirus vector.

The adenovirus is a double-stranded, linear DNA virus that does not replicate through an RNA intermediate. There are over 50 different human serotypes of adenovirus divided into 6 subgroups based on their genetic sequence.

20 Adenoviruses are double-stranded DNA non-enveloped viruses that are capable of in vivo, ex vivo and in vitro transduction of a broad range of cell types of human and non-human origin. These cells include respiratory airway epithelial cells, hepatocytes, muscle cells, cardiac myocytes, synoviocytes, primary mammary epithelial cells and post-mitotically terminally differentiated cells such as neurons. Adenoviral vectors are also capable of transducing non-
25 dividing cells.

Adenoviruses have been used as vectors for gene therapy and for expression of heterologous genes. The large (36 kb) genome can accommodate up to 8 kb of foreign insert DNA and is able to replicate efficiently in complementing cell lines to produce very high titres of up to 10¹² transducing units per ml. Adenovirus is thus one of the best systems to study the expression of
30 genes in primary non-replicative cells. The expression of viral or foreign genes from the adenovirus genome does not require a replicating cell. Adenoviral vectors enter cells by receptor mediated endocytosis. Once inside the cell, adenovirus vectors rarely integrate into

the host chromosome. Instead, they function episomally (independently from the host genome) as a linear genome in the host nucleus.

In one embodiment, the vector may be a modified vaccinia Ankara (MVA) vector. The MVA virus is related to Vaccinia virus, a member of the genera Orthopoxvirus in the family of Poxviridae. The MVA virus has been generated by 516 serial passages on chicken embryo fibroblasts of the Chorioallantois Vaccinia Ankara (CVA) virus. In the course of the attenuation process by repeated passaging to chicken derived material as production substrate, the MVA virus has lost approximately 15% of the genomic DNA at multiple sites (Mayr and Munz 1964 in Zentralbl Bakteriol Orig 195, 24-35; Meyer et al. 1991 in J Gen Virol 72 (Pt 5), 1031-1038). The MVA virus has been analysed to determine alterations in the genome relative to the wild-type CVA strain. Six major deletions of genomic DNA (deletion I, II, III, IV, V, and VI), totalling 31.000 base pairs, have been identified (Meyer, et al. 1991 in J Gen Virol 72 (Pt 5), 1031-1038). MVA does not replicate in human and non-human primate cells.

Alternatives to vaccinia vectors include avipox vectors such as fowlpox or canarypox known as ALVAC and strains derived therefrom which can infect and express recombinant proteins in human cells but are unable to replicate.

Examples of other vectors include *ex vivo* delivery systems, which include but are not limited to DNA transfection methods such as electroporation, DNA biolistics, lipid-mediated transfection and compacted DNA-mediated transfection.

The vector may be a plasmid DNA vector. As used herein, "plasmid" refers to discrete elements that are used to introduce heterologous DNA into cells for either expression or replication thereof.

In one embodiment, the vector may comprise more than one nucleic acid sequences, each of which encodes a different polypeptide as described for use in the present vaccine.

Suitably, one or more vectors may comprise multiple nucleic acid sequences which between them encode a combination of polypeptide as described for use in the present vaccine

In one embodiment, the vector may comprise more than one nucleic acid sequence, such that the vector comprises nucleic acid sequences which between them encode a combination of polypeptide as described for use in the present vaccine.

In order for the more than one nucleic acid sequences to be expressed, there may be two or more transcription units within the vector, one for each nucleic acid sequence. In one embodiment, an internal ribosome entry site (IRES) is used to initiate translation of the second

(and subsequent) coding sequence(s) in a polycistronic message (Adam et al 1991 J.Virol. 65, 4985).

5 Insertion of IRES elements into retroviral vectors is compatible with the retroviral replication cycle and allows expression of multiple coding regions from a single promoter (Adam et al (as above); Koo et al (1992) Virology 186:669-675; Chen et al 1993 J. Virol 67:2142-2148). IRES elements were first found in the non-translated 5' ends of picornaviruses where they promote cap-independent translation of viral proteins (Jang et al (1990) Enzyme 44: 292-309). When located between open reading frames in an RNA, IRES elements allow efficient translation of the downstream open reading frame by promoting entry of the ribosome at the IRES element
10 followed by downstream initiation of translation.

A review on IRES is presented by Mountford and Smith (TIG May 1995 vol 11, No 5:179-184). A number of different IRES sequences are known including those from encephalomyocarditis virus (EMCV) (Ghattas, I.R., et al., Mol. Cell. Biol., 11:5848-5859 (1991); BiP protein [Macejak and Sarnow, Nature 353:91 (1991)]; the Antennapedia gene of Drosophila (exons d and e) [Oh, et al., Genes & Development, 6:1643-1653 (1992)] as well as those in polio virus (PV) [Pelletier and Sonenberg, Nature 334: 320-325 (1988); see also Mountford and Smith, TIG 11, 179-184 (1985)].
15

The term "IRES" includes any sequence or combination of sequences which work as or improve the function of an IRES.

20 The IRES(s) may be of viral origin (such as EMCV IRES, PV IRES, or FMDV 2A-like sequences) or cellular origin (such as FGF2 IRES, NRF IRES, Notch 2 IRES or EIF4 IRES).

In order for the IRES to be capable of initiating translation of each polynucleotide it should be located between or prior to the polynucleotides in the vector genome.

25 The polynucleotide may encode a polypeptide which comprises two or more immunogenic polypeptides for use in the present vaccine, wherein each polypeptide is joined by a cleavage site. The cleavage site may be self-cleaving, such that when the polypeptide is produced, it is immediately cleaved into the individual immunogenic polypeptides without the need for any external cleavage activity.

30 Various self-cleaving sites are known, including the Foot-and-Mouth disease virus (FMDV) 2a self-cleaving peptide, which has the sequence shown:

SEQ ID NO:19

RAEGRGSLLCGDVEENPGP.

or

SEQ ID NO: 18

QCTNYALLKLAGDVESNPGP

5 A 'self-cleaving peptide' refers to a peptide which functions such that when the nascent polypeptide comprising two or more polypeptides for use in the present vaccine and the self-cleaving peptide is produced, it is immediately "cleaved" or separated into distinct and discrete ASFV polypeptides without the need for any external cleavage activity.

10 In embodiments wherein the polypeptide comprises multiple ASFV polypeptides for use in the present vaccine, for example, three or more ASFV polypeptides, there may be a self-cleaving polypeptide present between each of the ASFV polypeptides. Suitably, there may be a self-cleaving polypeptide present between the first and second ASFV polypeptides, the second and third ASFV polypeptides, etc.

15 The self-cleaving peptide may be a 2A self-cleaving peptide from an aphtho- or a cardiovirus. The primary 2A/2B cleavage of the aphtho- and cardioviruses is mediated by 2A "cleaving" at its own C-terminus. In aphthoviruses, such as foot-and-mouth disease viruses (FMDV) and equine rhinitis A virus, the 2A region is a short section of about 18 amino acids, which, together with the N-terminal residue of protein 2B (a conserved proline residue) represents an autonomous element capable of mediating "cleavage" at its own C-terminus.

20 Expression of a nucleic acid sequence may be controlled using control sequences, which include promoters/enhancers and other expression regulation signals. Prokaryotic promoters and promoters functional in eukaryotic cells may be used. Tissue specific or stimuli specific promoters may be used. Chimeric promoters may also be used comprising sequence elements from two or more different promoters.

25 Suitable promoting sequences are strong promoters including those derived from the genomes of viruses, such as polyoma virus, adenovirus, fowlpox virus, bovine papilloma virus, avian sarcoma virus, cytomegalovirus (CMV), retrovirus and Simian Virus 40 (SV40), or from heterologous mammalian promoters, such as the actin promoter, EF1a, CAG, TK, SV40, ubiquitin, PGK or ribosomal protein promoter. Alternatively, tissue-specific promoters such as rhodopsin (Rho), rhodopsin kinase (RhoK), cone-rod homeobox containing gene (CRX), neural retina-specific leucine zipper protein (NRL), Vitelliform Macular Dystrophy 2 (VMD2), Tyrosine hydroxylase, neuronal-specific neuronal-specific enolase (NSE) promoter, astrocyte-specific glial fibrillary acidic protein (GFAP) promoter, human α 1-antitrypsin (hAAT) promoter, phosphoenolpyruvate carboxykinase (PEPCK), liver fatty acid binding protein promoter, Flt-1 promoter, INF- β promoter, Mb promoter, SP-B promoter, SYN1 promoter, WASP promoter,

SV40 / hAlb promoter, SV40 / CD43, SV40 / CD45, NSE / RU5' promoter, ICAM-2 promoter, GPIIb promoter, GFAP promoter, Fibronectin promoter, Endoglin promoter, Elastase-1 promoter, Desmin promoter, CD68 promoter, CD14 promoter and B29 promoter may be used to drive transcription.

- 5 Transcription of a gene may be increased further by inserting an enhancer sequence into the vector. Enhancers are relatively orientation- and position-independent; however, one may employ an enhancer from a eukaryotic cell virus, such as the SV40 enhancer on the late side of the replication origin (bp 100-270) and the CMV early promoter enhancer. The enhancer may be spliced into the vector at a position 5' or 3' to the promoter, but is preferably located at a site
10 5' from the promoter.

The promoter can additionally include features to ensure or to increase expression in a suitable target cell. For example, the features can be conserved regions e.g. a Pribnow Box or a TATA box. The promoter may contain other sequences to affect (such as to maintain, enhance or decrease) the levels of expression of a nucleotide sequence. Suitable other sequences include
15 the Sh1-intron or an ADH intron. Other sequences include inducible elements, such as temperature, chemical, light or stress inducible elements. Also, suitable elements to enhance transcription or translation may be present.

VACCINE

The term 'vaccine' as used herein refers to a preparation which, when administered to a
20 subject, induces or stimulates a protective immune response. A vaccine can render an organism immune to a particular disease, for example in the present case ASF. The vaccine of the present invention thus induces an immune response in a subject that is protective against subsequent ASFV challenge.

The present vaccine may comprise a vector, recombinant polypeptide and/or recombinant
25 polynucleotide as described herein.

In one embodiment, the present vaccine comprises a plurality of vectors, recombinant polypeptides and/or recombinant polynucleotides as defined herein.

The term 'plurality' is used herein to mean more than one vector, recombinant polypeptide and/or recombinant polynucleotide as described herein. For example, a plurality may mean
30 two, three, four, five, six, seven, eight, nine, or ten recombinant polypeptides and/or recombinant polynucleotides as defined herein.

The vaccine may be useful in preventing African Swine Fever.

The vaccine composition may optionally comprise a pharmaceutically acceptable carrier, diluent, excipient or adjuvant. The choice of pharmaceutical carrier, excipient or diluent can be selected with regard to the intended route of administration and standard pharmaceutical practice. The vaccine composition may comprise as (or in addition to) the carrier, excipient or diluent, any suitable binder(s), lubricant(s), suspending agent(s), coating agent(s), solubilising agent(s), and other carrier agents that may aid or increase the delivery or immunogenicity of the vaccine.

Such a formulation may, for example, be in a form suitable for oral, intravenous, intramuscular, subcutaneous, intranasal, intradermal administration or suppository routes or implanting (e.g. using slow release molecules).

The vaccine may additionally comprise an adjuvant. Examples of adjuvants include but are not limited to aluminium salts, oil emulsions and bacterial components (e.g. LPS and liposomes).

METHODS OF PREVENTION/TREATMENT

The present invention also provides a method of preventing and/or treating ASF in a subject by administration of an effective amount of a vaccine of the invention.

The present invention further provides a vaccine of the first aspect of the present invention for use in treating and/or preventing ASF.

The term 'preventing' is intended to refer to averting, delaying, impeding or hindering the contraction of ASF. The vaccine may, for example, prevent or reduce the likelihood of an infectious ASFV entering a cell.

The term "treating" is intended to refer to reducing or alleviating at least one symptom of an existing ASF infection.

ADMINISTRATION

The vaccine may be administered in a convenient manner such as by the oral, intravenous, intramuscular, subcutaneous, intranasal, intradermal or suppository routes or implanting (e.g. using slow release molecules).

Typically, a veterinarian or producer will determine the actual dosage which will be most suitable for an individual subject or group of subjects and it may vary, for example, with the age, weight and response of the particular subject(s). The dosage is such that it is sufficient to reduce and/or prevent disease symptoms.

Those skilled in the art will appreciate, for example, that route of delivery (e.g., oral vs intravenous vs subcutaneous vs intratumoural, etc) may impact dose amount and/or required dose amount may impact route of delivery. For example, where particularly high concentrations of an agent within a particular site or location (e.g., within a tumour) are of interest, focused
5 delivery (e.g., in this example, intratumoural delivery) may be desired and/or useful. Other factors to be considered when optimizing routes and/or dosing schedule for a given therapeutic regimen may include, for example, the particular cancer being treated (e.g., type, stage, location, etc.), the clinical condition of a subject (e.g., age, overall health, etc.), the presence or absence of combination therapy, and other factors known to medical practitioners.

10 The dosage is such that it is sufficient to stabilise or improve symptoms of the disease.

The vaccine may be administered following a prime-boost regime. For example, after the first inoculation, the subject may receive a second boosting administration some time (e.g. 3 to 21, 3 to 14, 5 to 14 or 5 to 7 days) later. For example, the subject may receive a second boosting administration about 3, 5, 7, 14 or 21 days after the first inoculation.

15 In one embodiment, the first inoculation may be administered as one or more adenovirus vectors as described herein; and the second inoculation may be administered as one or more MVA vectors as defined herein. Typically the boosting administration is at a higher dose than the priming administration.

20 For all vertebrate use, the vector, immunogenic composition or vaccine may be administered parenterally, by injection, for example, either subcutaneously or intramuscularly. Additional formulations which are suitable for other modes of administration include suppositories and oral formulations. Where the immunogenic composition or vaccine is lyophilised, the lyophilised material may be reconstituted prior to administration, e.g. as a suspension.

25 SUBJECT

The subject may be any animal which is susceptible to ASF infection. ASF susceptible animals include domestic pigs, warhogs, bush pigs and ticks.

The subject vaccinated according to the present invention may be a domestic pig.

The subject may be susceptible to ASF infection.

30 Where the vaccine is used to treat an established infection, the subject may have been diagnosed as positive for the disease and/or show one or more symptom(s) associated with the infection.

This disclosure is not limited by the exemplary methods and materials disclosed herein, and any methods and materials similar or equivalent to those described herein can be used in the practice or testing of embodiments of this disclosure. Numeric ranges are inclusive of the numbers defining the range. Unless otherwise indicated, any nucleic acid sequences are written left to right in 5' to 3' orientation; amino acid sequences are written left to right in amino to carboxy orientation, respectively.

Where a range of values is provided, it is understood that each intervening value, to the tenth of the unit of the lower limit unless the context clearly dictates otherwise, between the upper and lower limits of that range is also specifically disclosed. Each smaller range between any stated value or intervening value in a stated range and any other stated or intervening value in that stated range is encompassed within this disclosure. The upper and lower limits of these smaller ranges may independently be included or excluded in the range, and each range where either, neither or both limits are included in the smaller ranges is also encompassed within this disclosure, subject to any specifically excluded limit in the stated range. Where the stated range includes one or both of the limits, ranges excluding either or both of those included limits are also included in this disclosure.

It must be noted that as used herein and in the appended claims, the singular forms "a", "an", and "the" include plural referents unless the context clearly dictates otherwise.

The terms "comprising", "comprises" and "comprised of" as used herein are synonymous with "including", "includes" or "containing", "contains", and are inclusive or open-ended and do not exclude additional, non-recited members, elements or method steps. The terms "comprising", "comprises" and "comprised of" also include the term "consisting of".

The publications discussed herein are provided solely for their disclosure prior to the filing date of the present application. Nothing herein is to be construed as an admission that such publications constitute prior art to the claims appended hereto.

The invention will now be further described by way of Examples, which are meant to serve to assist one of ordinary skill in the art in carrying out the invention and are not intended in any way to limit the scope of the invention.

EXAMPLES

Example 1 - Immunisation of pigs with pools of subunit ASFV polypeptides and challenge with ASFV

Various polynucleotides encoding different ASFV polypeptides were individually cloned in recombinant adenovirus and modified vaccinia Ankara (MVA) vectors. The cloned sequences were from the genotype I OURT88/3 strain except for EP153R and MGF360-11L which were from the genotype I strain Benin 1997/1.

5 Ten groups of pigs were immunised as shown in Table 1.

Pigs in experiments AR000737 and AR000742 were primed with 5E9 IU of adenovirus and 7.5E7 pfu of MVA. Pigs in experiments AR000750 and AR000858 were primed with 1.5×10^{10} infectious units of each recombinant adenovirus vector and boosted with 2×10^8 plaque forming units of each recombinant MVA vector.

10 Details of the different combinations of antigens, vectors used for the priming and boosting doses and induction of protection are provided in Table 1.

B602L, E183L and EP153R are present in all of the groups in which protection was observed after challenge. Protection was determined when animals did not reach the predetermined humane endpoints of the study.

15 However, both B602L and EP153R were also in Pool B of Experiment 2 in which no protection was observed. E183L was in Pool 5 of Experiment 4 in which protection was observed after challenge. Therefore, the combination of B602L and EP153R alone was not sufficient for protection alone, likewise E183L alone is not sufficient for protection.

20

*Materials & Methods***Materials and Methods****African swine fever viruses and cells**

5 The OUR T88/1 isolate was grown in primary macrophages from pig bone marrow. Virus stocks were prepared from a spleen suspension from an infected pig and titres were determined by limiting dilution using haemadsorption to detect virus infected cells and titres were calculated by the Spearman-Karber method

Pig immunisation and challenge

10 Pigs were either outbred cross-bred Large White and Landrace or outbred cross-bred White, Landrace and Hampshire from a high health status farm or. Pigs were of average size 20 kg at the start of experiments.

15 Recombinant adenoviruses (rAds) and modified vaccinia Ankara viruses were delivered by intramuscular injection in at most 5 ml total volume at two separate sites. MVA or rAd boosts were delivered to pigs 4 weeks after the initial rAd prime. Pigs were challenged intramuscularly with 10^4 HAD₅₀ Georgia 2007/1 virus four to eight weeks after the boost. Pigs were observed for development of clinical signs and these were scored using the scoring system used previously. Blood and tissue samples were collected to measure levels of virus replication.

20 All publications mentioned in the above specification are herein incorporated by reference. Various modifications and variations of the described methods and system of the invention will be apparent to those skilled in the art without departing from the scope and spirit of the invention. Although the invention has been described in connection with specific preferred embodiments, it should be understood that the invention as claimed should not be unduly limited to such specific embodiments. Indeed, various modifications of the described modes for carrying out the invention which are obvious to those skilled in the art are intended to be within
25 the scope of the following claims.

CLAIMS

1. An African swine fever virus (ASFV) subunit vaccine comprising:
- (i) one or more recombinant polynucleotides which encode polypeptides shown as SEQ ID NO: 1, 2 and 3 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 1, 2 or 3 or an immunogenic fragment thereof; wherein the total number of different ASFV polypeptides encoded by the one or more recombinant polynucleotides is 10 or fewer; or
- (ii) recombinant polypeptides shown as SEQ ID NO: 1, 2 and 3 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 1, 2 and 3 or an immunogenic fragment thereof; wherein vaccine comprises 10 or fewer different ASFV polypeptides.
2. A vaccine according to claim 1, wherein the number of different ASFV polypeptides encoded by the one or more recombinant polynucleotides is 9 or fewer, 8 or fewer, 7 or fewer, or 6 or fewer; or wherein the vaccine comprises 9 or fewer, 8 or fewer, 7 or fewer, or 6 or fewer ASFV polypeptides.
3. A vaccine according to claim 1 or 2 which comprises:
- (i) one or more further recombinant polynucleotides encoding an ASFV polypeptide selected from SEQ ID NO: 4-8 or an immunogenic fragment thereof, or a variant with at least 70% sequence identity to one of SEQ ID NO: 4-8 or an immunogenic fragment thereof; or
- (ii) one or more further polypeptides selected from SEQ ID NO: 4-8 or an immunogenic fragment thereof; or a variant with at least 70% sequence identity to one of SEQ ID NO: 4-8 or an immunogenic fragment thereof.
4. A vaccine according to any preceding claim which comprises:
- a) a recombinant polynucleotide(s) which encodes polypeptides comprising SEQ ID NO: 1-8 or an immunogenic fragment thereof or a variant with at least 70% sequence identity to SEQ ID NO: 1-8 or immunogenic fragments thereof; or recombinant polypeptides comprising SEQ ID NO: 1-8 or an immunogenic fragment thereof or variants with at least 70% sequence identity to SEQ ID NO: 1-8 or immunogenic fragments thereof;
- (b) a recombinant polynucleotide(s) which encodes polypeptides comprising SEQ ID NO: 1-3 and 6-8 or an immunogenic fragment thereof or variants with at least 70%

- sequence identity to SEQ ID NO: 1-3 and 6-8 or immunogenic fragments thereof; or recombinant polypeptides comprising SEQ ID NO: 1-3 and 6-8 or an immunogenic fragment thereof or variants with at least 70% sequence identity to SEQ ID NO: 1-3 and 6-8 or immunogenic fragments thereof; or
- 5 (c) a recombinant polynucleotide(s) which encode polypeptides comprising SEQ ID NO: 1-5 or an immunogenic fragment thereof or variants with at least 70% sequence identity to SEQ ID NO: 1-5 or immunogenic fragments thereof; or recombinant polypeptides comprising SEQ ID NO: 1-5 or an immunogenic fragment thereof or variants with at least 70% sequence identity to SEQ ID NO: 1-5 or immunogenic fragments thereof.
- 10 5. A vaccine according to claim 1 wherein the recombinant polynucleotides comprise SEQ ID NO: 9, 10 and 11 or a variant thereof with at least 70% sequence identity.
6. A vaccine according to claim 3 wherein the further polynucleotides comprise one or more of SEQ ID NO: 12-16 or a variant thereof with at least 70% sequence identity.
7. A vaccine according to any preceding claim wherein the polynucleotide is present in a
15 vector.
8. A vaccine according to claim 7 wherein the vector is selected from an adenovirus, a modified vaccinia Ankara vector and a pseudorabies virus vector.
9. A vaccine according to claim 7 or 8 wherein each of the recombinant polynucleotides is provided in the same vector.
- 20 10. A vaccine according to any preceding claim for use in treating and/or preventing African swine fever in a subject.
11. A method for treating and/or preventing African swine fever in a subject which comprises administering a therapeutically effective amount of the vaccine according to any of claims 1 to 9 to the subject.
- 25 12. Use of a recombinant polynucleotide, vector or recombinant polypeptide as defined in any of claims 1 to 9 in the manufacture of a medicament for the treatment and/or prevention of African swine fever in a subject.
13. A vaccine for use according to claim 10, a method according to claim 11 or use of a recombinant polynucleotide, vector or recombinant polypeptide according to claim 12 wherein
30 the subject is a swine subject.

14. A vaccine for use, a method or a use according to claim 13 wherein the subject is a domestic pig.
15. A vaccine for use, a method or a use according to any of claims 10 to 14 wherein the vaccine is administered according to a prime-boost procedure.
- 5 16. A vaccine for use, a method or a use according to claim 15 wherein the priming composition comprises one or more adenovirus vectors and the boosting composition comprises one or more modified vaccinia Ankara vectors.
- 10 17. A vaccine for use, a method or a use according to any of claims 10 to 16 wherein the vaccine is administered by oral, intravenous, intramuscular, subcutaneous, intranasal or intradermal administration.

FIGURE 1

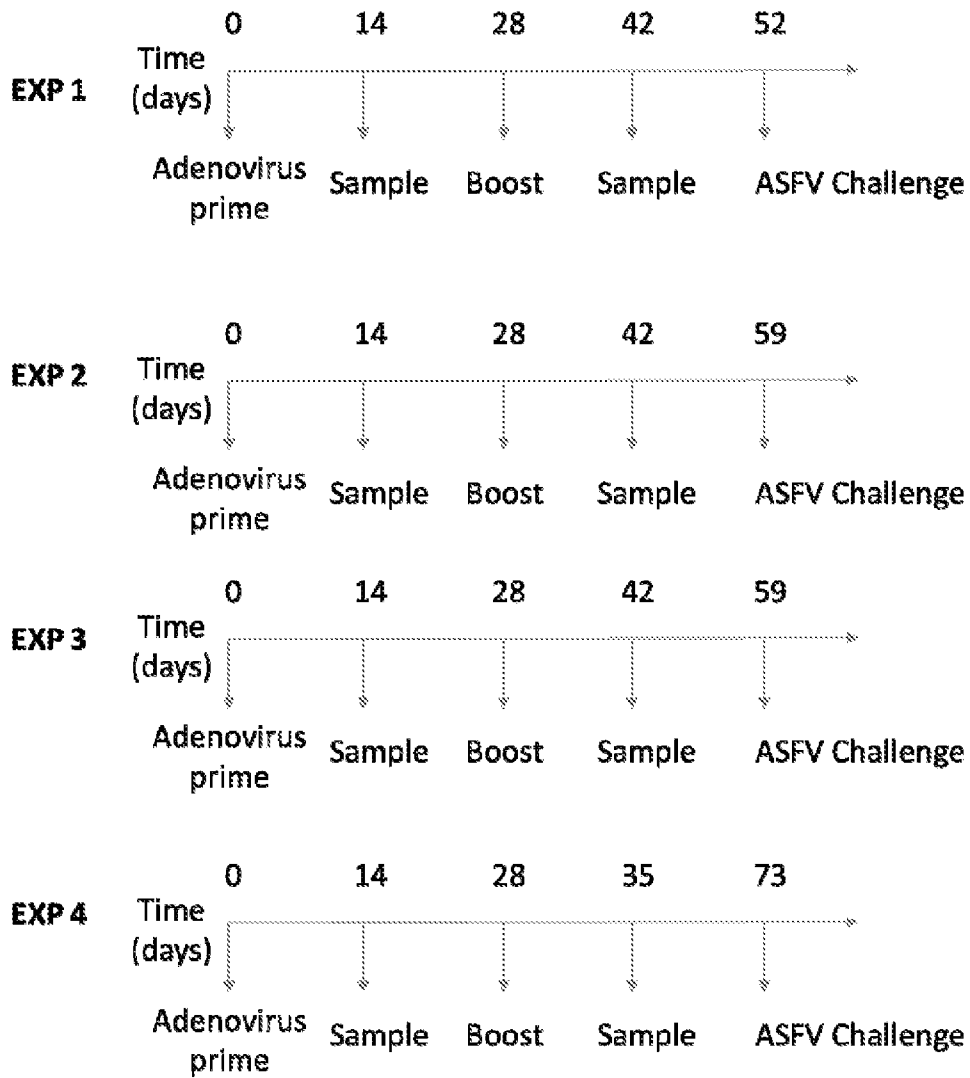


FIGURE 2

SEQ ID NO: 1 - B602L (OUR T88/3)

MAEFNIDELLKNVLEDPSTEISEETLKQLYQRTNPYKQFKNDSRVAFCSTNLREQYIRRLIMTS
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 KSKEAKTTIDSFLREHFVFDPNLHAQSAYTCASTCADTNVDTCASTCASTCASTCASTCA
 STCASTCASTCASTCASTCASTCASTGASTGASTCADTNVDTCASTCADTNVDTCASTC
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 TCASMCADTNVDTCASTCANTCASTEYTDLADPERIPLHIMQKTLNVPNELQADIDAITQTPQ
 GYRAAAHILQNIELHQSIKHMLENPRAFKPILFNTKITRYLSQHIPPQDTFYKWNYYIEDNYEEL
 RAATESIYPEKPDLEFAFIIDVVDSSNQKQVDEFYKYKQDQIFSEVSSIQLGNWTLGGSFKAN
 RERYNYFNQNNIIRILDRHEEDLKIGKEILRNTIYHKKAKNIQETGPDAPGLSIYNSTFHTDS
 GIKGLLSFKELKNLEKASGNIKKAREYDFIDDCEEKIKQLLKENLTPDEESELIKTKKQLNNAL
 EMLNVPDDTIRVDMWVNNNNKLEKEILYTKAEL

SEQ ID NO: 2 - E183L (p54) (OUR T88/3)

MDSEFFQPVYPRHYGECLSPVTPPSFFSTHMYTILIAIVVLVIIIVLIIYLFSSRKKKAAAIEEEDI
 QFINPYQDQQWAEVTPQPGTSKPAGATTASAGKPVTRPATNRPATNKPVTDNPVTDRLVMA
 TGGPAAAPAAASAHPTPEYTTVTQNTASQTMSAIENLRQRNTYTHKDLENSL

SEQ ID NO: 3 EP153R (Benin 1997/1)

MYFKKKYIGLIDKNCEKKILDDSSSTIKICYILIGILIGTNMITLIYNFIFWDNYIKCYRNNDKMFY
 CPNDWVGYNNICYYFSNGSFSKNYTAASNFCRQLNGTLANNDTNLLNLTKIYNNQSMYWVN
 NTVILRGDNKYSQKVNYTDLLFICGK

SEQ ID NO: 4 - B646L (OUR T88/3)

MASGGAFCLIANDGKADKILAQDLLNSRISNIKNVNSYGKPDPEPTLSQIEETHLVHFNAHF
 KPYVPVGFYENKVRPHTGTPTLGNKLTFGIPQYGDFHDMVGHHLGACHSSWQDAPIQGTA
 QMGAHGQLQTFPRNGYDWDNQTPLEGAVYTLVDPFGRPIVPGTKNAYRNLVYYCEYPGERL
 YENVRFDVNGNSLDEYSSDVTTLVRKFCIPGDKMTGYKHLVGQEVSVEGTSGPLLCNIHDLH
 KPHQSKPILTDENDTQRTCSHTNPKFLSQHFPENSHNIQTAGKQDITPITDATYLDIRRVHYS
 CNGPQTPKYYPPLALWIKLRFWFNENVNLAIPSVSIPFGERFITIKLASQKDLVNEFPGLFIRQSR
 FIPGRPSRRNIRFKPWFIPGVINEISLTNNELYINNLFVTPEIHNLVVKRVRFSLIRVHKTVHTN
 NNHHDEKLMSALKWPIEYMFGLKPTWNISDQNPQHHRDWHKFGHVVNAIMQPTHAEISFQ
 DRDTALPDACSSISDISPVTYPITLPIKNISVTAHGINLIDKFPSKFCSSYIPFHYGGNAIKTPDDP
 GAMMITFALKPREEYQPSGHINVSRAREFYISWDTDYVGSITTADLVVSASAINFLLQNGSAV
 LRYST

SEQ ID NO: 5 - CP204L (p30) (OUR T88/3)

MDFILNISMKMEVIFKTDLRSSSQVVFHAGSLYNWFSVEIINSGRIVTTAIKTLLSTVKYDIVKS
 AHYAGQGYTEHQAQEEWNMILHVLFEETESSASSESIHEKNDNETNECASSFETLFEQEPSSE
 EPKDSKLYMLAQKTVQHIEQYGKAPDFNKVIRAHNFIQTIHGTPLKEEKEVVRMLVIKLLKK
 K

SEQ ID NO: 6 - E199L (OUR T88/3)

MSCMPVSTKCNDIWVDFSC TGPSISELQKKEPKAWAAILRSHTNQQTAEEDNIIIGSICDKRGLC
 SKDEYAYSQYCACVNSGTLWAECAPCNGNKNAYKTTEQRNLTNKQCPSGLTICQNIAYEY
 RGSNISDLYQNFNCNSVINTFLINVMNHPFLTILILILIIIIYRLMPSSGGKHNDKLPPLSIF
 NLNNF

FIGURE 2 – CONTD.

SEQ ID NO: 7 - F317L (OUR T88/3)

MVETQMDKLGFLLNHIGKQVTTKVLNAHITQTMKEIILENHGVDGGAAKNVSKGKSSPKEK
KHWTEFESWEQLSKSKRSFKEYWAERNEIVNTLLL NWDNVRGAIKKFLDDDREWCGRINMIN
GVPEIVEIIPSPYRAGENIYFGSEAMMPADIYSRVANKPAMFVFHHPNLGSCCGMPSICDIST
TLRYLLMGWTAGHLIISNQVGMMLTVDKRIIVDLWANENPRWLMAQKILDIFMMLTSRRSLV
NPWTLRDLKILQDYGIEYIIFPSNDFFIYEDERLLMFSKKWTNFFTLHELLDDLETIETKASSTT

SEQ ID NO: 8 - MGF505-5R (OUR T88/3)

MFSLQEICRKNIYFLPDWLSEHVIQRLGLYWEKHGSLQRIGDDYVLIQQDLIIPINEALRMAGEE
GNDEVVQLLLLWEGNIHYAIIGALES DHYSLIRKLYDQIGDCHDILPLIQDPKIFEKCHELDKFC
NILCLVLHAVKNDMLCILQEYKMHLSGEDIQVVFETACRSQKNDIVSWMGQNAIYNSGVIFDI
AFDKMNVSLLSIGYTLLFNHHINNTNENINSLLTQHLEWAAGMGLLHFMLETLKYGGDVTIIV
LSEAVKYDHRKILDYFLRRKNLYQEDLEELLLLAI RADCSKKTLNLLLSYLNYSINNIRKKILQ
CVKEYETTVIKILWKRKINLIEPILADFIGYHSYTYMVDFMREFSIHPEKMIKMAARESREDLII
KFSKKVCKECPKDRHLHYLKSLVYTMRHKEGKQLLIYTIHNLKACHLESKEMFNLARFYARHN
AVIQFKSICHDL SKLNINIKNLLLECLGIAIKKNYFQLIKTIETDMRYE

SEQ ID NO: 17 EP153R (Genotype II)

MFSNKKYIGLINKKEGLKKKIDDY SILIIGILIGTNILSLIINIIGEINKPICYQND DKIFYCPKDWV
GYNNVCY YFGNEEK NYNNASNYCKQLNSTLTNNNTILVNLTKTLNLTKTYNHESNYWVNYS
LIKNESVLLRDSGYYKKQKHVSLLYICSK

FIGURE 3 – CONTD.

tgatacgtgtccataaaacgcaggtgacccacaccaacaataaccaccacgatgaaaaactaatgtctgctcttaaatggcccattgaatatatgtttat
aggattaaaaacctacgtggaacatctccgatcaaaaatcctcatcaaacaccgagattggcacaagttcggacatgttgtaacgccattatgcagcctact
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cggctcagctgtgctgcgttacgtacaccta

SEQ ID NO: 13 - CP204L (p30) (OUR T88/3)

Atggatttttttaaatatcatgaaaatggaggtcatctcaaacggattfaagatcatcttcacaagttgtgtttcatcgcggtgagctgtataattg
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cattcatggaacccctctaagggaagaagaaaaagaggtgtaagactcatgttcattaaacttttaaaaaaaaaataa

SEQ ID NO: 14 - E199L (OUR T88/3)

Atgtctgcatccagttccacgaatgcaatgatattgggtcagctttagctgtacaggcccttcgattccgagctgcaaaaaaggagcccaag
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SEQ ID NO: 15 - F317L (OUR T88/3)

Atggttgagacacaatggacaacttggtttctgtaaatcacataggttaaacggttaccactaagggtgcttagcaatgccatataactcaaacg
atgaaggagattatttggaaaatcatggtgtatgaggtggagccgcaaaaaattttcaaaagggaagcttccccaaaagaaaaaacattggac
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SEQ ID NO: 16 - MGF505-5R (OUR T88/3)

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gattgctatataaaaaaattactttcaacttatcaaaaatagaacggatgctgattgagtaa

FIGURE 4

Table 1

Experiment #	AR000737 1		AR000742 2		AR000750 3		AR000858 4				
Experiment Pool	Pool A	Pool B	Pool A	Pool B	Pool A	Pool B	Pool 1	Pool 2	Pool 3	Pool 4	Pool 5
	A151R	B602L	B602L	B602L	B602L		B602L	B602L	B602L	B602L	
	B646L	B646L	B646L	I329L	B646L	B646L	B646L	B646L	B646L	B602L	B646L
	CP204L	CP204L	CP204L	MGF505-4R	CP204L	CP204L	CP204L	CP204L	CP204L	CP204L	CP204L
	CP530R, C129R	E183L	E183L	MGF360-11L	E183L		E183L	E183L	E183L	E183L	E183L
	M448R, L8L	E199L	E199L	EP364R	E199L	E199L	E199L	E199L			E199L
	I73R, I215L	EP153R	EP153R	EP153R	EP153R		EP153R	EP153R	EP153R	EP153R	
	E146L, MGF110-5L	F317L	F317L	F317L	F317L	F317L	F317L	F317L			F317L
	MGF110-4L	MGF505-5R	MGF505-5R	MGF505-5R	MGF505-5R	MGF505-5R	MGF505-5R	MGF505-5R	MGF505-5R		MGF505-5R
Vectors used (Prime-Boost)	Ad-MVA	Ad-MVA	Ad-MVA	Ad-MVA	Ad-MVA	Ad-MVA	Ad-MVA	Ad-Ad	Ad-Ad	Ad-MVA	Ad-Ad
Protection	0/6	2/6	0/6	0/6	6/6	0/6	2/5	4/5	3/5	3/5	0/5

115

120

125

Thr Gln Lys Ser Lys Glu Ala Lys Thr Thr Ile Asp Ser Phe Leu Arg
 130 135 140

Glu His Phe Val Phe Asp Pro Asn Leu His Ala Gln Ser Ala Tyr Thr
 145 150 155 160

Cys Ala Ser Thr Cys Ala Asp Thr Asn Val Asp Thr Cys Ala Ser Thr
 165 170 175

Cys Ala Ser Thr Cys Ala Ser Thr Cys Ala Ser Thr Cys Ala Ser Thr
 180 185 190

Cys Ala Ser Thr Cys Ala Ser Thr Cys Ala Ser Thr Cys Ala Ser Thr
 195 200 205

Cys Ala Ser Thr Cys Ala Ser Thr Cys Ala Ser Thr Cys Ala Ser Thr
 210 215 220

Gly Ala Ser Thr Gly Ala Ser Thr Cys Ala Asp Thr Asn Val Asp Thr
 225 230 235 240

Cys Ala Ser Thr Cys Ala Asp Thr Asn Val Asp Thr Cys Ala Ser Thr
 245 250 255

Cys Ala Ser Thr Cys Ala Ser Thr Cys Ala Ser Thr Cys Ala Ser Thr
 260 265 270

Gly Ala Ser Thr Cys Ala Asp Thr Asn Val Asp Thr Cys Ala Ser Thr
 275 280 285

Cys Ala Asp Thr Asn Val Asp Thr Cys Ala Ser Thr Cys Ala Asp Thr
 290 295 300

Asn Val Asp Thr Cys Ala Ser Thr Cys Ala Asp Thr Asn Val Asn Thr
 305 310 315 320

Cys Ala Ser Met Cys Ala Asp Thr Asn Val Asp Thr Cys Ala Ser Thr
 325 330 335

Cys Ala Asn Thr Cys Ala Ser Thr Glu Tyr Thr Asp Leu Ala Asp Pro
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Glu Arg Ile Pro Leu His Ile Met Gln Lys Thr Leu Asn Val Pro Asn
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Glu Leu Gln Ala Asp Ile Asp Ala Ile Thr Gln Thr Pro Gln Gly Tyr
370 375 380

Arg Ala Ala Ala His Ile Leu Gln Asn Ile Glu Leu His Gln Ser Ile
385 390 395 400

Lys His Met Leu Glu Asn Pro Arg Ala Phe Lys Pro Ile Leu Phe Asn
405 410 415

Thr Lys Ile Thr Arg Tyr Leu Ser Gln His Ile Pro Pro Gln Asp Thr
420 425 430

Phe Tyr Lys Trp Asn Tyr Tyr Ile Glu Asp Asn Tyr Glu Glu Leu Arg
435 440 445

Ala Ala Thr Glu Ser Ile Tyr Pro Glu Lys Pro Asp Leu Glu Phe Ala
450 455 460

Phe Ile Ile Tyr Asp Val Val Asp Ser Ser Asn Gln Gln Lys Val Asp
465 470 475 480

Glu Phe Tyr Tyr Lys Tyr Lys Asp Gln Ile Phe Ser Glu Val Ser Ser
485 490 495

Ile Gln Leu Gly Asn Trp Thr Leu Leu Gly Ser Phe Lys Ala Asn Arg
500 505 510

Glu Arg Tyr Asn Tyr Phe Asn Gln Asn Asn Glu Ile Ile Lys Arg Ile
515 520 525

Leu Asp Arg His Glu Glu Asp Leu Lys Ile Gly Lys Glu Ile Leu Arg
530 535 540

Asn Thr Ile Tyr His Lys Lys Ala Lys Asn Ile Gln Glu Thr Gly Pro

35

40

45

Ile Tyr Leu Phe Ser Ser Arg Lys Lys Lys Ala Ala Ala Ala Ile Glu
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Glu Glu Asp Ile Gln Phe Ile Asn Pro Tyr Gln Asp Gln Gln Trp Ala
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Glu Val Thr Pro Gln Pro Gly Thr Ser Lys Pro Ala Gly Ala Thr Thr
85 90 95

Ala Ser Ala Gly Lys Pro Val Thr Gly Arg Pro Ala Thr Asn Arg Pro
100 105 110

Ala Thr Asn Lys Pro Val Thr Asp Asn Pro Val Thr Asp Arg Leu Val
115 120 125

Met Ala Thr Gly Gly Pro Ala Ala Ala Pro Ala Ala Ala Ser Ala His
130 135 140

Pro Thr Glu Pro Tyr Thr Thr Val Thr Thr Gln Asn Thr Ala Ser Gln
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Thr Met Ser Ala Ile Glu Asn Leu Arg Gln Arg Asn Thr Tyr Thr His
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Lys Asp Leu Glu Asn Ser Leu
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<213> Artificial Sequence

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25

30

Ile Gly Ile Leu Ile Gly Thr Asn Met Ile Thr Leu Ile Tyr Asn Phe
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Ile Phe Trp Asp Asn Tyr Ile Lys Cys Tyr Arg Asn Asn Asp Lys Met
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Phe Tyr Cys Pro Asn Asp Trp Val Gly Tyr Asn Asn Ile Cys Tyr Tyr
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Phe Ser Asn Gly Ser Phe Ser Lys Asn Tyr Thr Ala Ala Ser Asn Phe
85 90 95

Cys Arg Gln Leu Asn Gly Thr Leu Ala Asn Asn Asp Thr Asn Leu Leu
100 105 110

Asn Leu Thr Lys Ile Tyr Asn Asn Gln Ser Met Tyr Trp Val Asn Asn
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Ile Lys Asn Val Asn Lys Ser Tyr Gly Lys Pro Asp Pro Glu Pro Thr

35

40

45

Leu Ser Gln Ile Glu Glu Thr His Leu Val His Phe Asn Ala His Phe
50 55 60

Lys Pro Tyr Val Pro Val Gly Phe Glu Tyr Asn Lys Val Arg Pro His
65 70 75 80

Thr Gly Thr Pro Thr Leu Gly Asn Lys Leu Thr Phe Gly Ile Pro Gln
85 90 95

Tyr Gly Asp Phe Phe His Asp Met Val Gly His His Ile Leu Gly Ala
100 105 110

Cys His Ser Ser Trp Gln Asp Ala Pro Ile Gln Gly Thr Ala Gln Met
115 120 125

Gly Ala His Gly Gln Leu Gln Thr Phe Pro Arg Asn Gly Tyr Asp Trp
130 135 140

Asp Asn Gln Thr Pro Leu Glu Gly Ala Val Tyr Thr Leu Val Asp Pro
145 150 155 160

Phe Gly Arg Pro Ile Val Pro Gly Thr Lys Asn Ala Tyr Arg Asn Leu
165 170 175

Val Tyr Tyr Cys Glu Tyr Pro Gly Glu Arg Leu Tyr Glu Asn Val Arg
180 185 190

Phe Asp Val Asn Gly Asn Ser Leu Asp Glu Tyr Ser Ser Asp Val Thr
195 200 205

Thr Leu Val Arg Lys Phe Cys Ile Pro Gly Asp Lys Met Thr Gly Tyr
210 215 220

Lys His Leu Val Gly Gln Glu Val Ser Val Glu Gly Thr Ser Gly Pro
225 230 235 240

Leu Leu Cys Asn Ile His Asp Leu His Lys Pro His Gln Ser Lys Pro
245 250 255

Ile Leu Thr Asp Glu Asn Asp Thr Gln Arg Thr Cys Ser His Thr Asn
260 265 270

Pro Lys Phe Leu Ser Gln His Phe Pro Glu Asn Ser His Asn Ile Gln
275 280 285

Thr Ala Gly Lys Gln Asp Ile Thr Pro Ile Thr Asp Ala Thr Tyr Leu
290 295 300

Asp Ile Arg Arg Asn Val His Tyr Ser Cys Asn Gly Pro Gln Thr Pro
305 310 315 320

Lys Tyr Tyr Gln Pro Pro Leu Ala Leu Trp Ile Lys Leu Arg Phe Trp
325 330 335

Phe Asn Glu Asn Val Asn Leu Ala Ile Pro Ser Val Ser Ile Pro Phe
340 345 350

Gly Glu Arg Phe Ile Thr Ile Lys Leu Ala Ser Gln Lys Asp Leu Val
355 360 365

Asn Glu Phe Pro Gly Leu Phe Ile Arg Gln Ser Arg Phe Ile Pro Gly
370 375 380

Arg Pro Ser Arg Arg Asn Ile Arg Phe Lys Pro Trp Phe Ile Pro Gly
385 390 395 400

Val Ile Asn Glu Ile Ser Leu Thr Asn Asn Glu Leu Tyr Ile Asn Asn
405 410 415

Leu Phe Val Thr Pro Glu Ile His Asn Leu Phe Val Lys Arg Val Arg
420 425 430

Phe Ser Leu Ile Arg Val His Lys Thr Gln Val Thr His Thr Asn Asn
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Asn His His Asp Glu Lys Leu Met Ser Ala Leu Lys Trp Pro Ile Glu
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Tyr Met Phe Ile Gly Leu Lys Pro Thr Trp Asn Ile Ser Asp Gln Asn

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35 40 45

Thr Ala Ile Lys Thr Leu Leu Ser Thr Val Lys Tyr Asp Ile Val Lys
50 55 60

Ser Ala His Ile Tyr Ala Gly Gln Gly Tyr Thr Glu His Gln Ala Gln
65 70 75 80

Glu Glu Trp Asn Met Ile Leu His Val Leu Phe Glu Glu Glu Thr Glu
85 90 95

Ser Ser Ala Ser Ser Glu Ser Ile His Glu Lys Asn Asp Asn Glu Thr
100 105 110

Asn Glu Cys Ala Ser Ser Phe Glu Thr Leu Phe Glu Gln Glu Pro Ser
115 120 125

Ser Glu Glu Pro Lys Asp Ser Lys Leu Tyr Met Leu Ala Gln Lys Thr
130 135 140

Val Gln His Ile Glu Gln Tyr Gly Lys Ala Pro Asp Phe Asn Lys Val
145 150 155 160

Ile Arg Ala His Asn Phe Ile Gln Thr Ile His Gly Thr Pro Leu Lys
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<220>
<223> ASFV polypeptide, E199L (OUR T88/3)

<400> 6

Met Ser Cys Met Pro Val Ser Thr Lys Cys Asn Asp Ile Trp Val Asp
1 5 10 15

Phe Ser Cys Thr Gly Pro Ser Ile Ser Glu Leu Gln Lys Lys Glu Pro
20 25 30

Lys Ala Trp Ala Ala Ile Leu Arg Ser His Thr Asn Gln Gln Thr Ala
35 40 45

Glu Asp Asp Asn Ile Ile Gly Ser Ile Cys Asp Lys Arg Gly Leu Cys
50 55 60

Ser Lys Asp Glu Tyr Ala Tyr Ser Gln Tyr Cys Ala Cys Val Asn Ser
65 70 75 80

Gly Thr Leu Trp Ala Glu Cys Ala Phe Ala Pro Cys Asn Gly Asn Lys
85 90 95

Asn Ala Tyr Lys Thr Thr Glu Gln Arg Asn Ile Leu Thr Asn Lys Gln
100 105 110

Cys Pro Ser Gly Leu Thr Ile Cys Gln Asn Ile Ala Glu Tyr Arg Gly
115 120 125

Ser Gly Asn Ile Ser Asp Leu Tyr Gln Asn Phe Asn Cys Asn Ser Val
130 135 140

Ile Asn Thr Phe Leu Ile Asn Val Met Asn His Pro Phe Leu Thr Leu
145 150 155 160

Ile Leu Ile Ile Leu Ile Leu Ile Ile Ile Tyr Arg Leu Met Pro Ser
165 170 175

Ser Gly Gly Lys His Asn Asp Asp Lys Leu Pro Pro Pro Ser Leu Ile
180 185 190

Phe Ser Asn Leu Asn Asn Phe
195

<210> 7
<211> 317
<212> PRT
<213> Artificial Sequence

<220>
<223> ASFV polypeptide, F317L (OUR T88/3)

<400> 7

Met Val Glu Thr Gln Met Asp Lys Leu Gly Phe Leu Leu Asn His Ile
1 5 10 15

Gly Lys Gln Val Thr Thr Lys Val Leu Ser Asn Ala His Ile Thr Gln
20 25 30

Thr Met Lys Glu Ile Ile Leu Glu Asn His Gly Val Asp Gly Gly Ala
35 40 45

Ala Lys Asn Val Ser Lys Gly Lys Ser Ser Pro Lys Glu Lys Lys His
50 55 60

Trp Thr Glu Phe Glu Ser Trp Glu Gln Leu Ser Lys Ser Lys Arg Ser
65 70 75 80

Phe Lys Glu Tyr Trp Ala Glu Arg Asn Glu Ile Val Asn Thr Leu Leu
85 90 95

Leu Asn Trp Asp Asn Val Arg Gly Ala Ile Lys Lys Phe Leu Asp Asp
100 105 110

Asp Arg Glu Trp Cys Gly Arg Ile Asn Met Ile Asn Gly Val Pro Glu
115 120 125

Ile Val Glu Ile Ile Pro Ser Pro Tyr Arg Ala Gly Glu Asn Ile Tyr
130 135 140

Phe Gly Ser Glu Ala Met Met Pro Ala Asp Ile Tyr Ser Arg Val Ala
145 150 155 160

Asn Lys Pro Ala Met Phe Val Phe His Thr His Pro Asn Leu Gly Ser
165 170 175

Cys Cys Gly Gly Met Pro Ser Ile Cys Asp Ile Ser Thr Thr Leu Arg
180 185 190

Tyr Leu Leu Met Gly Trp Thr Ala Gly His Leu Ile Ile Ser Ser Asn
195 200 205

Gln Val Gly Met Leu Thr Val Asp Lys Arg Ile Ile Val Asp Leu Trp
210 215 220

Ala Asn Glu Asn Pro Arg Trp Leu Met Ala Gln Lys Ile Leu Asp Ile
225 230 235 240

Phe Met Met Leu Thr Ser Arg Arg Ser Leu Val Asn Pro Trp Thr Leu
245 250 255

Arg Asp Leu Lys Lys Ile Leu Gln Asp Tyr Gly Ile Glu Tyr Ile Ile
260 265 270

Phe Pro Ser Asn Asp Phe Phe Ile Tyr Glu Asp Glu Arg Leu Leu Met
275 280 285

Phe Ser Lys Lys Trp Thr Asn Phe Phe Thr Leu His Glu Leu Leu Asp
290 295 300

Asp Leu Glu Thr Ile Glu Thr Lys Ala Ser Ser Thr Thr
305 310 315

<210> 8

<211> 498

<212> PRT

<213> Artificial Sequence

<220>

<223> ASFV polypeptide, MGF505-5R (OUR T88/3)

<400> 8

Met Phe Ser Leu Gln Glu Ile Cys Arg Lys Asn Ile Tyr Phe Leu Pro
1 5 10 15

Asp Trp Leu Ser Glu His Val Ile Gln Arg Leu Gly Leu Tyr Trp Glu
20 25 30

Lys His Gly Ser Leu Gln Arg Ile Gly Asp Asp Tyr Val Leu Ile Gln
35 40 45

Gln Asp Leu Ile Ile Pro Ile Asn Glu Ala Leu Arg Met Ala Gly Glu
50 55 60

Glu Gly Asn Asp Glu Val Val Gln Leu Leu Leu Leu Trp Glu Gly Asn
65 70 75 80

Ile His Tyr Ala Ile Ile Gly Ala Leu Glu Ser Asp His Tyr Ser Leu
85 90 95

Ile Arg Lys Leu Tyr Asp Gln Ile Gly Asp Cys His Asp Ile Leu Pro
100 105 110

Leu Ile Gln Asp Pro Lys Ile Phe Glu Lys Cys His Glu Leu Asp Lys
115 120 125

Phe Cys Asn Ile Leu Cys Leu Val Leu His Ala Val Lys Asn Asp Met
130 135 140

Leu Cys Ile Leu Gln Glu Tyr Lys Met His Leu Ser Gly Glu Asp Ile
145 150 155 160

Gln Val Val Phe Glu Thr Ala Cys Arg Ser Gln Lys Asn Asp Ile Val
165 170 175

Ser Trp Met Gly Gln Asn Ile Ala Ile Tyr Asn Ser Gly Val Ile Phe
180 185 190

Asp Ile Ala Phe Asp Lys Met Asn Val Ser Leu Leu Ser Ile Gly Tyr
195 200 205

Thr Leu Leu Phe Asn His His Ile Asn Asn Thr Asn Glu Asn Ile Asn

210

215

220

Ser Leu Leu Thr Gln His Leu Glu Trp Ala Ala Gly Met Gly Leu Leu
225 230 235 240

His Phe Met Leu Glu Thr Leu Lys Tyr Gly Gly Asp Val Thr Ile Ile
245 250 255

Val Leu Ser Glu Ala Val Lys Tyr Asp His Arg Lys Ile Leu Asp Tyr
260 265 270

Phe Leu Arg Arg Lys Asn Leu Tyr Gln Glu Asp Leu Glu Glu Leu Leu
275 280 285

Leu Leu Ala Ile Arg Ala Asp Cys Ser Lys Lys Thr Leu Asn Leu Leu
290 295 300

Leu Ser Tyr Leu Asn Tyr Ser Ile Asn Asn Ile Arg Lys Lys Ile Leu
305 310 315 320

Gln Cys Val Lys Glu Tyr Glu Thr Thr Val Ile Ile Lys Ile Leu Trp
325 330 335

Lys Arg Lys Ile Asn Leu Ile Glu Pro Ile Leu Ala Asp Phe Ile Gly
340 345 350

Tyr His Ser Tyr Thr Tyr Met Val Asp Phe Met Arg Glu Phe Ser Ile
355 360 365

His Pro Glu Lys Met Ile Lys Met Ala Ala Arg Glu Ser Arg Glu Asp
370 375 380

Leu Ile Ile Lys Phe Ser Lys Lys Val Cys Lys Glu Pro Lys Asp Arg
385 390 395 400

Leu His Tyr Leu Lys Ser Leu Val Tyr Thr Met Arg His Lys Glu Gly
405 410 415

Lys Gln Leu Leu Ile Tyr Thr Ile His Asn Leu Tyr Lys Ala Cys His
420 425 430

Leu Glu Ser Lys Glu Met Phe Asn Leu Ala Arg Phe Tyr Ala Arg His
435 440 445

Asn Ala Val Ile Gln Phe Lys Ser Ile Cys His Asp Leu Ser Lys Leu
450 455 460

Asn Ile Asn Ile Lys Asn Leu Leu Leu Glu Cys Leu Gly Ile Ala Ile
465 470 475 480

Lys Lys Asn Tyr Phe Gln Leu Ile Lys Thr Ile Glu Thr Asp Met Arg
485 490 495

Tyr Glu

- <210> 9
- <211> 2025
- <212> DNA
- <213> Artificial Sequence

<220>
<223> nucleic acid sequence encoding ASFV polypeptide B602L (OUR T88/3)

<400> 9
atggcagaat ttaatattga tgagcttctc aaaaacgtat tggaggatcc ctctactgaa 60
atatccgaag aaacgcttaa acagctttat caaaggacga acccttacia acagttcaaa 120
aatgatagca ggggtggcctt ttgctctttt acaaatttgc gggagcagta tattcgacgt 180
cttataatga ctagctttat tggatatgtc ttcaaagctc tgcaggaatg gatgccttcc 240
tattcaaaac ctaccacac gacccaaact cttctcagtg agctaataac gttagttgat 300
actttgaaac aggaaactaa tgatgttccc tctgaatcgg tagtaaatac aattttatct 360
atagcggata gctgcaaaac ccagacgcag aaaagcaagg aagctaaaac aacgatcgat 420
agctttttac gagaacattt tgtgtttgat cctaactctc atgctcaaag tgcgtatact 480
tgtgcaagca cttgtgcaga taccaatgta gacacctgtg caagcacttg tgcaagcact 540
tgtgcaagca cttgtgcaag cacttgtgca agcacttgtg caagcacttg tgcaagcact 600
tgtgcaagca cttgtgcaag cacttgtgca agcacttgtg caagcacttg tgcaagcact 660
tgtgcaagca caggtgcaag cacaggtgca agcacttgtg cagataccaa ttagacacc 720

tgtgcaagca cttgtgcaga taccaatgta gacacctgtg caagcacttg tgcaagcact	780
tgtgcaagca cttgtgcaag cacttgtgca agcacagggtg caagcacttg tgcaagcact	840
aatgtagaca cctgtgcaag cacttgtgca gataccaatg tagacacctg tgcaagcact	900
tgtgcaagca ccaatgtaga cacctgtgca agcacttgtg cagataccaa tgtaaacact	960
tgtgcaagca tgtgtgcaga taccaatgta gacacctgtg caagcacttg tgcaaacacc	1020
tgtgcaagca cagaatacac cgatttagca gatcctgagc gcatcccttt acacatcatg	1080
caaaaaacat taaatgtgcc taatgagctt caggccgata ttgatgcaat tacccaaacc	1140
ccacagggtc atagggcagc agcccacata ttacaaaata tagaacttca tcaaagcatt	1200
aaacatatgc ttgaaaatcc gagggcgctt aaaccattc tctttaacac aaaaattact	1260
agatatcttt cgcagcatat tccacctcag gatacttttt ataagtggaa ttattacatt	1320
gaggataatt acgaagagtt gcgggcccgt acggaaagca tctaccaga aaagcccagc	1380
ctagagtttg ctttcattat ttatgatgtg gtggatagca gcaaccaaca aaaggttgat	1440
gaattttatt ataaatataa agaccagatt ttctcagagg tttcatccat tcaattaggc	1500
aactggacac tcctgggaag ctttaaggcc aacagagagc gctacaatta tttaataca	1560
aataatgaaa taataaacg gattttggac cgtcatgagg aagacctaaa gataggaaaa	1620
gagattctac gaaataccat ttaccacaaa aaagcaaaaa atatacaaga aaccggcccc	1680
gatgctccgg ggctctccat ctataattca acctttcaca cggatagcgg gattaaggga	1740
ctgctttcct ttaaggagct aaaaaaccta gaaaaagcat ctggaaatat caaaaaagct	1800
cgagagtatg attttataga cgactgcgaa gaaaaaatta agcaactgct tagtaaagaa	1860
aatttaacc cccgatgaaga aagcgagctg ataaaaaca aaaaacagtt aaataatgag	1920
cttgaaatgc tcaatgtgcc tgatgatacg atacgggtag atatgtgggt caacaataat	1980
aataaactcg aaaaagaat tttatataca aaagcagaat tgtaa	2025

<210> 10

<211> 552

<212> DNA

<213> Artificial Sequence

<220>

<223> nucleic acid sequence encoding E183L (p54) (OUR T88/3)

<400> 10

atggattctg aatTTTTTca accgTTTTat ccgcggcatt atggtgagtg tttgtcacca 60
 gtcacccac caagcttctt ctccacacat atgtatacta ttctcattgc tatcgtggtc 120
 ttagtcatta ttatcatcgt tctaattctat ctatttcttt caagaaagaa aaaagctgct 180
 gccgctattg aggaggaaga tatacagttt ataaatcctt atcaagatca gcaatgggca 240
 gaagtcactc cacaaccagg tacctctaaa ccggctggag cgactacagc aagtcagggc 300
 aaaccagtca cgggcagacc ggcaacaaac agaccagcaa caaacaaacc agtcacggac 360
 aaccagtta cggacagact agtcatggca actggcgggc cagcggccgc acctgcggcc 420
 gcgagtgtc atccgactga gccttacagc acagtcacta ctcagaacac tgcttcacaa 480
 acaatgtcgg ctattgaaaa tttacgacaa agaaacacct atacgataa agacctagaa 540
 aactccttgt aa 552

<210> 11
 <211> 462
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> nucleic acid sequence encoding EP153R (Benin 1997/1)

<400> 11
 atgtatttta agaaaaata catcggcttt attgataaga actgtgaaaa aaaaatatta 60
 gatgattcta gtacaataaa aatttgttac atattaattg gaatattgat tggaactaat 120
 atgataactc ttatttataa tttcatattc tgggataatt atataaaatg ttaccgaaat 180
 aatgataaaa tgttttactg tcctaattgat tgggttggat ataataatat ttgttactat 240
 tttagtaatg gtagtttttc taaaaattat acagctgcta gtaatttttg tagacaatta 300
 aatggtacac ttgctaataa tgataactaat ttattaaatc taactaaaat atataataat 360
 caatctatgt attgggttaa caatacggta atattacgtg gtgataataa atatagtcaa 420
 aaagttaact atacagattt attattttatt tgttgtaaat aa 462

<210> 12
 <211> 1941
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> nucleic acid sequence encoding B646L (OUR T88/3)

<400> 12

atggcatcag gaggagcttt ttgtcttatt gctaacgatg ggaaggccga caagattata	60
ttggccaag acttgcttaa tagcaggatt tctaacatta aaaatgtgaa caaaagttat	120
gggaaacccg accccgaacc cactttgagt caaatcgaag aaacacattt ggttcatttt	180
aatgcgcat ttaagcctta tgttccagta gggtttgaat acaataaagt acgcccgcac	240
acgggtacc ccaccttggg aaacaagctt acctttggta ttccccagta cggagacttt	300
ttccatgata tgggtgggcca ccatatattg ggtgcatgtc attcgtcctg gcaggatgct	360
ccgattcagg gcacggccca gatgggggccc catggtcagc ttcaaacggt tcctcgcaac	420
ggatatgact gggacaacca aacaccttta gagggcgccg tttacacgct ttagatccc	480
tttgggaagac ctattgtacc cggcacaaaag aatgcgctacc gaaacttggg ttactactgc	540
gaatacccg gagaacgact ttatgaaaac gtaagattcg atgtaaattg aaattccctg	600
gacgaatata gttcggatgt cacaacgctt gtgcgcaaat tttgcatccc aggggataaa	660
atgactggat ataagcactt ggtcggccag gaggtatcgg tggagggaac tagtggcct	720
ctcctatgca acattcatga tttgcacaag ccgcaccaa gcaaacctat tcttaccgat	780
gaaaatgata cgcagcgaac gtgcagccat accaaccgga aattcctttc acaacatttt	840
cccgagaact ctacaatat ccaaacagca ggtaaacaag atattactcc tattacggac	900
gcaacgtatc tggacataag acgtaatggt cattacagct gtaatggacc tcaaaccct	960
aaatactatc agccccctct tgcgctctgg attaagctgc gcttttggtt taacgagaac	1020
gtgaaccttg ctattccctc ggtatccatt cccttcggcg agcgctttat caccataaag	1080
cttgcacgc aaaaggattt ggtgaatgaa tttcctggac tttttatacg ccagtgcgct	1140
tttatacctg gacgccccag tagacgcaat atacgcttta aacctgggtt tatcccagga	1200
gtcattaatg aaatctcgct cacgaataat gaactttaca tcaataacct gtttgaacc	1260
cctgaaatac acaacctttt tgtaaaacgc gttcgatttt ccctgatacg tgtccataaa	1320
acgcaggtga cccacaccaa caataaccac cacgatgaaa aactaatgtc tgctcttaaa	1380
tggcccattg aatatatggt tataggatta aaacctacct ggaacatctc cgatcaaaat	1440
cctcatcaac accgagattg gcacaagttc ggacatggtt ttaacgcat tatgcagcct	1500
actcaccag cagagataag ctttcaggat agagatacag ctcttcaga cgcatgttca	1560

tctatatcgg atattagccc cgttacgtat ccgatcacat tacctattat taaaaacatt	1620
tccgtaactg ctcatgggat caatccttatc gataagtttc catcaaagtt ctgcagctct	1680
tacataccct tccactacgg aggcaatgca attaaaaccc ccgatgatcc gggtgcatg	1740
atgattacct ttgctttgaa gccacgggag gaataccaac ccagtgggtca tattaacgta	1800
tccagagcaa gagaatttta tattagttgg gacacggatt acgtggggtc tatcactacg	1860
gctgatcttg tggatcggc atctgctatt aactttcttc ttcttcagaa cggttcagct	1920
gtgctgcggt acagtaccta a	1941

<210> 13
 <211> 585
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> nucleic acid sequence encoding CP204L (p30) (OUR T88/3)

<400> 13	
atggatttta ttttaaatat atccatgaaa atggagggtca tcttcaaaac ggatttaaga	60
tcattctcac aagttgtggt tcatgcbggg agcttgtata attggttttc tgttgagatt	120
atcaatagcg gtagaattgt tacgaccgct ataaaaacat tgctcagtac tgtaagtat	180
gatattgtga aatctgctca tatatatgca gggcaagggt atactgaaca tcaggctcaa	240
gaagaatgga atatgattct gcatgtgctg tttgaagagg agacagaatc ctcagcatca	300
tcggaaagca ttcataaaaa aatgataat gaaaccaatg aatgcbgcatc ctcttttgaa	360
acattgtttg agcaagagcc ctcatcagag gaacctaaag actccaagct gtatatgctt	420
gcacaaaaga ctgtgcaaca tattgaacaa tatggaaagg cacctgattt taacaagggt	480
attagagcac ataactttat tcaaaccatt catggaaccc ctctaaagga agaagaaaaa	540
gaggtggtaa gactcatggt cattaaactt ttaaaaaaaaa aataa	585

<210> 14
 <211> 600
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> nucleic acid sequence encoding E199L (OUR T88/3)

<400> 14

atgtcttgca tgccagtttc cacgaaatgc aatgatattt gggctgactt tagctgtaca 60
ggcccttcga tttccgagct gcaaaaaaag gagcccaagg cctgggccgc tattttacgc 120
tcgcatacaa atcaacaaac ggcggaggat gacaatatta ttgggagcat atgcgataaa 180
cggggattgt gctcaaagga tgagtatgcg tatagccagt attgtgcctg tgtgaactcc 240
ggcaccctat gggctgaatg tgcgtttgct ccgtgtaatg gaaataaaaa tgcctataaa 300
acaacggagc aaagaaatat tttgaccaac aagcagtgcc cctccggact caccatatgt 360
cagaacattg cagaatacag aggctcgggc aatatttccg acctatacca aaatttcaac 420
tgcaacagcg ttataaatac gtttttaatt aatgtgatga atcatccttt tttaacctt 480
atattaatca ttttgattct tataattatt taccgtttga tgccttccag tgggggtaaa 540
cacaatgacg ataagttgcc ccctccatct cttatttttt caaacctaaa caatttttaa 600

<210> 15
<211> 954
<212> DNA
<213> Artificial Sequence

<220>
<223> nucleic acid sequence encoding F317L (OUR T88/3)

<400> 15
atggttgaga cacaatgga caaacttggt tttctgctaa atcacatagg taaacaggtt 60
accactaagg tgcttagcaa tgcccatata actcaaacga tgaaggagat tattttggaa 120
aatcatggtg tagatggtgg agccgcaaaa aatgtttcaa aagggaagtc ttccccaaaa 180
gaaaaaaaaac attggaccga gttcgaatcc tgggaacagc tcagcaagtc taaaagaagt 240
ttcaaggaat actgggcgga gcgtaatgag attgtgaaca ctctgttgct taattgggac 300
aatgttcggg gtgcatcaa aaaatTTTTG gacgatgacc gtgaatggtg cggccgcatt 360
aatatgataa acggtgtacc cgagatagtg gaaatcattc caagccccta tagggcagga 420
gagaacattt attttggcag cgaggctatg atgcctgctg atatttatag cagggtggcc 480
aacaagcctg ctatgtttgt gtttcacacg catcctaatt tgggttcatg ttgtggagga 540
atgccttcca tatgtgacat ttctacaacg ctgcttatc tacttatggg gtggaccgcc 600
gggcatctaa tcatttcttc gaatcaagta ggaatgctca cggttgataa gagaattatt 660
gttgatttgt gggccaatga gaatccgcgc tggctcatgg cgcaaaaaat attagatatt 720

tttatgatgc tcacttcgcg tagaagcctg gtaaaccctt ggaccctgag agacctaata 780
aaaatattac aagactatgg tattgagtat atcatttttc cttcgaatga cttttttatt 840
tatgaagacg aacgtctttt aatgttttca aaaaaatgga ccaacttttt tacgtttacat 900
gagttattgg atgacctcga aactattgag acaaaggcat cgtccacaac atag 954

<210> 16
<211> 1497
<212> DNA
<213> Artificial Sequence

<220>
<223> nucleic acid sequence encoding MGF505-5R (OUR T88/3)

<400> 16
atgttctccc tccaggagat ctgtcgaaag aacatctact ttctacctga ctggctcagt 60
gagcatgtga ttcagcgact aggtctgtac tgggaaaaac atggttctct tcagcgaatc 120
ggagacgact atgtacttat acaacaggat ctcatcatcc ccatcaatga agctctaaga 180
atggcagggg aggaggggaa tgatgaggtg gtacaactcc tattactatg ggaggggaaac 240
attcattatg ccatcatagg agctttggag agtgaccatt atagcctaata acgtaagctc 300
tatgaccaa tcggagactg tcacgacatc cttcccttaa ttcaagacc aaaaatcttt 360
gaaaaatgcc atgaattaga taaattttgt aacattttat gtctcgtatt acacgccgta 420
aaaaacgata tgctttgcat tcttcaagag tataaaatgc atctaagtgg agaggatatt 480
caagtgggtg ttgaaacagc atgccgttca caaaaaaacg atattgtgtc atggatggga 540
caaatattg caatatacaa ctccggagtt atttttgata ttgcctttga taagatgaat 600
gtgtccttat tatctatagg gtacacgctt cttttcaatc atcatataaa taatacgaac 660
gaaaatatta attctttatt gacacaacat cttgaatggg ctgccggcat gggccttctt 720
cattttatgc tggaaacttt aaagtatggc ggggatgtaa cgataatagt tttgtctgag 780
gccgtaaaat atgaccacag aaagatttta gattattttc tccgtcga aaacttgatc 840
caagaagatc ttgaagaact attattgttg gcgatacgtg cagattgttc taaaagacc 900
ttaaacttgt tattatctta cttaactat tccataaaca atatccgtaa aaaaatatta 960
caatgtgtaa aagaatatga aacgaccgtt attataaaaa ttctatggaa aagaagata 1020
aatctgatag agcccatttt ggcagacttt ataggatatc atagctatac ctatatggta 1080

gatTTtatgc gcgagTtttc catccatccg gaaaaaatga tcaaaatggc tgcgcgagaa 1140
 tcgagggagg acttaatcat aaaatTttcc aaaaaagttt gcaaagagcc taaagataga 1200
 cttcactatc tcaaaagctt agtgtatact atgcgacata aagaaggcaa acaactgtta 1260
 attatacaa tccataactt atacaaagct tgtcatctag agagtaaaga aatgtttaat 1320
 ttggcacgat tttatgcacg gcataatgca gtgatccagt tcaaatcgat ttgtcacgat 1380
 ctctccaagc tgaatattaa tatcaaaaac ttgttgtagg aatgtttagg tattgctatt 1440
 aaaaaaatt actttcaact tatcaaaaca atagaaacgg atatgCGtta tgagtaa 1497

<210> 17
 <211> 158
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> ASFV polypeptide, 17 EP153R (Genotype II)

<400> 17

Met Phe Ser Asn Lys Lys Tyr Ile Gly Leu Ile Asn Lys Lys Glu Gly
 1 5 10 15

Leu Lys Lys Lys Ile Asp Asp Tyr Ser Ile Leu Ile Ile Gly Ile Leu
 20 25 30

Ile Gly Thr Asn Ile Leu Ser Leu Ile Ile Asn Ile Ile Gly Glu Ile
 35 40 45

Asn Lys Pro Ile Cys Tyr Gln Asn Asp Asp Lys Ile Phe Tyr Cys Pro
 50 55 60

Lys Asp Trp Val Gly Tyr Asn Asn Val Cys Tyr Tyr Phe Gly Asn Glu
 65 70 75 80

Glu Lys Asn Tyr Asn Asn Ala Ser Asn Tyr Cys Lys Gln Leu Asn Ser
 85 90 95

Thr Leu Thr Asn Asn Asn Thr Ile Leu Val Asn Leu Thr Lys Thr Leu
 100 105 110

Asn Leu Thr Lys Thr Tyr Asn His Glu Ser Asn Tyr Trp Val Asn Tyr

115

120

125

Ser Leu Ile Lys Asn Glu Ser Val Leu Leu Arg Asp Ser Gly Tyr Tyr
130 135 140

Lys Lys Gln Lys His Val Ser Leu Leu Tyr Ile Cys Ser Lys
145 150 155

<210> 18
<211> 20
<212> PRT
<213> Foot-and-mouth disease virus

<400> 18

Gln Cys Thr Asn Tyr Ala Leu Leu Lys Leu Ala Gly Asp Val Glu Ser
1 5 10 15

Asn Pro Gly Pro
20

<210> 19
<211> 20
<212> PRT
<213> Foot-and-mouth disease virus

<400> 19

Arg Ala Glu Gly Arg Gly Ser Leu Leu Thr Cys Gly Asp Val Glu Glu
1 5 10 15

Asn Pro Gly Pro
20