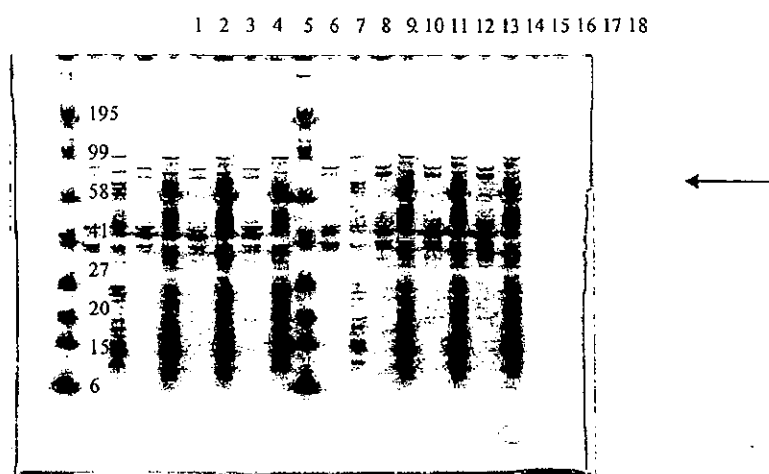
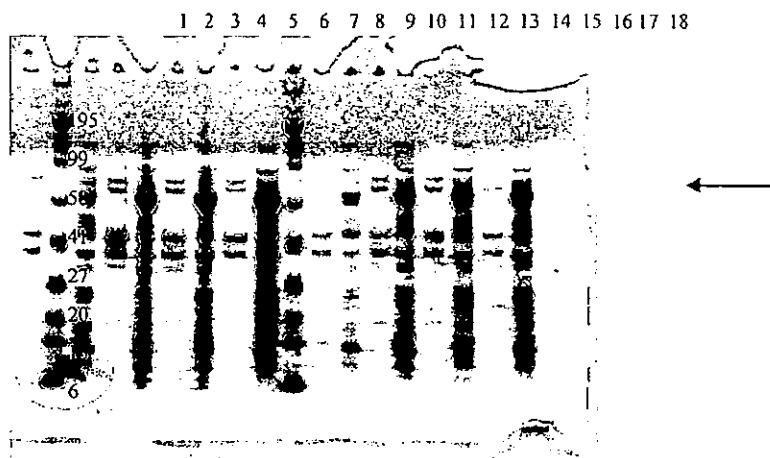


Fig 1 –SDS page analysis of 1/3PD-PRAME with or without His-tail in pET21 vector
(Example/construct Nos 3 and 4)



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Fig 2 -SDS page analysis of 1/3PD-PRAME codon optimized with or without His-tail in pET26 vector (Example/constructNos 3a and 4a)



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Figure 3

CD4 response (AS01B adjuvant)

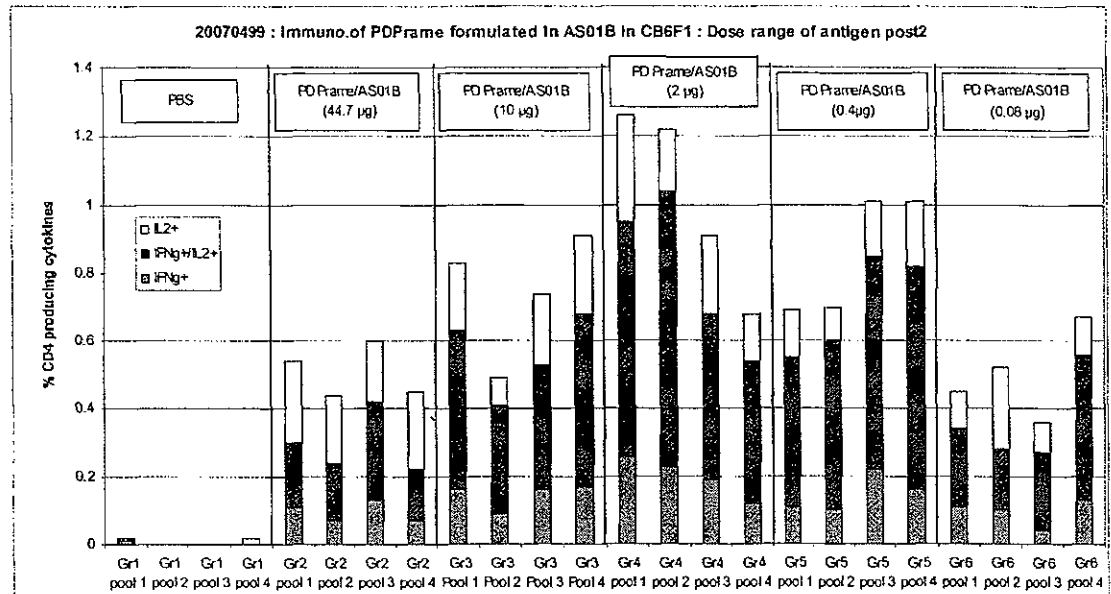


Figure 4

CD8 response (AS01B adjuvant - 20070499)

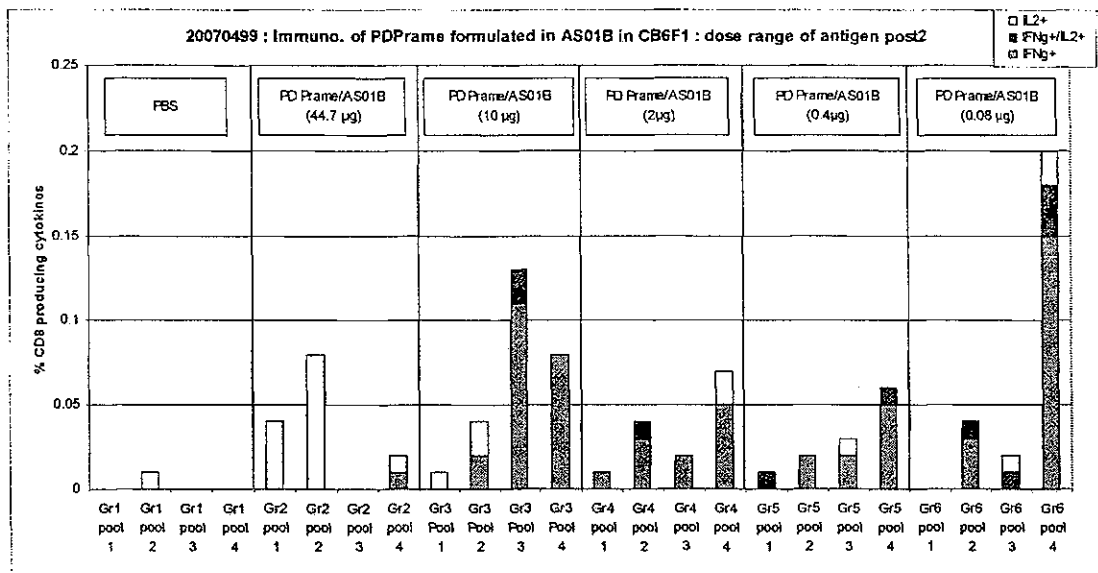
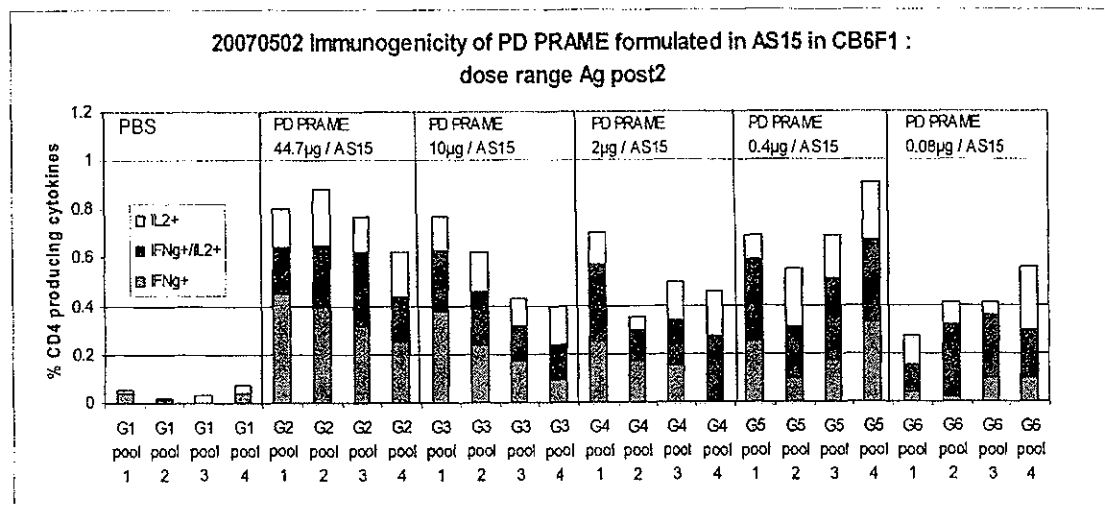


Figure 5

CD4 response (AS15 adjuvant - 20070502)



5

Figure 6

CD8 response (AS15 adjuvant - 20070502)

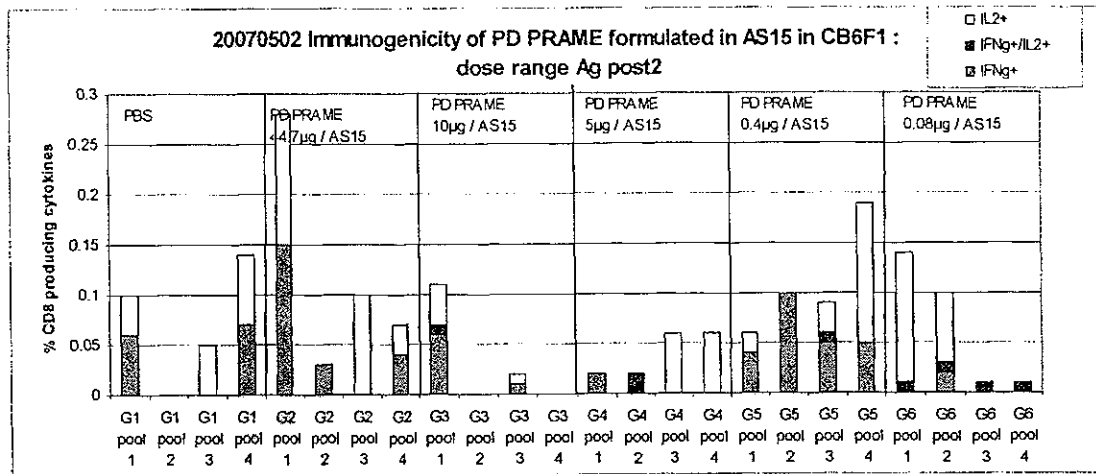


Figure 7

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1      MDPSSSSNM ANTQMKSDKI IIAHRGASGY LPEHTLESKA LAFAQQADYL
51     EQDLAMTKDG RLVVIHDHFL DGLTDVAKKF PHRHRKDGRY YVIDFTLKEI
5      101     QSLEMTENFE TMERRRLWGS IQSRYISMSV WTSPRRLVEL AGQSLLKDEA
151    LAIAALELLP RELFPPLFMA AFDGRHSOTL KAMVQAWPFT CLPLGVLMKG
201    QHLHLETFKA VLDGLDVLLA QEVPRRWKL QVLDLRKNSH QDFWTWWSGN
251    RASLYSFPEP EAAQPMKKR KVDGLSTEAE QPFIPVEVLV DLFLKEGACD
301    ELFSYLIEKV KRKKNVLRCL CKKLKIFAMP MQDIKMILKM VQLDSIEDLE
10     351    VTCTWKLPTL AKFSPYLGOM INLRRLLLSH IHASSYISPE KEEQYIAQFT
401    SQFLSLOCLO ALYVDSLFFL RGRLDQLLRH VMNPLETISI TNCRLSEGDV
451    MHLQSQSPVS QLSVLSLSGV MLTDVSPEPL QALLERASAT LQDLVFDECG
501    ITDDOLLALL PSLSHCSOLT TLSFYGNSIS ISALQSLLQH LIGLSNLTHV
551    LYPVPLESYE DIHGTLLHLR LAYLHARLRE LLCELGRPSM VWLSANPCPH
15     601    CGDRTFYDPE PILCPCFMPN (TSG) / (LE) / HHHHHH

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No signal sequence used for those constructions

Black = AA 20-127 of protein D

Black - highlighted = 1-Met, 2-Asp and 3-Pro

20 Double underlined = PRAME protein; amino acids 1-509 of PRAME (509 aas total)

Black - underlined = 6 his tail with a linker sequence shown in parentheses that varied or was present depending of the clone

Figure 8
Alignment between LipoD-MAGE3-His (SEQ ID NO:43) and pD1/3-PRAME-His (SEQ ID NO:10)

		1	60
5	LipoD-MAGE3-His	(1) MDPKTLALSLLAAGVLACGSSSSNMANTQMKSDKLTTHRCAGCYLPENTLESKALAPA	
	pD1/3-PRAME-His	(1) MDP-----SSSSNMANTQMKSDKLTTHRCAGCYLPENTLESKALAPA	
		61	120
	LipoD-MAGE3-His	(61) QADYLEQDLAMTQDGRLVVTHDHFLLDGLTDVAKKFPHRRKDCRYVVIDFTLKEIQSL	
	pD1/3-PRAME-His	(45) QADYLEQDLAMTQDGRLVVTHDHFLLDGLTDVAKKFPHRRKDCRYVVIDFTLKEIQSL	
10		121	180
	LipoD-MAGE3-His	(121) MTENPTFMLE-----QRSQHCKPEEGEARGEEIGLVGAQAPATE---QE--	
	pD1/3-PRAME-His	(105) MTENPTFMRRRLWGSIQSRYISMVWTSRRRLRLAGQSLKDESLAIALELLPRELF	
		181	240
	LipoD-MAGE3-His	(165) -----ASSSSSTIVEET-----EG-----EVPAAESP	
15	pD1/3-PRAME-His	(165) PPLFMAAFDRHSQTLKAMVQAWPFTCLPLGVLMKGQHLHLETFKAVLDGLVLLAQEVR	
		241	300
	LipoD-MAGE3-His	(188) L-----PQ-SIPGASSIPPTMNYPLISQSYEDSSNQEE-----EIPSTEP	
	pD1/3-PRAME-His	(225) PRRWLQVLDLRKNQHDFWFWGNRASISFPEPSSAGCPMTKKRKVDGLSTEEQPF	
		301	360
20	LipoD-MAGE3-His	(227) DSEFQAALSRKVAHLHFLLEKYRAREP-----	
	pD1/3-PRAME-His	(285) PVEVLVDLFKEGACDEEFSYLTEKVKRKKNVRLCCKKLKIFAMPQDIKMILKMVQLD	
		361	420
	LipoD-MAGE3-His	(257) -----VT-K-----A-EMIGSYGNWQYFFPVIFSKASSS-----LQLVFGIBL	
	pD1/3-PRAME-His	(345) SIEDLEVTCIWKLP LAKFSPYLGGMLNLRLLLSHIHSSYISPEKEEQYIAQFTSQF	
25		421	480
	LipoD-MAGE3-His	(295) EVDPEGHLYIFFTCLGLSYDGLGDNQMPKAGLLIIVLAARIAGDCAPEEKINEELSV	
	pD1/3-PRAME-His	(405) SIQCQALAYDGLFFLRGRDQLLRHVMPLETLSITNCRSEGGVMHLSQSFSVSQLSV	
		481	540
	LipoD-MAGE3-His	(355) LEQFEGREDSILGDEPKKLTQ-----HFVQENYLEYRQVPGSDPACYEFL	
30	pD1/3-PRAME-His	(465) LSIISGVMLTDVSPHFLQALLERASATLQDLVFDECGITDQLLALLPSLSHCSQLTTLSE	
		541	600
	LipoD-MAGE3-His	(401) GPRVETSIVKYLHIVKISGGPHISYTP-----LHEWMLR	
	pD1/3-PRAME-His	(525) YGNISISLQSLQIQLGSENLTMLYPVPLESYEDINGTLHLERLAYLHARLRRLICE	
		601	642
35	LipoD-MAGE3-His	(438) EGE-----E-----GGHIIIIII	
	pD1/3-PRAME-His	(585) LGRPSMVWLSANPCPHCGDRFTFYDPEPILCPCFMPNIIIIII	

Figure 9

Alignment between the shared sequence of the original protein D from *Haemophilus influenzae* (SEQ ID NO:41), the LipoD-MAGE3-His (SEQ ID NO:45) for use as fusion partner proteins of the present invention

5

10

15

		1	50
Protein D_H influenzae	(1)	MKLKTHALSELLAAGVLAGCSSHSSNMANTQM SSDKITTAHRCASGYLPEH	
LipoD-MAGE3-His	(1)	MDPKTHALSLIPLAAGVLAGCSSHSSNMANTQM KSKITTAHRCASGYLPEH	
Consensus	(1)	M KTLALSLLAAGVLAGCSSHSSNMANTQM	KSKITTAHRCASGYLPEH
		51	100
Protein D_H influenzae	(51)	TLESKALAFACQADYLEQDLAMTKDGRLLVVIHDFLDGLTDVAKKFP PHRH	
LipoD-MAGE3-His	(51)	TLESKALAFACQADYLEQDLAMTKDGRLLVVIHDFLDGLTDVAKKFP PHRH	
Consensus	(51)	TLESKALAFACQADYLEQDLAMTKDGRLLVVIHDFLDGLTDVAKKFP	PHRH
		101	127
Protein D_H influenzae	(101)	RKDGRYYVIDFTLKEIQSLEMTENFET	
LipoD-MAGE3-His	(101)	RKDGRYYVIDFTLKEIQSLEMTENFET	
Consensus	(101)	RKDGRYYVIDFTLKEIQSLEMTENFET	

Figure 10

Alignment between the shared sequence of the original protein D from *Haemophilus influenzae* (SEQ ID NO:41), the LipoD-MAGE3-His (SEQ ID NO:45) and the pD1/3-PRAME-His (SEQ ID NO:44) for use as fusion partner proteins of the present invention

		1	50
Protein D_H influenzae	(1)	MKLSTSLLAAGVLAGCSSHSSNMANTQMKSDKIIIAHRGASGYLPEH	
LipoD-MAGE3-His	(1)	MDPKTIAVSLDAAGVLAGCSSHSSNMANTQMKSDKIIIAHRGASGYLPEH	
pD1/3-PRAME	(1)	-----MSSHSSNMANTQMKSDKIIIAHRGASGYLPEH	
Consensus	(1)	M KTLALSLLAAGVLAGCSSHSSNMANTQMKSDKIIIAHRGASGYLPEH	
		51	100
Protein D_H influenzae	(51)	TLESKALAFQAQADYLEQDLAMTKDGRLVVIHDFLDGLTDVAKKPPHRH	
LipoD-MAGE3-His	(51)	TLESKALAFQAQADYLEQDLAMTKDGRLVVIHDFLDGLTDVAKKPPHRH	
pD1/3-PRAME	(33)	TLESKALAFQAQADYLEQDLAMTKDGRLVVIHDFLDGLTDVAKKPPHRH	
Consensus	(51)	TLESKALAFQAQADYLEQDLAMTKDGRLVVIHDFLDGLTDVAKKPPHRH	
		101	127
Protein D_H influenzae	(101)	RKDGRYVIDFTLKEIQSLEMTENFET	
LipoD-MAGE3-His	(101)	RKDGRYVIDFTLKEIQSLEMTENFET	
pD1/3-PRAME	(83)	RKDGRYVIDFTLKEIQSLEMTENFET	
Consensus	(101)	RKDGRYVIDFTLKEIQSLEMTENFET	

Figure 11

SDS page analysis of pD1/3-PRAME with or without secretion signal (SS) and His-tail (His) in pET21 vector.

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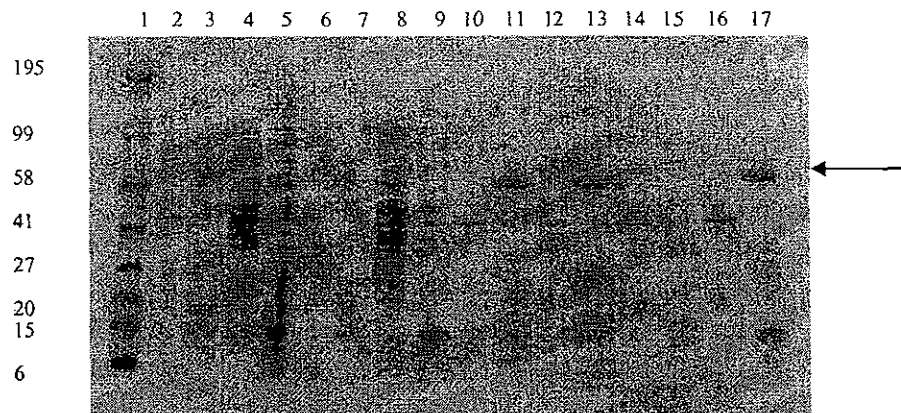


Figure 12

Sequence of protein D 1/3 MAGE-A3-His (SEQ ID NO;43)

N term MDP	protD 1/3	Met Asp	Mage 3	GlyGly 7xHis	C term
5	2	124	3	314	

```

MDPKTLALSLLAAGVLAGCSSHSSNMANTOMKSDKIIIAH 40
RGASGYLPEHTLESKALAFQAQQADYLEQDLAMTKDGRLLV 80
10  IHDHFLDGLTDVAKKFPHRHRKDGRYYVIDFTLKEIQSLE 120
MTENFETMDLEQRSQHCKPEEGLEARGEALGLVGAQAPAT 160
EEQEAASSSSTLVEVTLGEVPAAESPDPPQSPQGASSLPT 200
TMNYPLWSQSYEDSSNQEEEGPSTFPDLESEFQAALSRKV 240
AELVHFLLLKYRAREPVTKAEMLGSVVGNWQYFFPVIFSK 280
15  ASSSLQLVFGIELMEVDPIGHLIYFATCLGLSYDGLLGDN 320
QIMPKAGLLIIVLAI IAREGDCAPEEKIWEELSVLEVFEG 360
REDSILGDPKKLLTQHFVQENYLEYRQVPGSDPACYEFLW 400
GPRALVETSYVKVLHMHMVKISGGPHISYPPLHEWVLREGE 440
EGGHHHHHHH 451

```

Underlined = signal sequence of Protein D including 1-Met and the substitutions 2-Asp and 3-Pro for the native aa 2-Lys and 3-Thr of protein D

Double underlined = amino acids 20 to 127 of Protein D

MD = unrelated amino acids Met-Asp at aa 128-129 to create a cloning site

GG = unrelated amino acids Gly-Gly at 442-443

Dotted underlined = fragment of MAGE3; amino acids 3-314 of MAGE3 (312 aas total)

HHHHHHH = 7 his tail