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C07K 14/13 (2006.01), *C07K 14/135* (2006.01),
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(54) Titre : PROCEDES ET COMPOSITIONS POUR EMPECHER CERTAINS PHENOMENES ASSOCIES AVEC LA FUSION AVEC LA MEMBRANE, EN PARTICULIER LA TRANSMISSION DU VIH

(54) Title: METHODS AND COMPOSITIONS FOR INHIBITION OF MEMBRANE FUSION-ASSOCIATED EVENTS, INCLUDING HIV TRANSMISSION

HIV1LAI (DP-178; SEQ ID:1)	YTSLIHSLIEESQNQQEKNEQELLELDKWASLWNWF
HIV1SF2 (DP-185; SEQ ID:3)	YTNTIYNLLEESQNQQEKNEQELLELDKWASLWNWF
HIV1RF (SEQ ID:4)	YTGIIYNLLEESQNQQEKNEQELLELDKWANLWNWF
HIV1MN (SEQ ID:5)	YTSLIYSLLEKSQTQQEKNEQELLELDKWASLWNWF
HIV2ROD (SEQ ID:6)	LEANISKSLEQAQIQQEKNMYELQKLNSWDIFGNWF
HIV2NIHZ (SEQ ID:7)	LEANISQSLEQAQIQQEKNMYELQKLNSWDVFTNWL
DP180 (SEQ ID:2)	SSESFTLLEQWNNWKLQLAEQWLEQINEKHYLEDIS
DP118 (SEQ ID:10)	QQLLDVVKRQQEMLRLTVWGTKNLQARVTAIEKYLKDQ
DP125 (SEQ ID:8)	CGGNNLLRAIEAQQHLLQLTVWGIKQLQARILAVERYLKDQ
DP116 (SEQ ID:9)	LQARILAVERYLKDQQQ

(57) Abrégé/Abstract:
The present invention relates to peptides which exhibit potent anti-retroviral activity. The peptides of the invention comprise DP178 (SEQ ID:1) peptide corresponding to amino acids 638 to 673 of the HIV-1_{LAI} gp4I protein, and fragments, analogs and homologs

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(74) Agent: OSLER, HOSKIN & HARCOURT LLP

(57) Abrégé(suite)/Abstract(continued):
of DP178. The invention further relates to the uses of such peptides as inhibitory of human and non-human retroviral, especially HIV, transmission to uninfected cells.

CORRECTED VERSION*

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PCT

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(54) Title: METHODS AND COMPOSITIONS FOR INHIBITION OF MEMBRANE FUSION-ASSOCIATED EVENTS, INCLUDING HIV TRANSMISSION

(57) Abstract

The present invention relates to peptides which exhibit potent anti-retroviral activity. The peptides of the invention comprise DP178 (SEQ ID:1) peptide corresponding to amino acids 638 to 673 of the HIV-1_{LAI} gp41 protein, and fragments, analogs and homologs of DP178. The invention further relates to the uses of such peptides as inhibitory of human and non-human retroviral, especially HIV, transmission to uninfected cells.

* (Referred to in PCT Gazette No. 44/1996, Section II)

The embodiments of the invention in which an exclusive property or privilege is claimed are defined as follows:

1. An isolated peptide having the formula:

X-YTSLIHSLIEESQNQQEKNEQELLELDKWASLWNAF-Z;

X-YTSLIHSLIEESQNQQEKNEQELLELDKWASLANWF-Z;

X-YTSLIHSLIEESQNQQEKNEQQLLELDKWASLWNWF-Z;

X-YTSLIHSLIEESQNQQEKNEQELLQLDKWASLWNWF-Z;

X-YTSLIHSLIEESQNQQEKNQQELLQLDKWASLWNWF-Z;

X-YTSLIHSLQEESQNQQEKNEQELLELDKWASLWNWF-Z;

X-YTSLIHSLIEQSQNQQEKNEQELLELDKWASLWNWF-Z;

X-YTSLIHSLIQESQNQQEKNEQELLELDKWASLWNWF-Z;

X-YTSLIHSLIEESQNQQEKNEQQLLELDKWASLWNWF-Z;

X-YTSLIQSLIEESQNQQEKNEQQLLELDKWASLWNWF-Z;

X-YTSLIHSLIEESQNQQEKNEQELLELDKWASLFNFF-Z;

X-YTSLIHSLIEESQNLQEKNEQELLELDKWASLWNWF-Z;

X-YTSLIHSLIEESQNQQEKLEQELLELDKWASLWNWF-Z;

X-YTSLIHSLIEESQNQQEKNEQELLEFDKWASLWNWF-Z;

X-YTSLIHSLIEESQNQQEKNEQELLELDKWASPWNWF-Z;

X-YTSLIHSLIEESQNQQEKNEQELLELDKWASLWNSF-Z;

X-LRAIEAQQHLLQLTVWQIKQLQARILAV-Z;

X-NKSLEQIWNNMTWMEWDREINNYTSLIHSLIEESQN-Z;

X-NKSLEQIWNNMTWMEWDREINNYTSLIHSLIEESQNQQEKNEQELLELDKWASLW
NWF-Z;

X-SLEQIWNNMTWMEWDREINNYTSLIHSLIEESQNQQ-Z;

X-LEQIWNNMTWMEWDREINNYTSLIHSLIEESQNQQE-Z;

X-EQIWNNMTWMEWDREINNYTSLIHSLIEESQNQQEK-Z;

X-QIWNNMTWMEWDREINNYTSLIHSLIEESQNQQEKN-Z;

X-IWNNMTWMEWDREINNYTSLIHSLIEESQNQQEKNE-Z;

X-WNNMTWMEWDREINNYTSLIHSLIEESQNQQEKNEQ-Z;

X-NNMTWMEWDREINNYTSLIHSLIEESQNQQEKNEQE-Z;

X-NMTWMEWDREINNYTSLIHSLIEESQNQQEKNEQEL-Z;

X-MTWMEWDREINNYTSLIHSLIEESQNQQEKNEQELLELDKWASLWNW-Z;

X-MTWMEWDREINNYTSLIHSLIEESQNQQEKNEQELL-Z;

X-TWMEWDREINNYTSLIHSLIEESQNQQEKNEQELLE-Z;

X-WMEWDREINNYTSLIHSLIEESQNQQEKNEQELLEL-Z;

X-WMEWDREINNYTSLIHSLIEESQNQQEKNEQELLE-Z;

X-WMEWDREINNYTSLIGSLIEESQNQQEKNEQELLE-Z;

X-MEWDREINNYTSLIHSLIEESQNQQEKNEQELLELD-Z;

X-EWDREINNYTSLIHSLIEESQNQQEKNEQELLELDK-Z;

X-WDREINNYTSLIHSLIEESQNQQEKNEQELLELDKW-Z;

X-DREINNYTSLIHSLIEESQNQQEKNEQELLELDKWA-Z;

X-REINNYTSLIHSLIEESQNQQEKNEQELLELDKWAS-Z;

X-EINNYTSLIHSLIEESQNQQEKNEQELLELDKWASL-Z;

X-INNYTSLIHSLIEESQNQQEKNEQELLELDKWASLW-Z;

X-NYTSLIHSLIEESQNQQEKNEQELLELDKWASLWNW-Z;

X-TSLIHSLIEESQNQQEKNEQELLELDKWASLWNWFN-Z;

X-SLIHSLIEESQNQQEKNEQELLELDKWASLWNWFNI-Z;

X-LIHSLIEESQNQQEKNEQELLELDKWASLWNWFNIT-Z; or

X-YTSLIHSLIEESQNQQEKNEQELLELDKWASLWNWFNITNWLWLIKFI-Z;

in which X comprises an amino group, an acetyl group, a 9-fluorenylmethoxy-carbonyl group, a hydrophobic group, or a macromolecular carrier group; and Z comprises a carboxyl group, an amido group, a hydrophobic group, or a macromolecular carrier group, wherein the peptide exhibits antiviral activity.

2. The peptide of claim 1, wherein X is an amino group and Z is a carboxyl group.

3. The peptide of claim 1, wherein the hydrophobic group is a carbobenzoxyl, dansyl or t-butyloxycarbonyl hydrophobic group.

4. The peptide of claim 1, wherein X is an acetyl group or a 9-fluorenylmethoxy-carbonyl group; and Z is a t-butyloxycarbonyl hydrophobic group or an amido group.

5. The peptide of claim 4, wherein X is an acetyl group and Z is an amido group.

6. The peptide of claim 1, wherein Z is a lipid-fatty acid conjugate, a polyethylene glycol, or a carbohydrate moiety.

7. The peptide according to any one of claims 1 to 6, wherein one bond linking adjacent amino acids is a non-peptide bond, further wherein the non-peptide bond is an imino, ester, hydrazine, semicarbazide, or azo bond.

8. The peptide according to any one of claims 1 to 7, wherein at least one amino acid residue is in a D-isomer configuration.

9. The peptide according to any one of claims 1 to 8 for use as an antiviral agent in the treatment of HIV-1 infection.

10. Use of the peptide according to any one of claims 1 to 8 for the manufacture of a medicament for the treatment of HIV-1 infection.

11. Use of the peptide according to any one of claims 1 to 8 in the treatment of HIV-1 infection.

12. A pharmaceutical composition for use in inhibiting transmission of an HIV virus to a cell, comprising the peptide according to any one of claims 1 to 8 as active component and a pharmaceutically acceptable carrier.

13. An isolated nucleic acid encoding the peptide of claim 1.

14. A vector comprising a nucleic acid encoding the peptide of claim 1.

15. A peptide produced by expression of a vector comprising a nucleic acid encoding the peptide of claim 1.

RECTIFIED SHEET (RULE 91)

HIV1LAI	(DP-178; SEQ ID:1)	YTSLIHSLIEESQNQQEKNEQELLELDKWASLWNWF
HIV1SF2	(DP-185; SEQ ID:3)	YTNTIYNLLEESQNQQEKNEQELLELDKWASLWNWF
HIV1RF	(SEQ ID:4)	YTGIIYNLLEESQNQQEKNEQELLELDKWANLWNWF
HIV1MN	(SEQ ID:5)	YTSLIYSLLEKSQTQQEKNEQELLELDKWASLWNWF
HIV2ROD	(SEQ ID:6)	LEANISKSLEQAQIQQEKNMYELQKLNSWDIFGNWF
HIV2NIHZ	(SEQ ID:7)	LEANISQSLEQAQIQQEKNMYELQKLNSWDVFTNWL
DP180	(SEQ ID:2)	SSESFTLLEQWNNWKLQLAEQWLEQINEKHYLEDIS
DP118	(SEQ ID:10)	QQLLDVVKRQQEMLRLTVWGTKNLQARVTAIEKYLKDQ
DP125	(SEQ ID:8)	CGGNNLLRAIEAQQHLLQLTVWGIKQLQARILAVERYLKDQ
DP116	(SEQ ID:9)	LQARILAVERYLKDQQQ

FIG.1

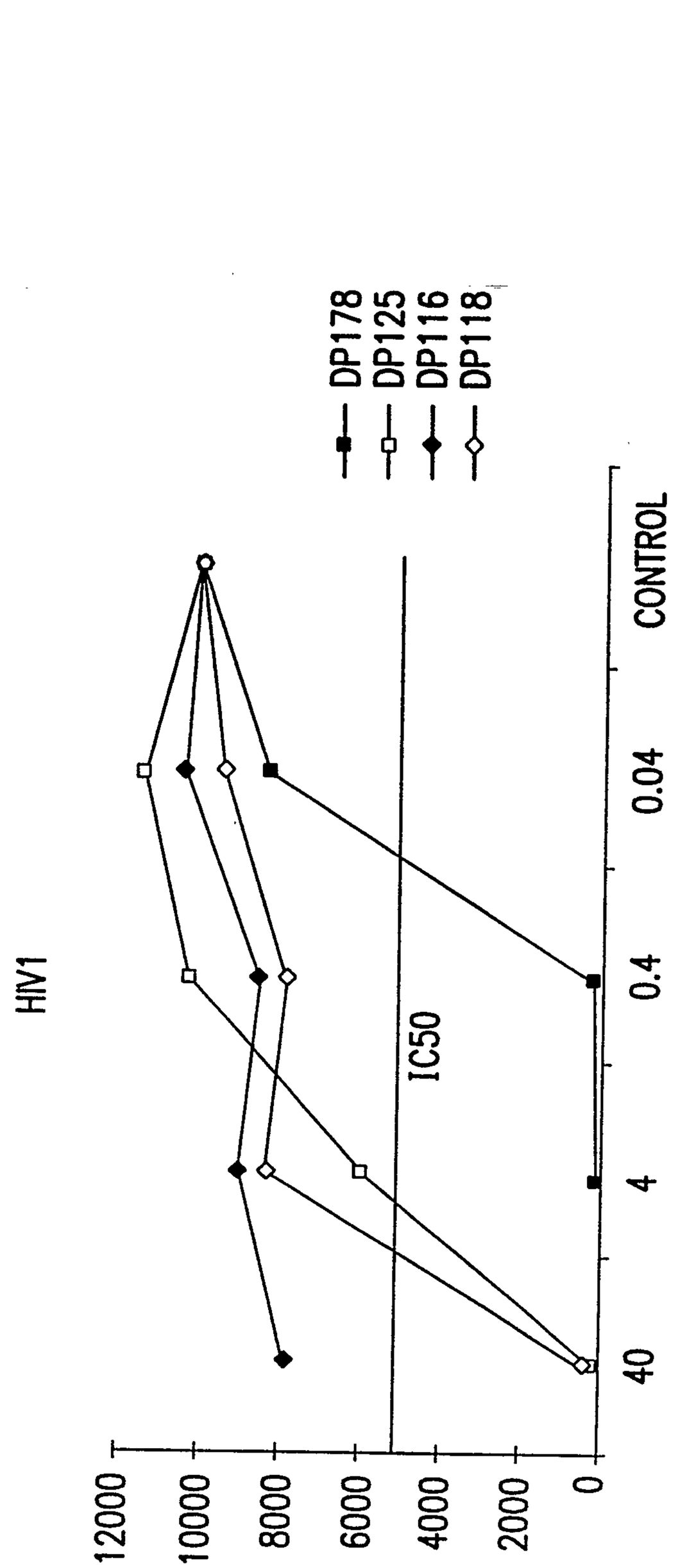
DP178
DP125
DP116
DP118
HIV1
IC50
CONTROL
0.04
0.4
4
40
CONC. ug/ml
12000
10000
8000
6000
4000
2000
0
REVERSE TRANSCRIPTASE UNITS

FIG.2

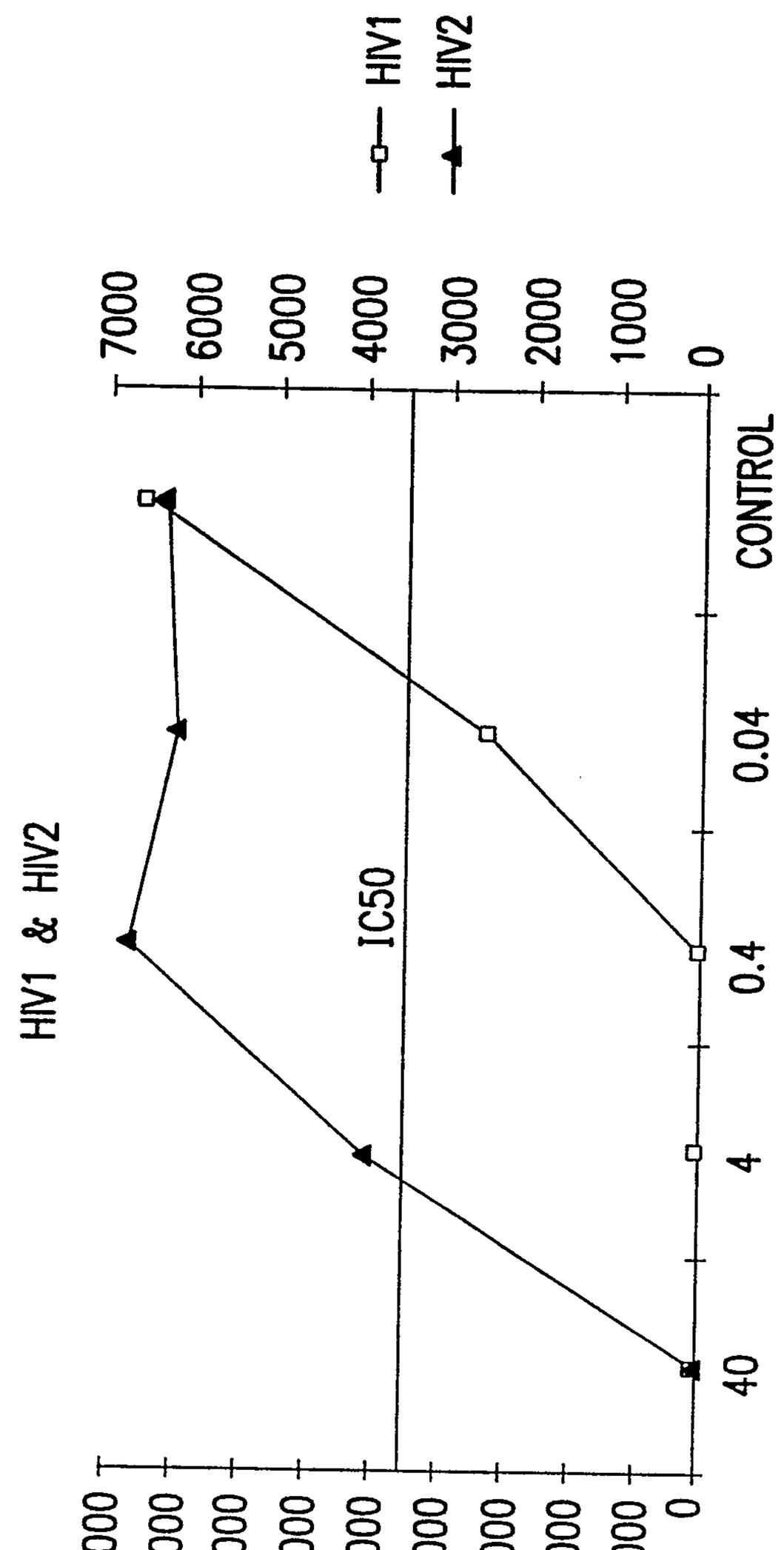
HIV1
HIV2
7000
6000
5000
4000
3000
2000
1000
0
CONTROL
HIV1 & HIV2
IC50
0.04
0.4
4
40
PD178 CONC. ug/ml
9000
8000
7000
6000
5000
4000
3000
2000
1000
0

FIG.3

Number of Syncytia/well: concentration in μg/ml (micrograms/ml)

DP178	10	5	1	0.2	0.1	0.05	0.025	0.0125	Control
Syncytia									
HIV1LAI	0	0	0	0	0	0	0	0	67
HIV1MN	0	0	0	0	0	ND	ND	ND	34
HIV1RF	0	0	0	0	0	ND	ND	ND	65
HIV1SF2	0	0	0	0	0	ND	ND	ND	58

DP125	10	5	1	0.2	0.1	0.05	0.025	0.0125	Control
Syncytia									
HIV1LAI	0	0	54	69	80	75	79	82	67
HIV1MN	0	0	30	36	ND	ND	ND	ND	34
HIV1RF	0	0	67	63	ND	ND	ND	ND	65
HIV1SF2	0	0	9	66	ND	ND	ND	ND	58

DP116	10	5	1	0.2	0.1	0.05	0.025	0.0125	Control
Syncytia									
HIV1LAI	75	ND	ND	ND	ND	ND	ND	ND	67
HIV1MN	35	ND	ND	ND	ND	ND	ND	ND	34
HIV1RF	81	ND	ND	ND	ND	ND	ND	ND	65
HIV1SF2	81	ND	ND	ND	ND	ND	ND	ND	58

FIG.4A

DP180	40	20	10	5	2.5	1.25	0.625	0.3125	Control
Syncytia									
HIV1LAI	50	>45	>45	>45	>45	>45	>45	>45	58

DP185	40	20	10	5	2.5	1.25	0.625	0.3125	Control
Syncytia									
HIV1LAI	0	0	0	0	0	0	0	ND	60

FIG.4B

HIV1

	Number of Syncytia/well: concentration in ng/ml (nanograms/ml)							
DP178	20	10	5	2.5	1.25	0.625	0.3125	Control
Syncytia HIV1	0	0	0	0	0	14	20	48
DP116	20	10	5	2.5	1.25	0.625	0.3125	Control
Syncytia HIV1	ND	48	ND	ND	ND	ND	ND	ND

HIV2

	Number of Syncytia/well: concentration in μg/ml (micrograms/ml)							
DP178	20	10	5	2.5	1.25	0.625	0.3125	Control
Syncytia HIV2	50	54	55	57	63	77	78	76
DP116	20	10	5	2.5	1.25	0.625	0.3125	Control
Syncytia HIV2	ND	58	ND	ND	ND	ND	ND	ND

FIG.5

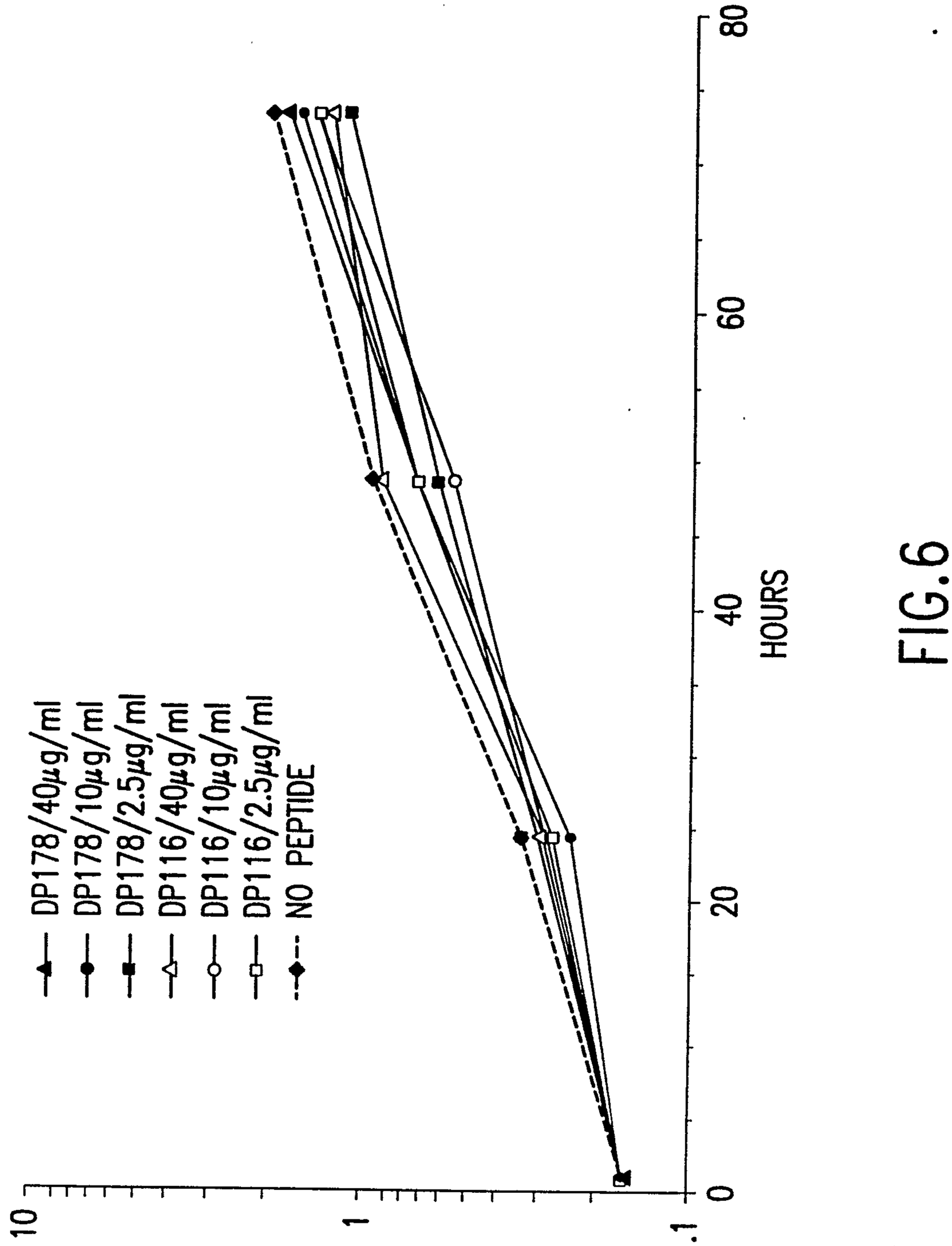
CELL CONCENTRATION X 10^6/ml
10
1
.1
0
20
40
HOURS
60
80
DP178/40μg/ml
DP178/10μg/ml
DP178/2.5μg/ml
DP116/40μg/ml
DP116/10μg/ml
DP116/2.5μg/ml
NO PEPTIDE

FIG.6

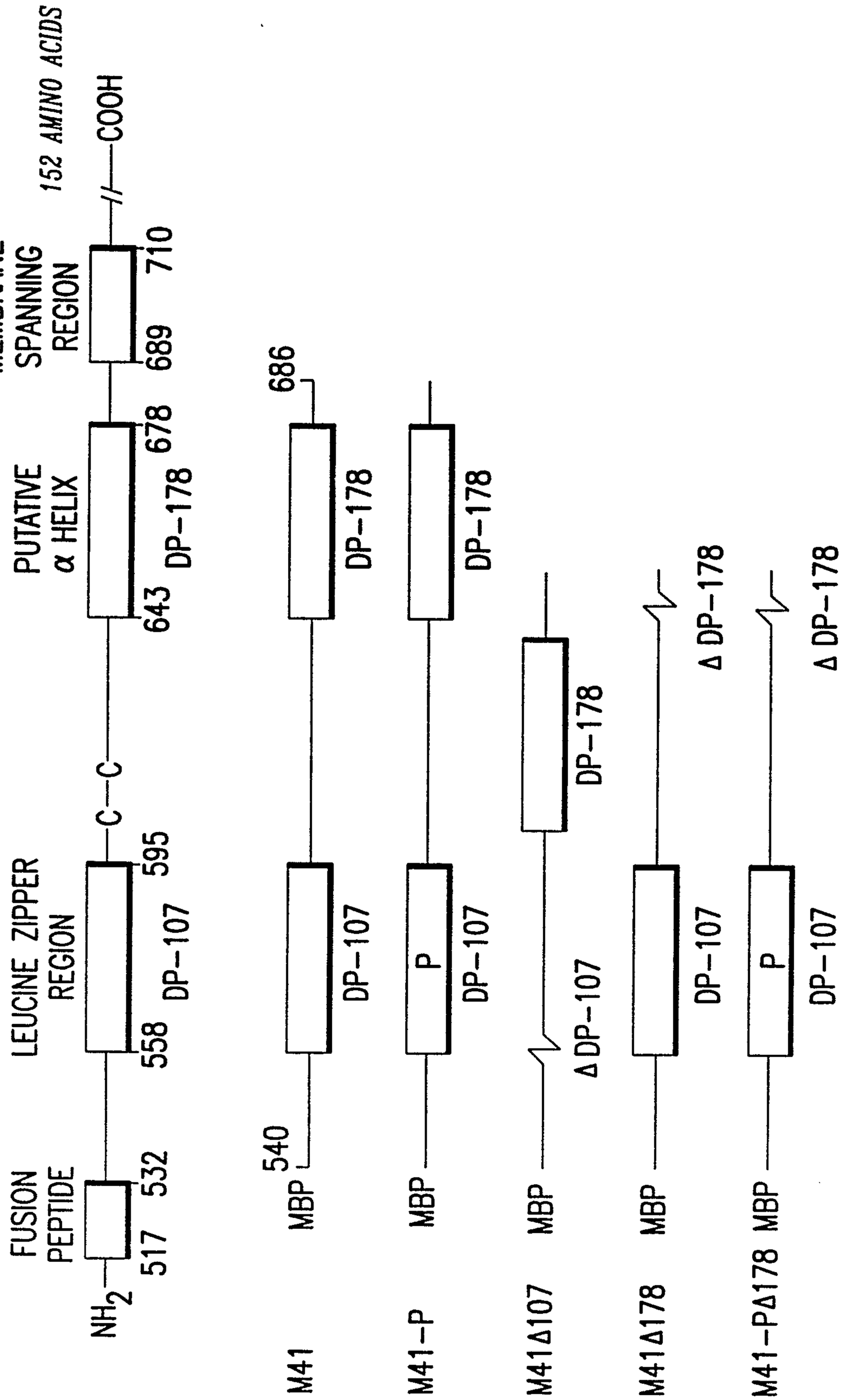
152 AMINO ACIDS
COOH
MEMBRANE SPANNING REGION
710
689
PUTATIVE α HELIX
678
643
DP-178
686
DP-178
DP-178
LEUCINE ZIPPER REGION
C—C
595
558
DP-107
DP-107
P
DP-107
DP-178
ΔDP-107
DP-107
P
DP-107
FUSION PEPTIDE
NH_2
532
517
540
MBP
M41
MBP
M41-P
MBP
M41Δ107
MBP
M41Δ178
MBP
M41-PΔ178
MBP

FIG.7

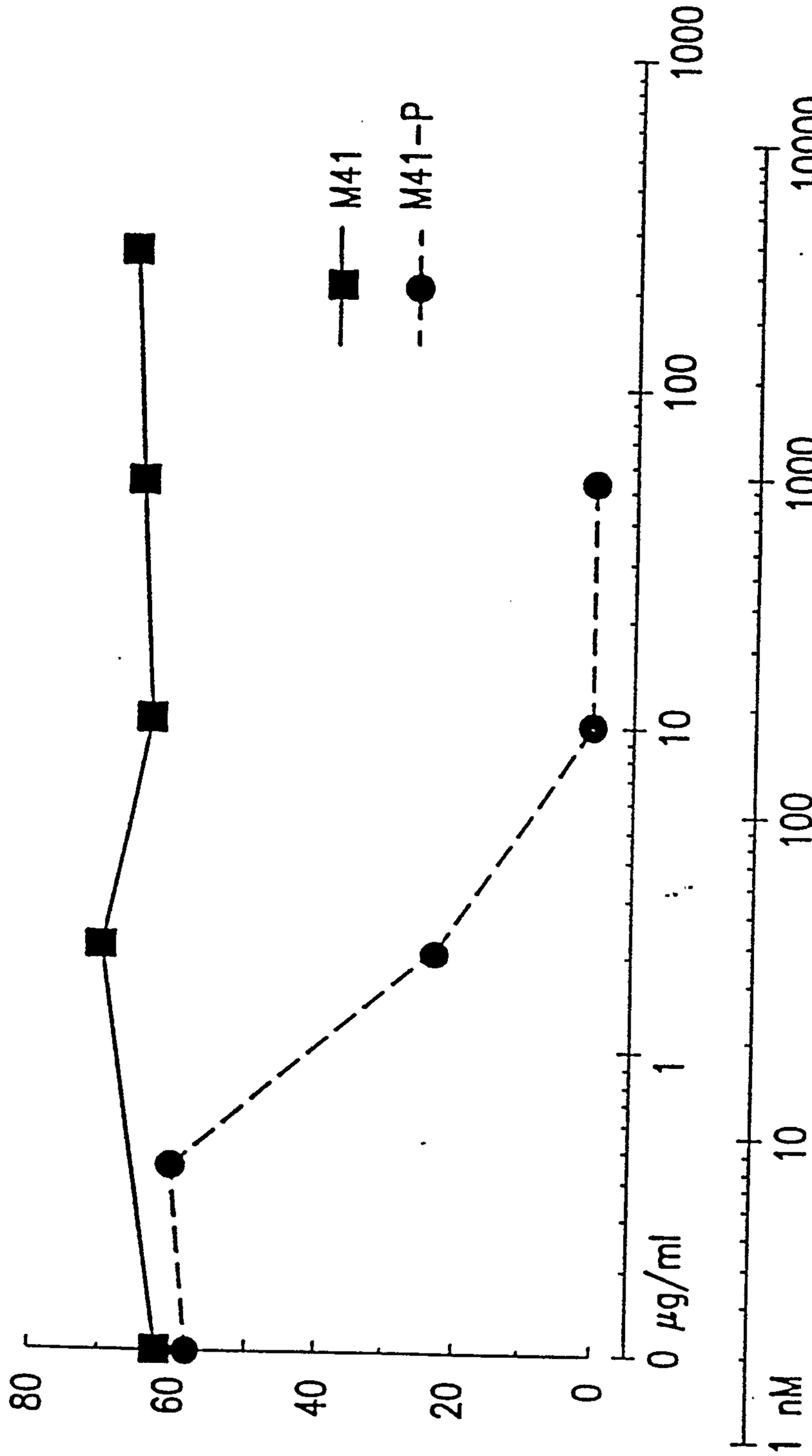
M41
M41-P
NUMBER OF SYNCYTIA
80
60
40
20
0
0 µg/ml
1 nM
10
1
10
100
1000
10000
1000
100
10
1

FIG.8

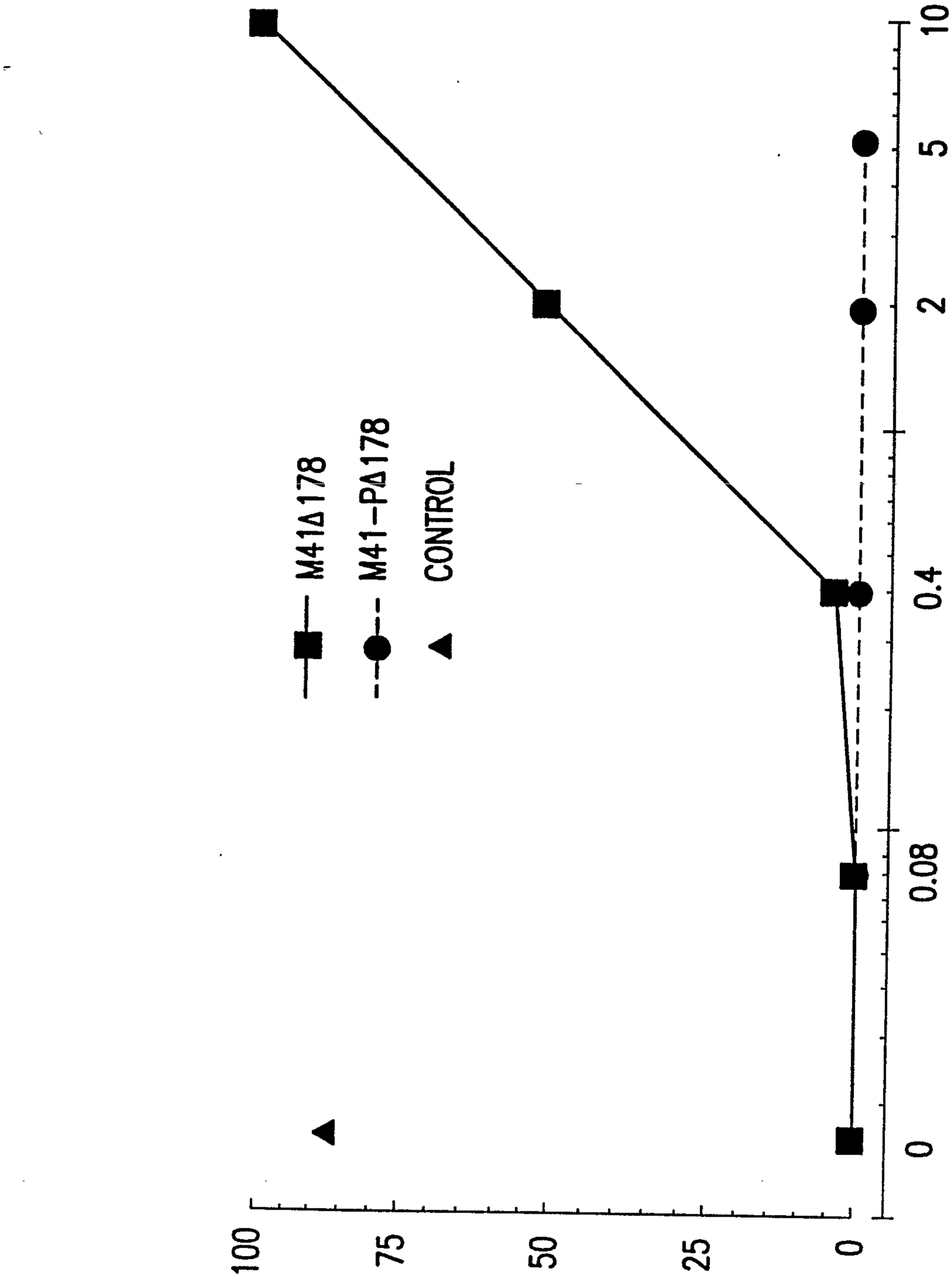
M41Δ178
M41–PΔ178
CONTROL
NUMBER OF SYNCYTIA
100
75
50
25
0
10
5
2
0.4
0.08
0
μg/ml

FIG.9

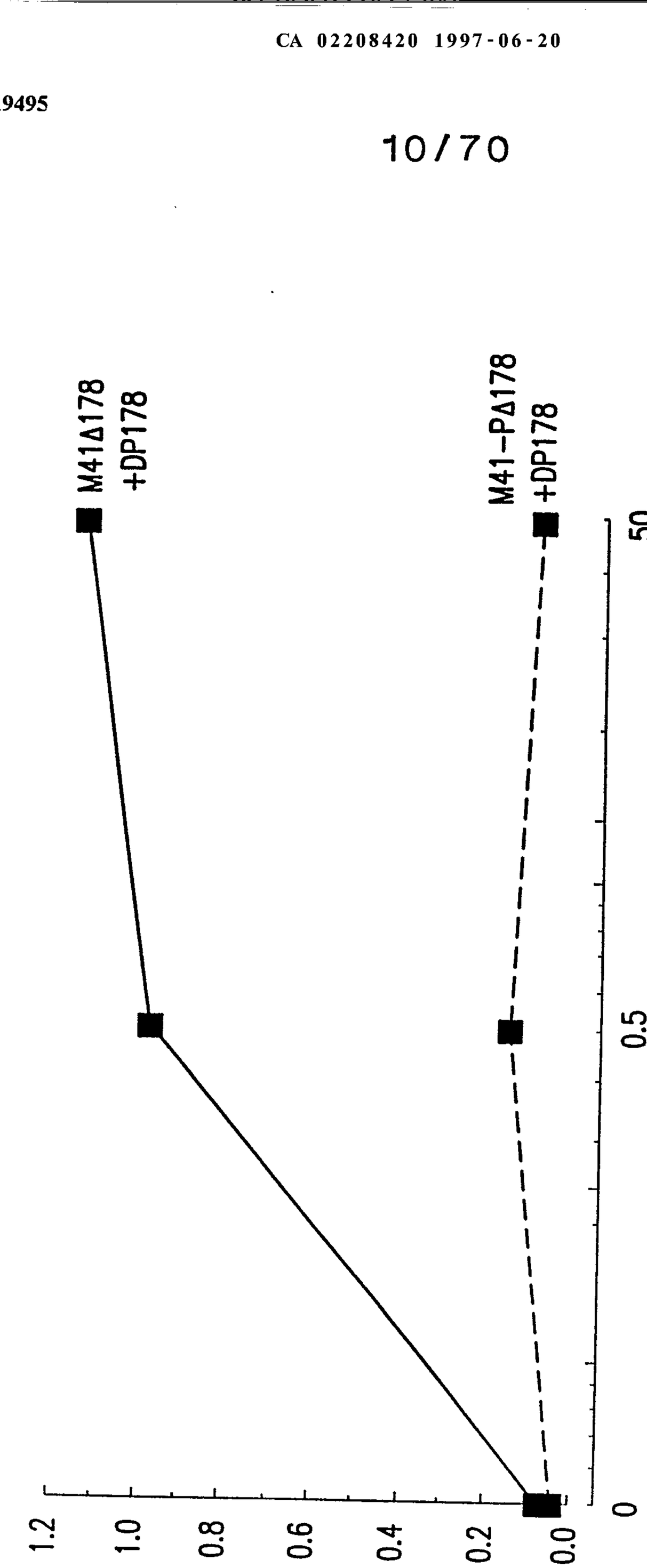
M41Δ178
+DP178
M41-PΔ178
+DP178
50
0.5
0
PEPTIDE CONCENTRATION (μg/ml)
1.2
1.0
0.8
0.6
0.4
0.2
0.0
OD (490 nm)

FIG.10

WO 96/19495 PCT/US95/16733

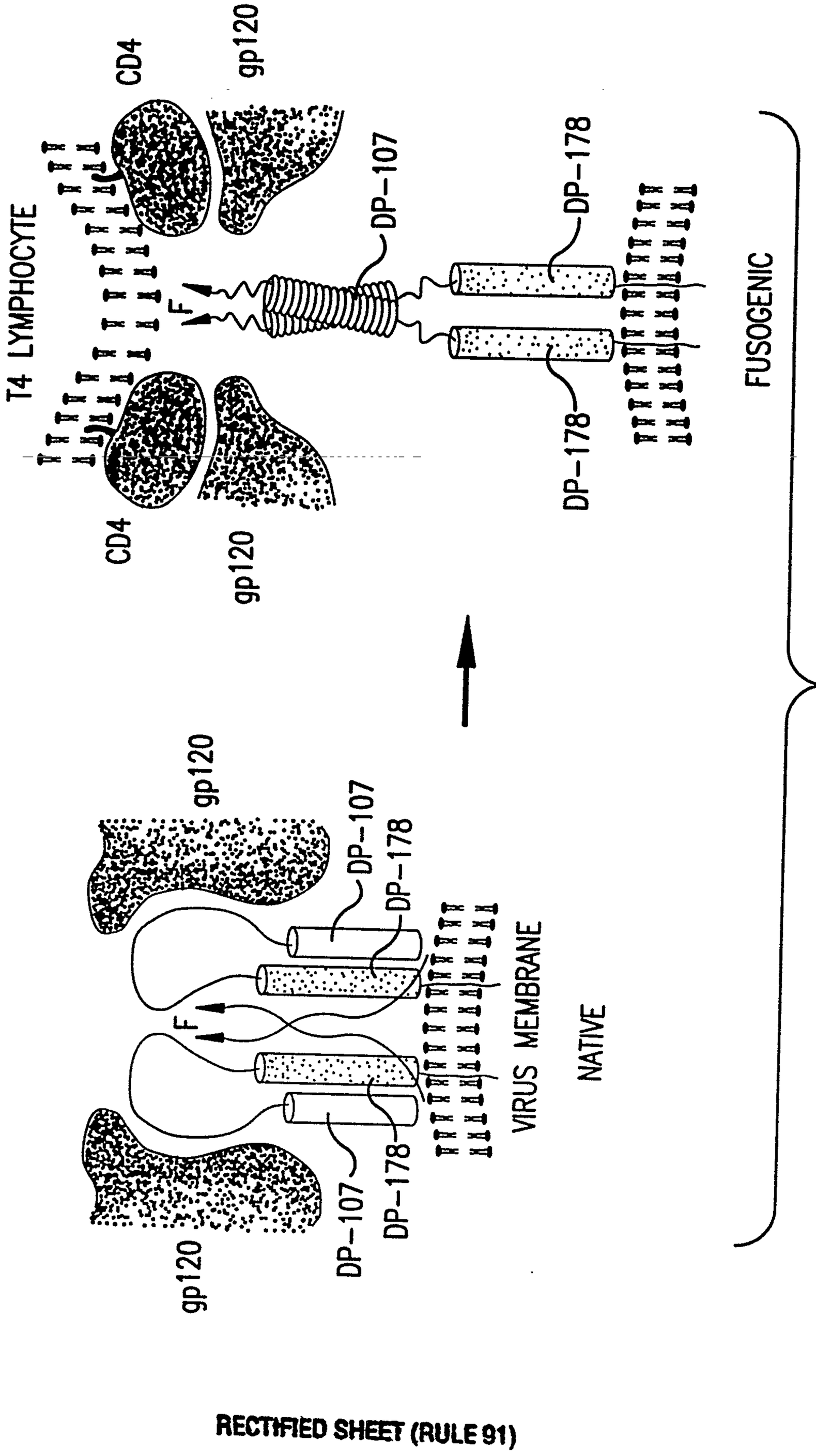

FIG.11A

WO 96/19495 PCT/US95/16733

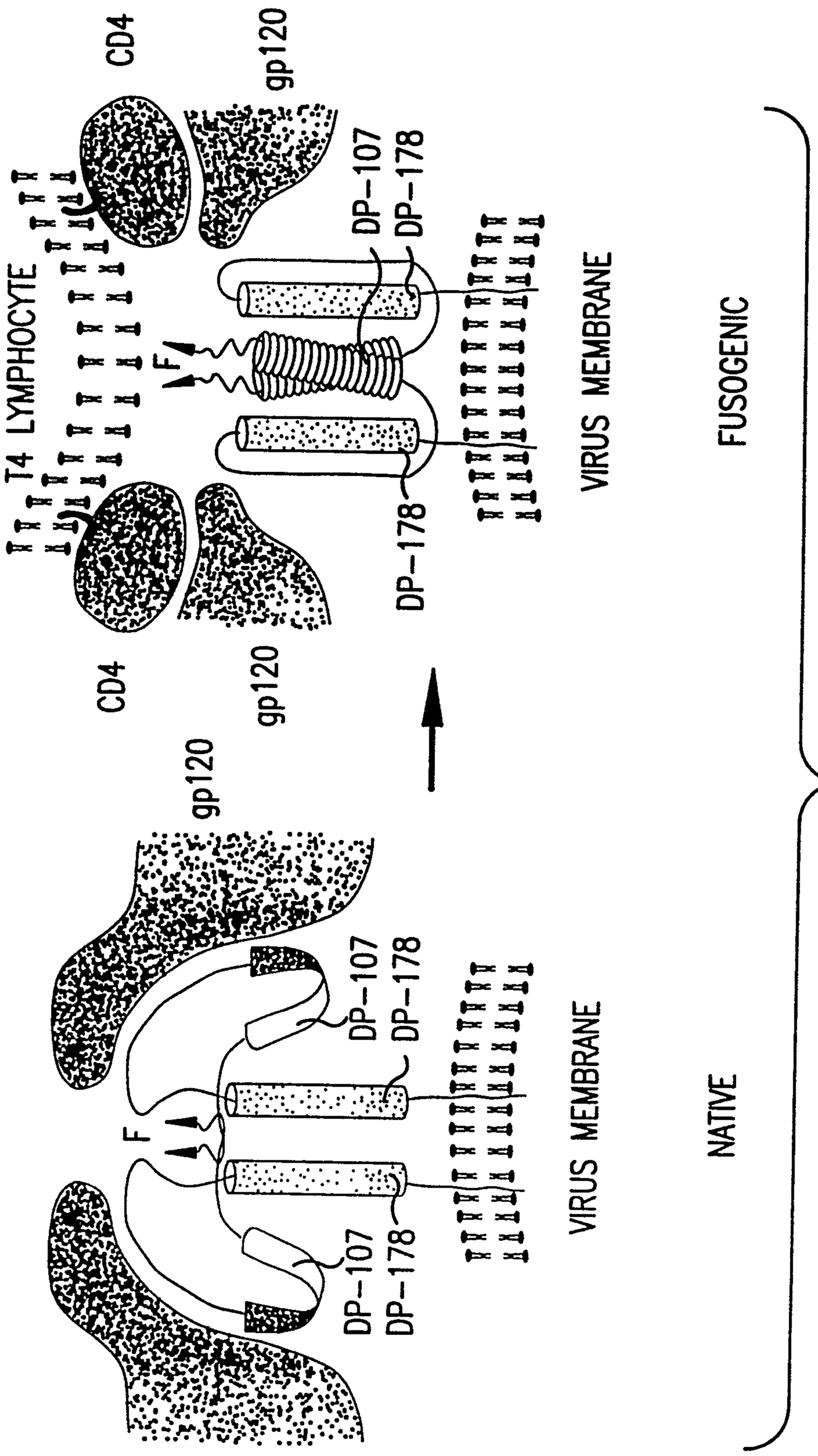

FIG.11B

| Sequence | Positions | | |

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RECTIFIED SHEET (RULE 91)

| Sequence | Positions | | |

| Sequence | Positions | | |

| Sequence | A | D | A | D | Positions | | |

WO 96/19495

PCT/US95/16733

Hybrid Motif

[EFIKLNQSTVWY] {CFMP}

Parent Motif

[ILQTV] {CDFIMPST}
[EKLNQV] {CFKMPS}
[EFKLQWY] {CFGMPRVY}
[EFILNQSWY] {CFGMPRVY}

[FILTV] {ACFLMPTVW}

Positions

| Sequence | A | | | D | | | D | | A | | D | | A | | D | | A

CA 02208420 1997-06-20

RECTIFIED SHEET (RULE 91)

| Sequence | Positions | | |

Hybrid Motif

[AEFIKLMNQRSTVWY] {CFP}
{CDGHP} {CFP}
=

Parent Motif

[LMNV] {CFGIMPTW}
[ILQTV] {CDFIMPST}
[EKLNQV] {CFKMPS}
[EFKLQWY] {CFGMPRVY}
[EFILNQSWY] {CFGMPRVY}
[IKLT] {CFGHIMPRVWY}
[AILNV] {CDFGHILPVWY}
[ELR] {ACFGMPVWY}
[FILTV] {ACFLMPTVW}

Positions

Sequence	A		D			A			D			A			D			A			D			A			D		
GCN4 (gcn4 yeast)	M	K	Q	L	E	D	K	V	E	E	L	S	K	N	Y	H	L	E	N	E	V	A	R	L	K	K	L	E	D
DP-107 (env_hv1bru)L1=D		N	N	L	L	R	A	I	E	A	Q	Q	H	L	Q	L	T	V	W	G	I	K	Q	L	Q	A	R	I	L
DP-107 (env_hv1bru)L2=D	N	N	L	L	R	A	I	E	A	Q	Q	H	L	L	Q	L	T	V	W	G	I	K	Q	L	Q	A	R	I	L
DP-178 (env_hv1bru)Y1=A	Y	T	S	L	I	H	S	L	I	E	E	S	Q	N	Q	E	K	N	E	Q	E	L	L	E	L	D	K	W	A
DP-178 (env_hv1bru)Y1=D			Y	T	S	L	I	H	S	L	I	E	E	S	Q	N	Q	Q	E	K	N	E	Q	E	L	L	E	L	D
C-FOS (fos_human)	T	D	T	L	Q	A	E	T	D	Q	L	E	D	E	K	S	A	L	Q	T	E	I	A	N	L	L	K	E	
C-JUN (top1_human)	I	A	R	L	E	E	A	E	T	L	K	A	Q	N	S	E	L	A	S	T	A	N	M	L	R	E	Q		
C-MYC (myo_human)	E	Q	K	L	I	S	E	E	D	L	L	R	K	R	E	Q	L	K	H	K	L	E	Q	L	R	N	S		
FLU LOOP 36	I	E	K	T	N	E	T	V	E	K	K	F	H	Q	I	E	K	E	F	S	E	V	E	G	R	I	Q	D	L	E	K	Y

FIG.18

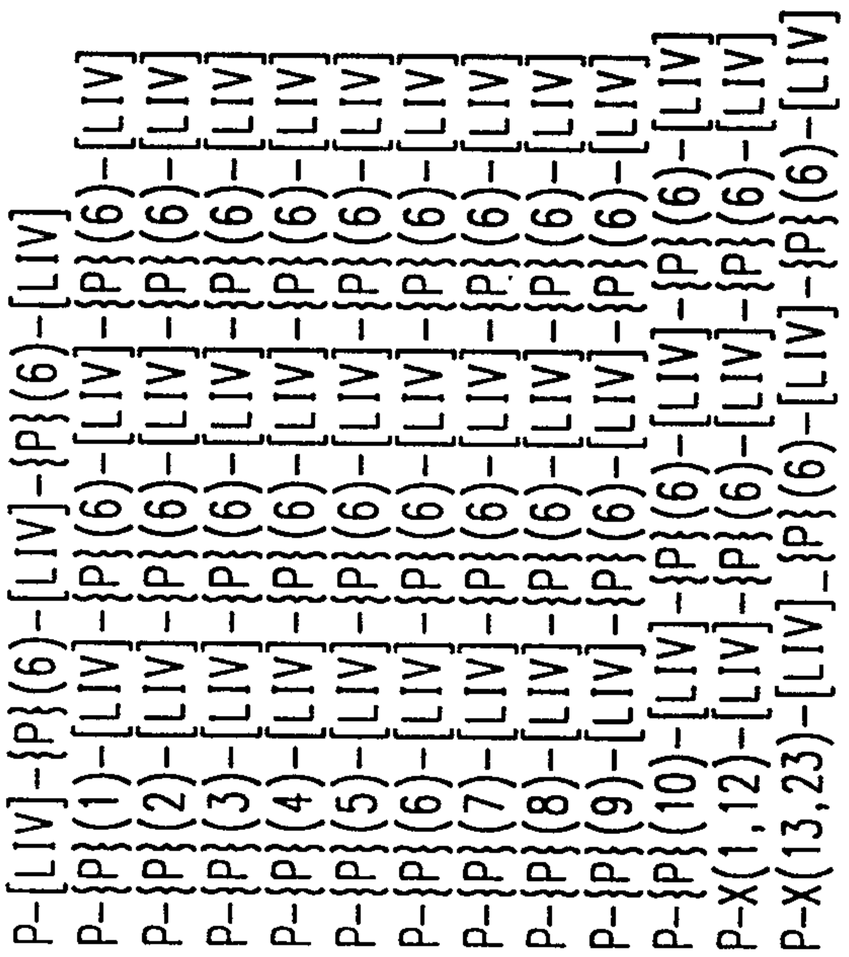

P–[LIV]–{P}(6)–[LIV]
P–{P}(1)–[LIV]–{P}(6)–[LIV]
P–{P}(2)–[LIV]–{P}(6)–[LIV]
P–{P}(3)–[LIV]–{P}(6)–[LIV]
P–{P}(4)–[LIV]–{P}(6)–[LIV]
P–{P}(5)–[LIV]–{P}(6)–[LIV]
P–{P}(6)–[LIV]–{P}(6)–[LIV]
P–{P}(7)–[LIV]–{P}(6)–[LIV]
P–{P}(8)–[LIV]–{P}(6)–[LIV]
P–{P}(9)–[LIV]–{P}(6)–[LIV]
P–{P}(10)–[LIV]–{P}(6)–[LIV]
P–X(1,12)–[LIV]–{P}(6)–[LIV]
P–X(13,23)–[LIV]–{P}(6)–[LIV]

FIG.19

Fusion ♥ALLMOTI5♥
Peptide ♠107x178x4♠
♥.....FLGFLG A AGSTMGARSM TLTVQARQ ♠LL SGIVQQQ *DP107-NNL*

LRAIEAQQHL LQLTVWGIKQ LQARILAVER YLKDQ-DP107 QLLG♠♥ I WGC

♠107x178x4♠
♥ALLMOTI5♥ *LVS Coiled-Coil*
SGKLICT TAVP ♥WNASWS NKSLEQIWNN MTWM *E ♠WDRELNN *DP178-*

YTSLIHSL IEESQNQQEK NEQELLELDK *WASLWNWF-DP178* NI

♦Transmembrane Region♦
TNWLWYIK♠ ♦IF IMIVGGLVGL RIVFAVLSIV NRVRQGYS♥ PL

✣P23LZIPC✣
SFQTHLPTPR GPDR ✣PEGIEE EGGERDRDRS IRLVNGSLAL IWDDLRSL✣ CL

♥ALLMOTI5♥ ♠107x178x4♠
F ♥SYHRLRDLL LIVTRIVELL GRRGW ♠EALKY WWNLLQYWSQ

ELKNSAVSLL NAT♠ AIAVAEG TDRVIEVVQG A♥ CRAIRHIPR

RIRQGLERIL L

FIG. 20

Fusion ♥ALLMOTI5♥
Peptide ♠107x178x4♠
♥.....FLGFL LGVGSAIAS GVA ♠VSKVLHL EGEVNKIKSA

♣P1&12LZIPC♣
LLSTNKAVVS LSNGVSVLTS KVLDLKNYID KQ♠♥ LL ♣PIVNKQ

♠107x178x4♠
SC ♠SISNIETV I♣ EFQQKNNRLLEITREFSVNAG♠ VTTPVSTMLTNSELLSL

♣P1&12LZIPC♣
♥ALLMOTI5♥
INDM ♣PI ♥TNDQ KKLMSNNVQI V♣ RQQSYSI♣ MS IIKEEVLAYV

VQ♥ LPLYGVID TPCWKLHTSP LCTTNTKEGS NICLTRTDRG WYCDNAGSVS

FFPQAETCKV QSNRVFCDTM NSLTLPSEIN LCNVDIFNPK

YDCKIMTSKT DVSSSVITSL GAIVSCYGKT KCTASNKNRG

IIKTFSNGCDYVSNKGMDTV SVGNTLYYVN KQEGKSLYVK G

♣P7, 12, & 23LZIPC♣
♠107x178x4♠ ♥ALLMOTI5♥
EPIINFYDPLVF ♣PSDE ♠FDASISQVNEKINQSLAF ♥I♣ RKSDELL♣

♦Transmembrane Region♦
IINVNA♠ GK STTN ♦IMITTI IIVIIVILLS LIAVGLLLY♥ C♦

KARSTPVTLS KDQLSGINNI AFSN

FIG. 21

Fusion
Peptide ♥ALLMOTI5♥ ♠107x178x4♠
.....ELGELG ♥AAGTA MGAAA ♠TALTVQSQHLLAGILQQQKNLLAAV

♠107x178x4♠
EAQ♠ QQM ♠LKLTIWGVKNLNARVTALEKYLEDQARLN♠ AWG♥ CA

LVS Coiled-Coil
♥ALLMOTI5♥ ♠107x178x4♠
WKQVCHTTVP WQWNNRTPDW ♥NNMT *WLE ♠WERQISYLEGNIT

♠107x178x4♠
TQLEEARAQEEKNLD♠ AYQKLSS* WSDFWSW♥ FDF ♠SKWLN ♦ILK

♦Transmembrane Region♦
IGELDVLGIIGLRLLYTV♦ YS♠ CIARVRQGYS PLSPQIHIHP WKGQPDNAEG

PGEGGDKRKN SSEPWQKESG TAEWKSNWCK RLTNWCSISS IWLYNS

♥ALLMOTI5♥
♥CLTL LVIHLRSAFQY IQYGLGELKA AAQEAVVALA RLAQNAGYQIWL♥

ACRSAYRA IINSPRRVRQ GLEGILN

FIG. 22

Fusion
Peptide ♥ALLMOTI5♥ ♠107x178x4♠
LVS Coiled-Coil
.....FAG ♥VVL AGVALGVATA AQITAGIALHQ ♠*SNLNAQAIQ

SLRTSLEQSNKAIEEIREATQETVIA* VQGVQDY♠ VNNEL♥ VP

♥ALLMOTI5♥
♠107x178x4♠
✣P6 & 12LZIPC✣
AMQHMSCELVGQRLGLRLLRYYTELLSIFGPSLRD ✣PISA ♠♥EISIQALIYAL

GGEIHKILEKLGYSGSD♠ MIALESRGIKTKI♥ THVDLPGKF IILSISY

✣P1 & 12LZIPC✣
✣PTLSEVKGVIVHRLEAV✣ SYNIGSQEWYTTVPRYIATNGYLISNFDESSCVFVS

ESAICSQNSL YPMSPLLQQC IRGDTSSCAR TLVSGTMGNK FILSKGNIVA

NCASILCKCY STSTIINQSP DKLLTFIASD TCPLVEIDGA TIQVGGRQYP

LVS Coiled-Coil
♥ALLMOTI5♥
✣P12 & 23LZIPC✣
DMVYEGKVAL G ✣PAISLD ♥RL*DVGTNLGNALKKLDDAKVLI✣

♦Transmembrane Region♦
DSS✣ NQILETVR RS♥* SFN ♦EGSLL SVPILSCTAL ALLLLIYCC♦

K RRYQQTLKQH TKVDPAFKPD LTGTSKSYVR SL

FIG. 23

Fusion ♥ALLMOTI5♥
Peptide
♠107x178x4♠
♥.....EIGAI IGSVALGVA TAAQITAASA LIQANQNAAN ♠ILRLKESITA

TIEAVHEVTDGLSQLAVA♠ VG KM♥ QQFVNDQFNNTAQELDCIKITQQV

♥ALLMOTI5♥
GVELNLYLTELTTV FGPQITSPAL ♥TQLTIQALYNAGGNMDYLLTKLGVG

♣P1 & 12LZIPC♣
NNQLSSLIGSGLIT GN♥ ♣PILYDSQT QLLGIQVTLP SVGNLNNMRATYLET

LSVST TKGFASALVP KVVTQVGSVI EELDTSYCIE TDLDLYCTRI VTFPMSPGIY

SCLNGNTSAC MYSKTEGALT TPYMTLKGSV IANCKMTTCR CADPPGIISQ

♥ALLMOTI5♥
♠107x178x4♠
NYGEAVSLID RHSCN ♠♥VLSLD GITLRLSGEF DATYQKNISI LDSQVIVTG

LVS Coiled-Coil
♦Trans-
N LDISTELGNV NNSISNALDK LEESNSKLDK VNVKLTSTSA ♦LIT YIA

membrane Region♦
LTAISLVCGILSLV♥♠ LACYLMY♦ KQKAQQKTLLWLGNNTLGQMRATTKM

FIG. 24

Fusion ♥ALLMOTI5♥
Peptide ♠107x178x4♠ *LVS Coiled-Coil*
.....FFGGV ♠IG ♥TIALG *VATSAQITAAVALVEAKQARSDIEKLKE

AIRDTNKAVQSVQSSIGNLIVAIKSVQ* DYVNKE♥♠ IVPSIARLGCEAAG

♥ALLMOTI5♥
♠107x178x4♠
LQLGIALTQH ♠♥YSELTNIFGDNIGSLQEKGIKLQGIASLYRTNITE♥♠

♣P5 & 12LZIPC♣
IFTTSTVDKYDIYDLLFTESIKVRVIDVDLNDYSITLQVRL ♣PLLTRLLNTQIYR

VDSISYNI♣ QNREWYI♣ PLPSHIMTKGAFLGGADVKECIEAFSSYIC

PSDPGFVLNHEMESCLSGNISQCPRTVVKSDIVPRYAFVNGGVVANCITT

TCTCNGIGNRINQPPDQGVKIITHKECNTIGINGMLFNTNKEGTLAFYTP

♥ALLMOTI5♥
♠107x178x4♠
♣P6 & 23LZIPC♣
NDITLNNSVALD ♣PIDI ♠SIELN ♥KAKSDLEESKEWI♣ RRSNQKL♣

♦Transmembrane Region♦
DSIGNWHQSSTT ♦IIIV♠ LIM IIILFIINVT II♦ IIAVKYY♥ R

IQKRNRVDQN DKPYVLTNK

FIG. 25

Fusion
Peptide
.....GLFGAI AGFIENGWEGMIDGWYGFRHQNSEGTG

♠107x178x4♠
♥ALLMOTI5♥
LVS Coiled-Coil
*Q ♥AADLKST ♠QAAIDQINGKLNRVIEKTNEKFHQIEKEFSEVEGRIQ
DLEKYVEDTKIDL* WSYNAELLVALENQHTI♠ DLT♥ DSEMNKLFEKTR

RQLRENAEEMGNGCFKIYHKCDNACIESIRNGTYDHDVYRDEALNNRFQIKG

VELKSGYKDWILWISFAISCFLLCVVLLGFIMWACQRGNIRCNICI

FIG. 26

WO 96/19495 PCT/US95/16733

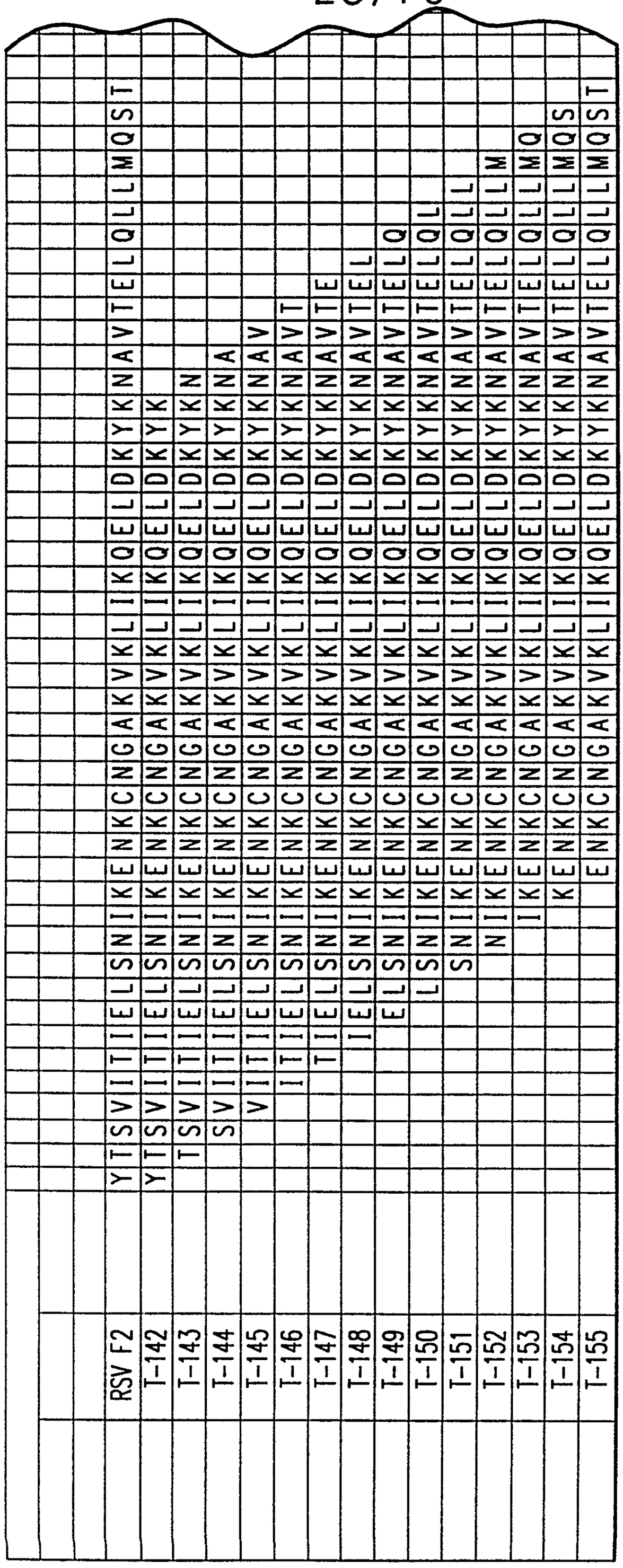


FIG.27A

	FUSION ARRAY																
	PURIFIED																
		IC50 (XTT)															
	AV	(µg/ml)	CD														
RSV F2	++	39	++														
T-142	++	31	+++														
T-143	+	114	++														
T-144	++	40	+														
T-145	-	281	-														
T-146	-	204	-														
T-147	-	354	-														
T-148	-	336	-														
T-149	-	342	+														
T-150	+/-	116	+														
T-151	+/-	117	++														
T-152	-	280	+														
T-153	+/-	118	++														
T-154	-	253	+														
T-155																	

FIG.27B

WO 96/19495

PCT/US95/16733

RSV PEPTIDE#	AVG. IC50 (XTT) UG/ML
T-22 IELSNIKENKCNGTDAKVKLIKQELDKYKNAVTELQLLMQST	>500
T-23 IELSNIKENKCNGTDAKVKLIKQELDKYKNAVTELQLLMQST	>500
T-24 ENKCNGTDAKVKLIKQELDKYKNAVTELQLLMQST	>500
T-25 DAKVKLIKQELDKYKNAVTELQLL	>500
T-26 CNGTDAKVKLIKQELDKYKNAVTELQLL	>500
T-27 SNIKENKCNGTDAKVKLIKQELDKYKNAVTELQLL	>500
T-68 VSKGYSALRTGWYTSVIIEELSNIKENKENKUNGTDAKVKLIKQELDKYKN	165
T-334 AFIRKSDELLHNV	26
T-371 YTSVIITIELSNIKENKUNGTDAKVKLIKQELDKYKNA	>500
T-372 TSVIITIELSNIKENKUNGTDAKVKLIKQELDKYKNA	NOT TESTED
T-373 SVIITIELSNIKENKUNGTDAKVKLIKQELDKYKNAVTELQLL	>500
T-374 SNIKENKUNGTDAKVKLIKQELDKYKNAVTELQLL	>500
T-375 IKENKUNGTDAKVKLIKQELDKYKNAVTELQLLMQS	>500
T-575 AVSKGYLSALRTGWYTSVIITIELSNIKENKUNGTDA	>100

FIG.27C

WO 96/19495 PCT/US95/16733

RSV DP-107-LIKE REGION (F1)	RSV
	F1-107
	T-120
	T-121
	T-122
	T-123
	T-124
	T-125
	T-126
	T-127
	T-128
	T-129
	T-130
	T-131
	T-132
	T-133
	T-134
	T-135
	T-136
	T-137
	T-138
	T-139
	T-140
	T-141

FIG.27D

FUSION ASSAY																								
	PURIFIED			204	354	347	126	95	84	89	89	206	343	177	118	272	307	187	60	194	99	38	86	160	204
		IC50 XTT																						
			(µg/ml)																					
			CD	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+/-	+/-	+/-
		RSV	AV																					
		F-107		-	-	-	+/-	+	+	+	+	-	-	-	+/-	-	+/-	+/-	+	-	+	+	++	+	-	-
		T-120																							
		T-121																							
		T-122																							
		T-123																							
		T-124																							
		T-125																							
		T-126																							
		T-127																							
		T-128																							
		T-129																							
		T-130																							
		T-131																							
		T-132																							
		T-133																							
		T-134																							
		T-135																							
		T-136																							
		T-137																							
		T-138																							
		T-139																							
		T-140																							
		T-141																							

FIG.27E

| RSV | | |

WO 96/19495 PCT/US95/16733

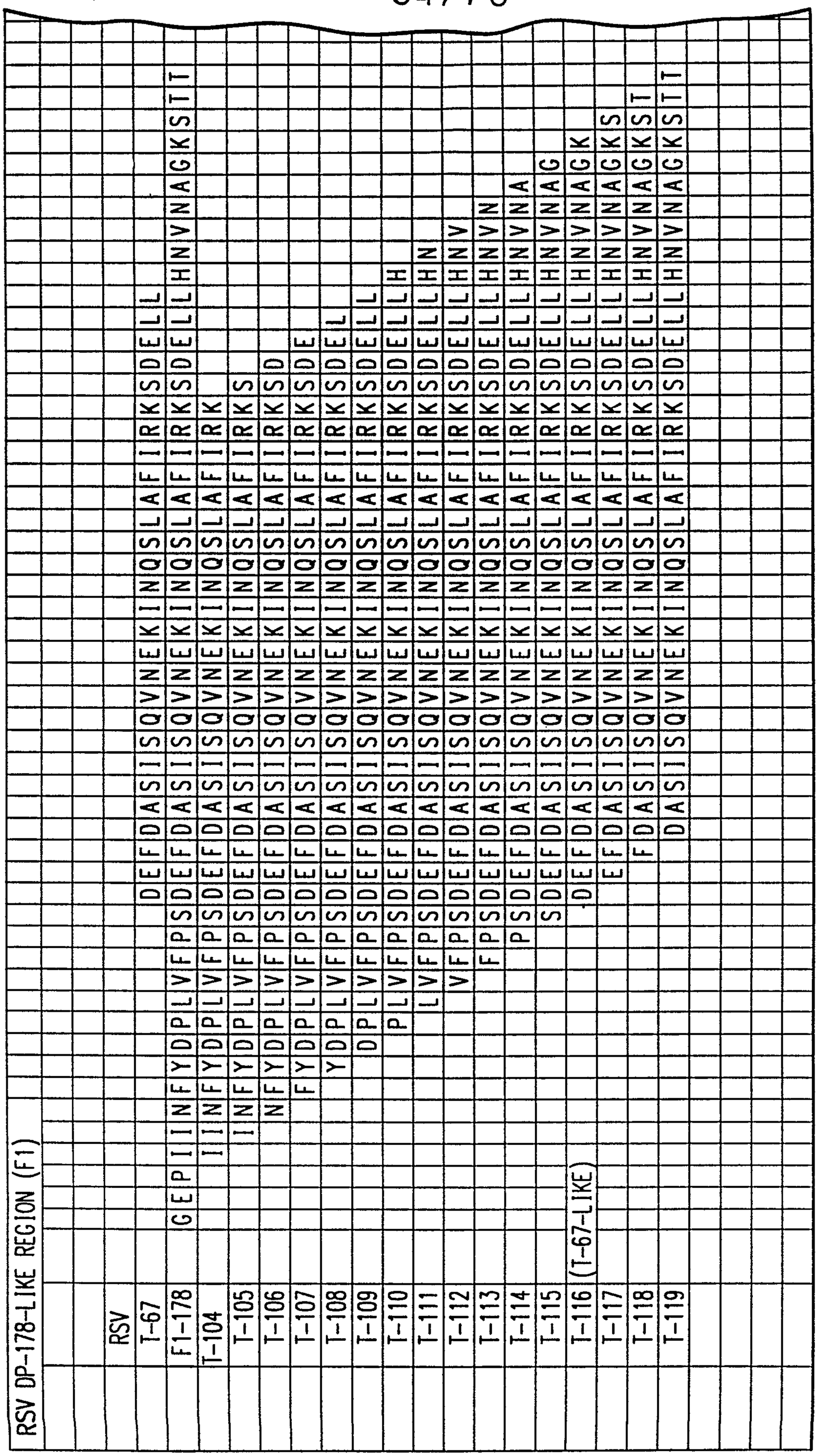


FIG. 28A

	FUSION ASSAY																									
	PURIFIED			CD +/−											+/−	+/−	+/−	+/−	+/−	+/−	+/−	+/−				
	IC50																									
RSV	AV	(μg/ml) (XTT)																								
T-67	++	37																								
F1-178																										
T-104	+	95																								
T-105	+	86																								
T-106	-	186																								
T-107	++	20																								
T-108	+++	6																								
T-109	+++	8																								
T-110	++	30																								
T-111	+++	9																								
T-112	+++	8																								
T-113	+++	6																								
T-114	+++	5																								
T-115	+++	6																								
T-116	+++	9																								
T-117	+++	14																								
T-118	+++	5																								
T-119	+++	6																								

FIG.28B

FIG.28C

| RSV | | |

FIG.29A

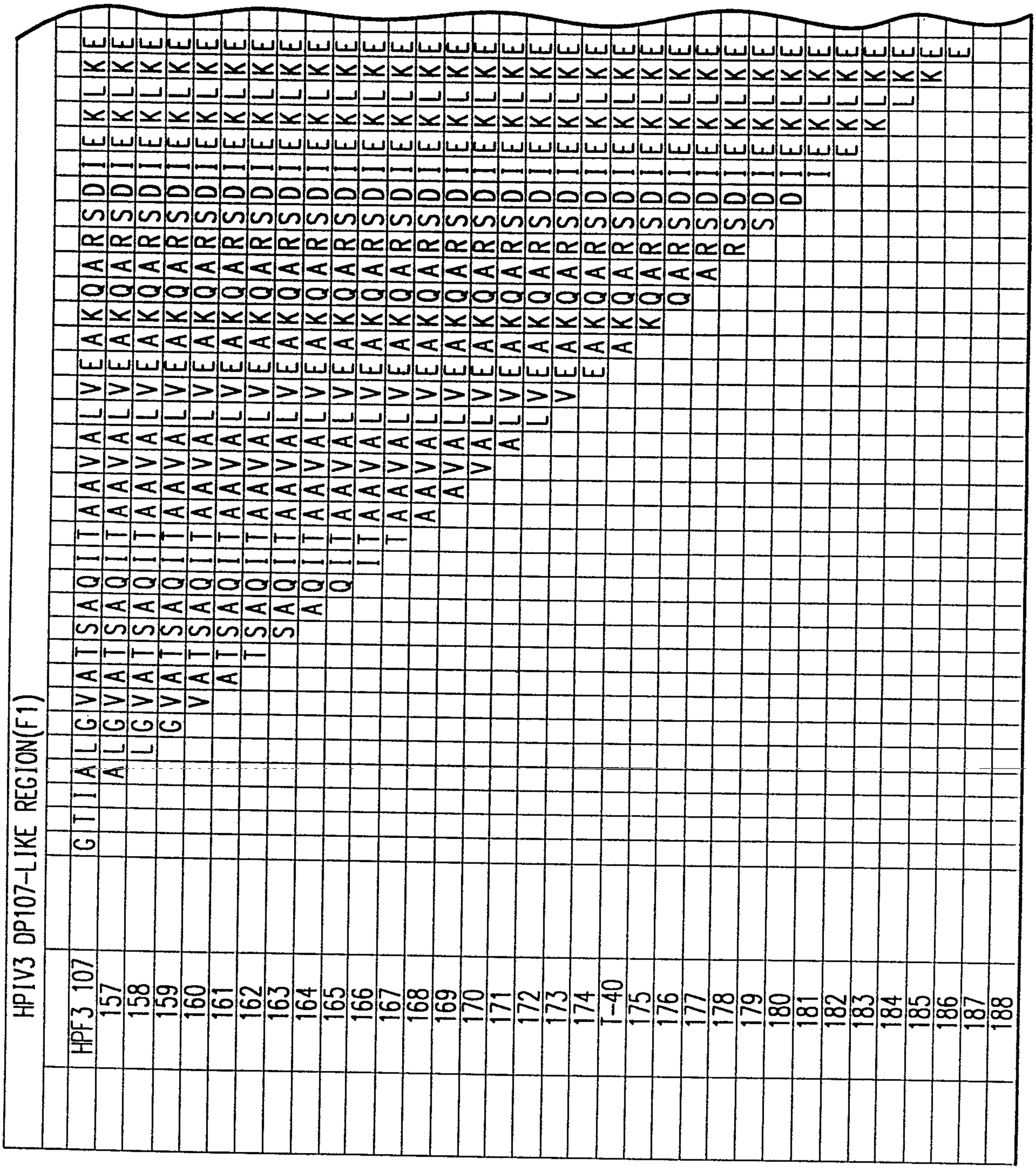
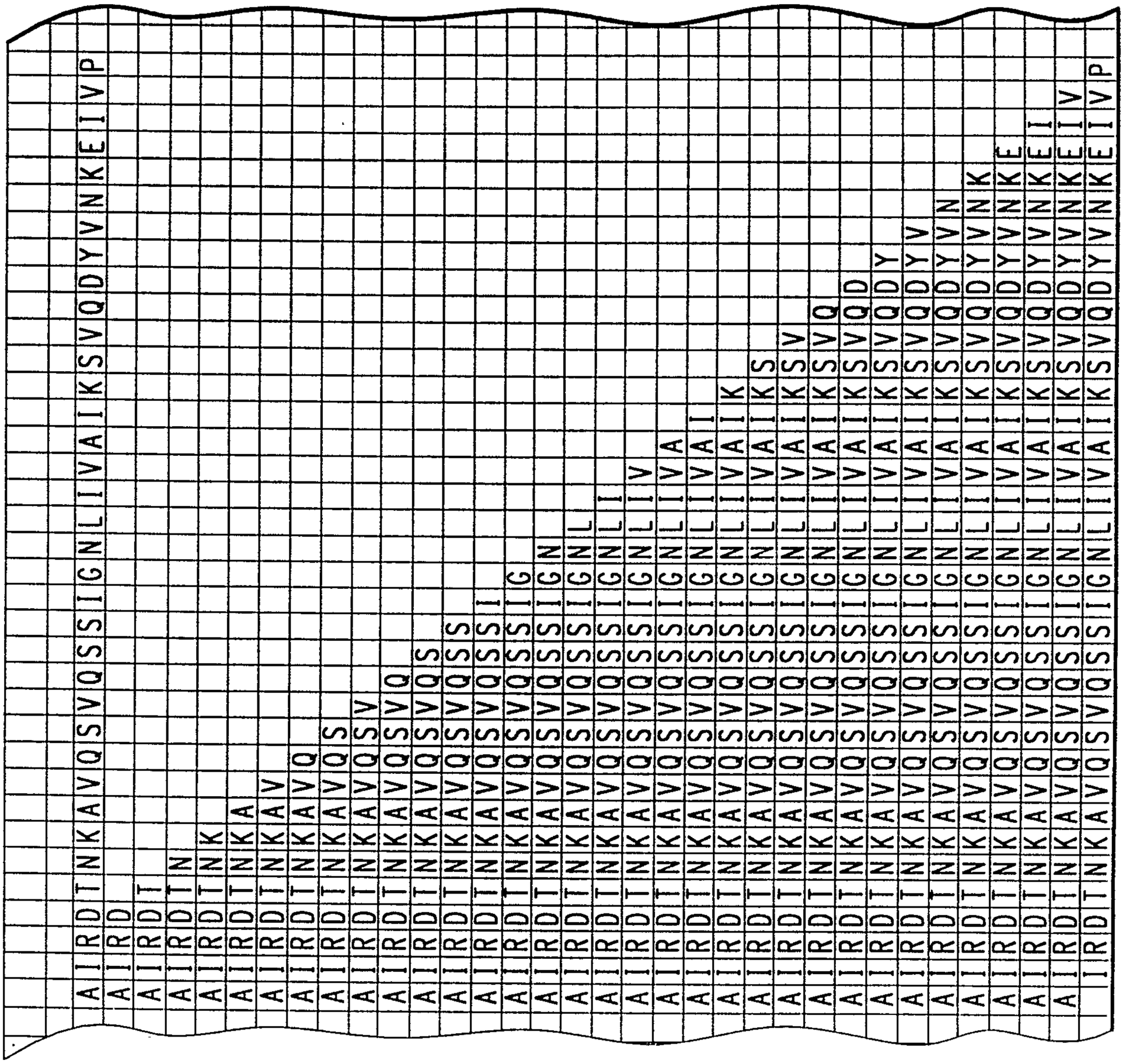

HPIV3 DP107-LIKE REGION(F1)
HPF3 107
157
158
159
160
161
162
163
164
165
166
167
168
169
170
171
172
173
174
T-40
175
176
177
178
179
180
181
182
183
184
185
186
187
188

FIG.29B



| | | |

WO 96/19495

PCT/US95/16733

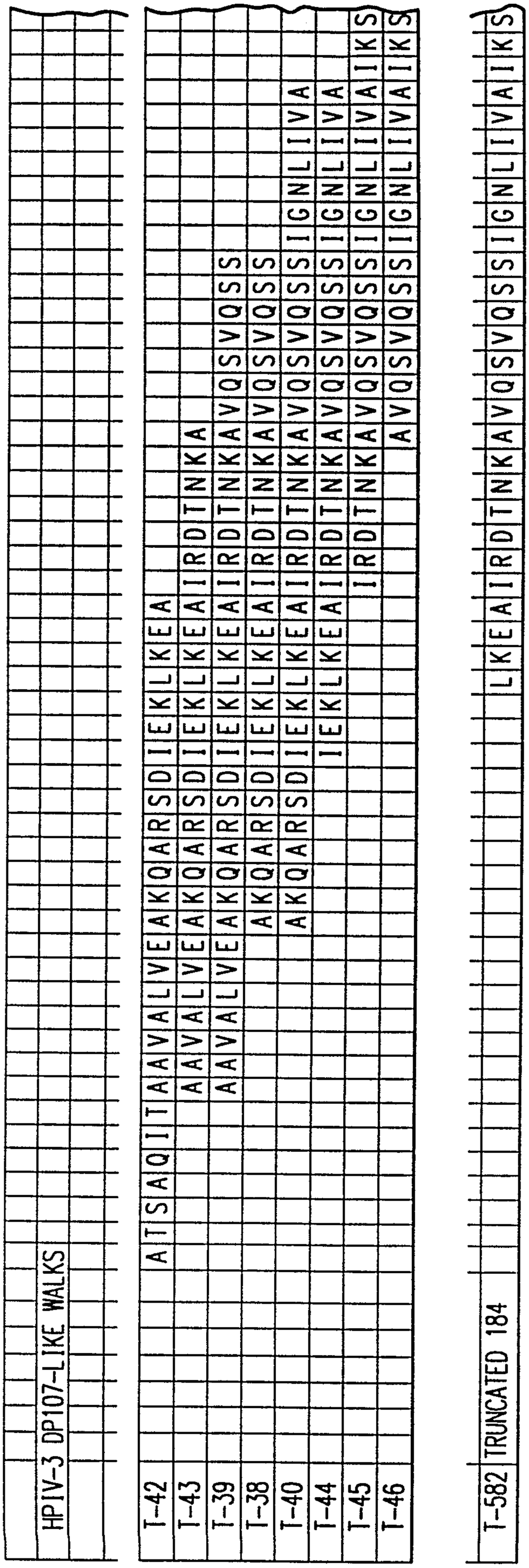


FIG.29D

WO 96/19495 PCT/US95/16733

	AVG. IC50		
			643.000 µg/ml*
			655.000 µg/ml*
			720.000 µg/ml*
			700.000 µg/ml*
			58.798 µg/ml
			225.140 µg/ml
			485.000 µg/ml*
			548.000 µg/ml*
			137.654 µg/ml*

VQDY
VQDYVNKEIV

FIG.29E

WO 96/19495

PCT/US95/16733

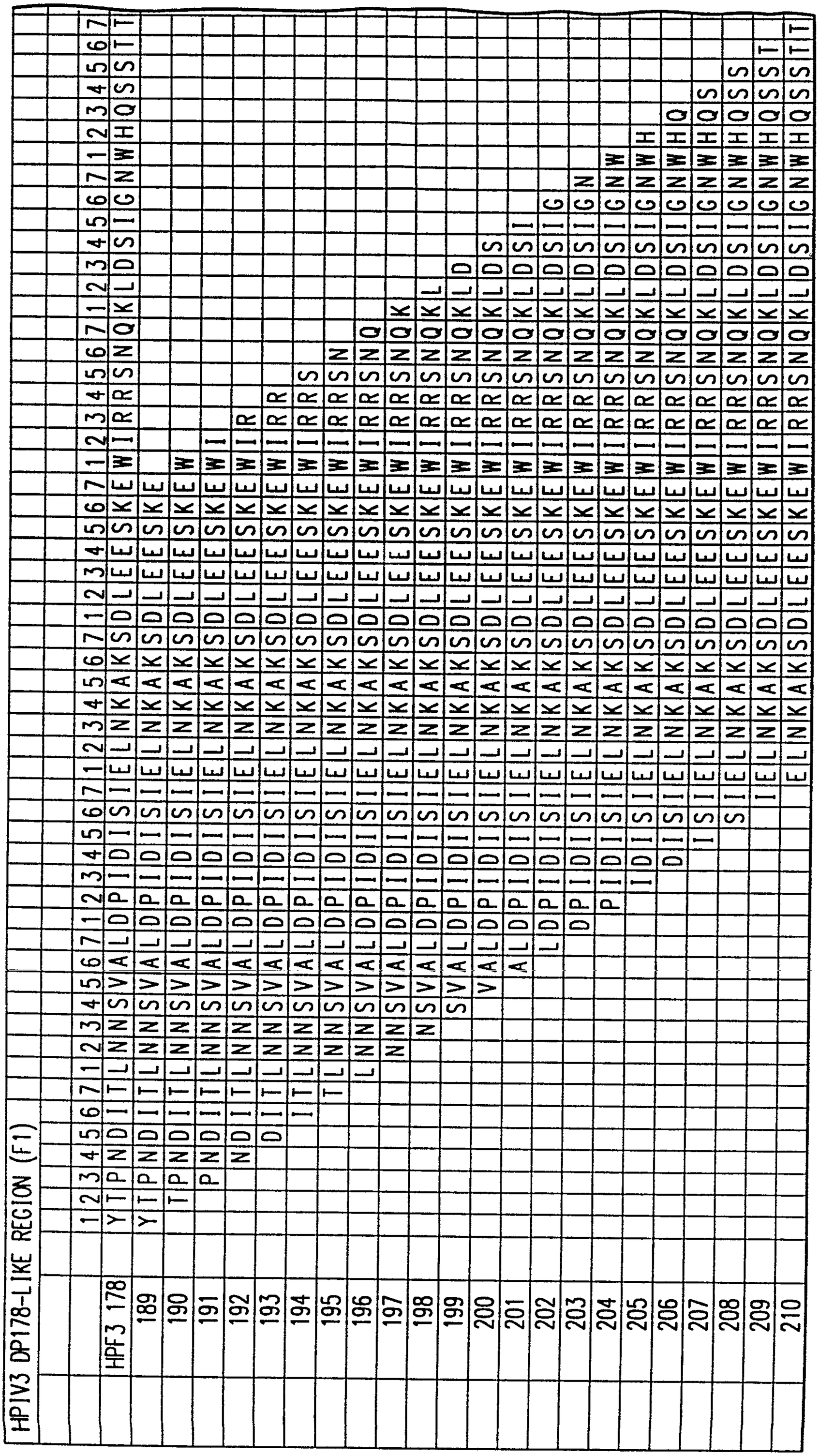


FIG.30A

			CD	-	-	-	-	-	-	±	+	±	+	+	±	+	+	+	+	+	+	+	+	+	+	+	+
		IC50																									
		HP1V3 178	AV	(μg/ml)																							
		189	-	827*																							
		190	-	775*																							
		191	-	612*																							
		192	-	699*																							
		193	-	525*																							
		194	+	61*																							
		195	+	49																							
		196	++	3.1																							
		197	++	71.15																							
		198	+++	0.325																							
		199	++	2.3*																							
		200	++	1.0*																							
		201	+++	0.224*																							
		202	+++																								
		203	+++	0.390*																							
		204	+++	0.213																							
		205	+++	0.174																							
		206	++	2.0*																							
		207	++	2.0*																							
		208	+	37*																							
		209	++	1.0*																							
		210	++	2.0*																							

FIG.30B

CA 02208420 1997-06-20

PCT/US95/16733

T-269	TRUNCATED 201 EWIRRSNQKLDSI	457.500UG/ML
T-626	205 MUTANT IDISIELNKAKSDLEESKEWIKKSNQKLDSIGNWH	209.589NG/ML
T-383	RMKQLEDKVEELLSKLEWIRRSNQKLDSI	NOT DONE
T-577	DQQIKQYKRLLDRLIIPLYDGLRQKDVIVSNQESN	133.793UG/ML*
T-578	YSELTNIFGDNIGSLQEKGIKLQGIASLYRTNITEI	107.177UG/ML*
T-579	TSITLQVRLPLLTRLLNTQIYRVDSISYNIQNREWY	NOT DONE

FIG.30C

RECTIFIED SHEET (RULE 91)

FUSION ♥ALLMOTI5♥
PEPTIDE ♠107x178x4♠
.....RNKRGVFVLGFLGFLATAGSAMGAAS ♠♥ XXXXAQSRTLLAGIVQQQQQ
LLDVVKRQQELLRLTVWGTKNLQTRVTAIEKYLKDQAQL♠NAWG♥ CAF

♥ALLMOTI5♥
*LVS PREDICTED COILED-COIL
RQVCHTTVPWPNASLTPDW *NND ♥TWQEWERKVDFLEENITALLEEAQIQQ

♠107x178x4♠
EKNMY ♠ELQKLNSWD* VF♥ GNXXXXXXXXXXXXXXXXXXXXXXXXXXX♠

IYIVMLAKLRQGYRPVFSSPPSYFQXTHTQQDPALPTREGKEGDGGEGGGNSSWP

WQIEYIHF

FIG. 31

MTRRRVLSVVVLLAALACRLGAQTPEQPAPPATTVQPTATRQQTSFPFRVCELSSHGDLFRFSSD

♠ 107x178x4♠

IQCPSFGTRENHTEGLLMVFKDNIIPYSF ♠ KVRSYTKIVTNILIYNGWYADSVTNRHE♠

EKFSVDSY ETDQMDTIYQ CYNAVKMTKD GLTRVYVDRD GVNITVNLKP TGGLANGVRR

YASQTELYDA PGWLIWTYRT RTTVNCLITD MMAKSNSPFD FFVTTTGQTV EMSPFYDGKN

KETFHERADS FHVRTNYKIV DYDNRGTNPQ GERRAFLDKG TYTLSWKLEN RTAYCPLQHW

QTFDSTIATE TGKSIHFVTD EGTSSFVTNT TVGIELPDAF KCIEEQVNKT HEKYEAVQD

RYTKGQEAIT YFITSGGLLL AWLPLTPRSL ATVKNLTELT TPTSSPPSSP SPPAPSAARG

STPAAVLRRR RRDAGNATTP VPPTAPGKSL GTLNNPATVQ IQFAYDSLRR QINRMLGDLA

RAWCLEQKRQ NMVLRELTKI NPTTVMSSIY GKAVAAKRLG DVISVSQCVP VNQATVTLRK

SMRVPGSETM CYSRPLVSFS FINDTKTYEG QLGTDNEIFL TKKMTEVCQA TSQYYFQSGN

♠107x178x4♠

EIHVYNDYHH FKTIELDGIA TLQTFISLNT ♠SLIENIDFASLELYSRDEQRASNVFD *LE♠

LVS PREDICTED COILED COIL TM Potential

GIFREYNFQAQNIAGLRKDLDNAVSN* GRNQ FVDGLGELMDSLGSVG QSITN

♣P12LZIPC♣

TM Potential TM Potential

LVSTVGGLFSSLVSGFISF FK N ♣PFGGMLILVLVAGVVILVISL♣ TRRTRQMS

QQPVQMLYPG IDELAQQHAS GEGPGINPIS KTELQAIMLA LHEQNQEQKR AAQRAAGPSV

ASRALQAARDRFPGLRRRRY HDPETAAALL GEAETEF

FIG. 32

```
MMDPNSTSED VKFTPDPYQV PFVQAFDQAT RVYQDLGGPS QAPLPCVLWP VLPEPLPQGQ

LTAYHVSTAP TGSWFSAPQP APENAYQAYA APQLFPVSDI TQNQQTNQAG GEAPQPGDNS

TVQTAAAVVF ACPGANQGQQ LADIGVPQPA PVAAPARRTR KPQQPESLEE CDSELEI

@DNA BINDING@          ♠107x178X4♠     +DIMERIZATION+
@KRY KNRVASRKCRAK      ♠FK@ Q           +LLQHYREVAAAKSSENDRLRLLLKQ♠

MCPSLDVD+ SI IPRTPDVLHE DLLNF
```

FIG. 33

FUSION
PEPTIDE ♥ALLMOTI5♥ *LVS COILED-COIL*
FAG ♥VVLAGAALGVATAAQITAGIALHQSML*NSQAIDNLRASLETTN

QAIEAIRQAGQEMI*LAVQGVQDYINN♥ ELIPSMNQLSCDLIGQKLGLKLLRYYT

♣P23LZIPC♣
♣P6,12LZIPC♣
♠107X178X4♠
♥ALLMOTI5♥
EILSLFGPSLRD ♣PISA ♠♥EISIQLSYALGGDINKV♣ LEKLGYSGGDL♣

♣P1,12LZIPC♣
LGILES♠ RGIKARI♥ THVDTESYFIVLSIAY ♣PTLSEIKGVIVHRLEGV♣ SY

NIGSQEWYTTVPKYVATQGYLISNFDESSCTFMPEGTVCSQNALYPMSPLLQECL

RGSTKSCARTLVSGSFGNRFILSQGNLIANCASILCKCYTTGTIINQDPDKILTYIAA

♣P23LZIPC♣
♣P12LZIPC♣
♥ALLMOTI5♥
LVS COILED-COIL
DHCPVVEVNGVTIQVGSRRYPDAVYLHRIDLGP ♣P ♥IS *LERLDVGTNLGN

♦TRANSMEMBRANE REGION♦
AIAKLEDAKELL♣ ESSDQI*L♣ RSMK ♦GLSSTSIVYILI♥ AVCLGGLIGIP

ALICCC♦ RGRCNKKGEQVGMSRPGLKPDLTGTSKSYVRSL

FIG. 34

WO 96/19495 PCT/US95/16733

Pre S1 and Pre S2
MGQNLSTSNPLGFFPDHQLDPAFRANTANPDWDFNPNKDTWPDANKVGAGAFG

LGFTPPHGGLLGWSPQAQGILQTLPANPPPASTNRQSGRQPTPLSPPLRNTHPQAM

QWNSTTFHQTLQDPRVRGLYFPAGGSSSGTVNPVLTTASPLSSIFSRIGDPALN

MAJOR SURFACE ANTIGEN(HBs)
FUSION
PEPTIDE
♣P12 & 23LZIPC♣
MENITSG **FLG ♣PLL** VLQAGFFLLTRILTI♣ PQSLDSWWTSLNFLGGTTVCLG

♣P12 & 23LZIPC♣
QNSQSPTSNHSPTSCPPTC ♣PGYRWMCLRRFIIFLFILLLCLIFLLVLLDYQGML♣

PVCPLIPGSSTTSTGPCRTCMTTAQGTSMYPSCCCTKPSDGNCTCIPIPSSWAFGKF

♦TRANSMEMBRANE REGION♦
LWEWASARFSWLS ♦**LLVPFVQWFVGLSPTVWLSVI**♦ WMMWYWGPSL

♦TRANSMEMBRANE REGION♦

♦**YSILSPFLPLLPIFFCLWVYI**♦

FIG. 35

FUSION ♥ ALLMOTI5 ♥ ♠107x 178x4♠
PEPTIDE *LVS COILED COIL
AIQLIPLFVG LGI ♥TTAVSTGAAGLGVS ♠IT *QYTKLSHQLISDV

QAISSTIQDLQDQVDSLAEVVLQ* NRRGLDLLTAE♠ QGGI♥

CLALQEKCCFYANKSGIVRDKIKNLQDDLERRRRQLIDNPFWTSFHG

FLPYVMPLLGPLLCLLLVLSFGPIIFNKLMTFIKHQIESIQAKPIQVHYH

TRANSMEMBRANE REGION
RLEQEDSGGSYLTLT.....??????????????????????....

FIG. 36

MKAQKGFTLI ELMIVVAIIG ILAAIAPGQ

♠107x178x4♠
♥ALLMOTI5♥
♠♥YQDYTARTQVTRAVSEVSALKTAAESAILEGKEIVSSA♠ T♥

PK DTQYDIGFT

♠107x178x4♠
♥ALLMOTI5♥
♠♥ESTLLDGSGKSQIQVTDNQDGTVELVATLGKSSGS♠ AIKGAVITSR♥

KNDGV WNCKITKTPT AWKPNYAPAN CPKS

FIG. 37

WO 96/19495 PCT/US95/16733

MNTLQKGFTL IELMIVIAIV GILAAVALPA YQDYTARAQV

SEAILLAEGQ KSAVTEYYLN HGIWP

♠**107x178x4**♠
♥**ALLMQTI5**♥
♠♥**KDNTSAGVASSSSIKGKYVKEVKVENGVVTAT**♠

MNSSNVNKEIQGKKLSLWAKRQDGSVKW♥

FCGQP VTRNAKDDTV TADATGNDGK IDTKHLPSTC RDNFDAS

FIG. 38

MKKTLLGSLI LLAFAGNVQA DINTETSGKV TFFGKVVENT

CKVKTEHKNL SVVLNDVGKN SLSTKVNTAM PTPFTITLQN

CDPTTANGTA NKANKVGLYF Y

♠107x178x4♠
♥ALLMOTI5♥
♠♥SWKNVDKENNFTLKNEQTTADYATNVNI♠

QLMESNGTKAISVVGKETE♥

DF MHTNNNGVAL NQTHPNNAHI SGSTQLTTGT NELPLHFIAQ

YYATNKATAG KVQSSVDFQI AYE

FIG. 39

MNKKLLMNFF IVSPLLLATT ATDFTPVP

♠107x178x4♠
♥ALLMOTI5♥
♠♥LSSNQIIKTAKASTNDNIKDLLDWYSSGSDTFTNS♠♥

EVLDNSL GSMRIKNTDG SISLIIFPSP YYSPAFTKGE KV

♠107x178x4♠
♠DLNTKRTKKSQHTSEGTYIHFQISGVT♠

N TEKLPTPIEL PLKVKVHGKD SPLKYG

♣P12LZIPC♣
♣PKFDKKQLAISTLDFEIRHQLTQI♣

HGLYRSSDKT GGYWKITMND GSTYQSDLSK KFEYNTEKPP

INIDEIKTIE AEIN

FIG. 40

♥ALLMOTI5♥
MKKTAFILLL FIALTLTTSP L ♥VNG

♠107x178x4♠
LVS PREDICTED COILED-COIL
S ♠EKSEEINEKDLRKKSELQRNALSNLRQIY YYNEKAITENKESDD♠

QFLENTLL♥ FKG FFTGHPW

♠107x178x4♠
♠YNDLLVDLGSKDATNKYKGKKVDLYGAY♠

YGYQCAGGTPNKTACMYGGVTLHDN NRLTEEKKVP INLWIDGKQTTV

♣P12LZIPC♣
♣PIDKVKTSKKEVTVQELDL♣ QARHYLHGK FGLYNSDSFGGKVQ

♣P12LZIPC♣
RGLIVF HSSEGSTVSY DLFDAQGQY ♣P DTLLRIYRDN KTINSENLHI♣

DLYLYTT

FIG. 41

♥ALLMOTI5♥
MKKTAFTLLL FIALTLTTSP L ♥VNGS

♠107x178x4♠
♠EKSEEINEKDLRKKSELQGTALGNLKQIYYYNEKAKTENKESHD♠ Q♥

FLQHTILFKG FFTDHSWYND LLVDFDSKDI VDKYKGKKVDLYGAYY

GYQC AGGTPNKTAC MYGGVTLHDN NRLTEEKKVPINL WLDGKQNTV

♠107x178x4♠
♥ALLMOTI5♥
♣P12LZIPC♣
♣P ♥L ♠ETVKTNKKNVTVQELDLQARRYL♣ QEKYNLYN♠

SDVFDGKVQR♥ GLIVF HTSTE

♣P23LZIPC♣
♣PSVNYDLFGAQGQYSNTLLRIYRDNKTINSENMHI♣ DIYLYTS

FIG. 42

MKNITFIFFILLASPLYANGDRLYRADSRPPDEIKRFRSLMPRGNEYFDRGT

♥ALLMOTI5♥
♥QMNINLYDHARGTQTGFVRYDDGYV

♠107x178x4♠
♠STSLSLRSAHLAGQYILSGYSLTIYIVI♠ ANMFNVNDVISVY♥

SP HPYEQEVSAL GGIPYSQIYG WYRVNFGVID ERLHRNREYR

DRYYRNLNIA PAEDGYRLAG FPPDHQAWRE EPWIHHAPQG

CGDSSRTITG DTCNE

♥ALLMOTI5♥
♥ETQNLSTIYLREYQSKVKRQIFSDYQSEVDIYNRIRDEL♥

FIG. 43

MMFSGFNADY EASSSRCSSA SPAGDSLSYY HSPADSFSSM

GSPVNAQDFC TDLAVSSANF IPTVTAISTS PDLQWLVQPA

LVSSVAPSQT RAPHPFGVPA PSAGAYSRAG VVKTMTGGRA

LVS PREDICTED COILED-COIL
QSIGRRGKVE QLSPEEEEKR RIRRE *RNKMA AAK

♠107x178x4♠
♥ALLMOTI5♥
♥CRNRRREL ♠TDTLQAETDQLEDEKSALQTEIANLLKEKEKL♥

EFILAAHR* PACKIPDDL GFPEEMSVAS LDLTGGLPEV

ATPESEEAFT LPLLNDPEPK PSVEPVKSIS SMELKTEPFD

DFLFPASSRP SGSETARSVP DMDLSGSFYA LPLLNDPEPK

PSVEPVKSIS SMELKTEPFD DFLFPASSRP SGSETARSVP

DMDLSGSFYA GSSSNEPSSD SLSSPTLLAL

FIG. 44

SGWESYYKTEGDEEAEEEQEENLEASGDYKYSGRDSLIFLVDASKA

MFESQSEDELTPFDMSIQCIQSVYISKIISSDRDLLAVVFYGTEKDKNS

VNFKNIYVLQELDNPGAKRILELDQFKGQQGQKRFQDMMGHGSDY

SLSEVLWVCANLFSDVQFKMSHKRIMLFTNEDNPHGNDSAKASRAR

TKAGDLRDTGIFLDLMHLKKPGGFDISLFYRDIISIAEDED

♠107x178x4♠

♥ALLMOTI5♥

LVS PREDICTED COILED-COIL

♥LRVH *FEE ♠SSKLEDLLRKVRAKETRKRALSRLKLKLNKDIV* ISV

GIYNLVQKAL♥ KPPPIKLYRETN♠ EPVKTKTRTFNTSTGGLLLPSDTKR

SQIYGSRQIILEKEETEELKRFDDPGLMLMGFKPLVLLKKHHLRPSLFVYPE
ESLVIGS STLFSALLIKCLEKEVAALCRYTPRRNIPPYFVALVPQEEELDDQK
IQVTPPGFQLVFLPFADDKRKMPFTEKIMATPEQVGKMKAIVEKLRFTYRS
DSFENPVLQQHFRNLEALALDLME

♣PI2LZIPC♣

♣PEQAVDLTLPKVEAMNKRL♣ GSLVDEFKELVYPPDYNPEGKVTKR
KHDNEGSGSKRPKVEYSEEELKTHISKGTLGKFTVPMLKEACRAYGLKSG
LKKQELLEALTKHFQD

FIG. 45

GGGALSPQHSAVTQGSIIKNKEGMDAKS

♠107x178x4♠
♥ALLMOTI5♥
♥♠LTAWSRTLVTFKDVFVDFTREEWKLLDT♠ AQQIVYRNV
MLENYKNLVSLGYQLT♥ KPDVILRLEKGEEPWLVEREIHQETHPD
SETAFEIKSSVSSRSIFKDKQSCDIKMEGMARNDLWYLSLEEVWKCR
DQLDKYQENPERHLRHQLIHTGEKPYECKECGKSFSRSSHLIGHQKT
HTGEEPYECKECGKSFSWFSHLVTHQRTHTGDKLYTCNQCGKSFVH
SSRLIRHQRTHTGHKPYECPECGKSFRQSTHLILHQRTHVRVRPYECN
ECGKSYSQRSHLVVHHRIHTGLKPFECKDCGKCFSRSSHLYSHQRTH
TGEKPYECHDCGKSFSQSSALIVHQRIHTGEKPYECCQCGKAFIRKN
DLIKHQRIHVGAETYKCNQCGIIFSQNS

♣P23LZIPC♣
♣PFIVHQIAHTGEQFLTCGNQCGTALVNTSNLIGQTNHI♣ RENAY

FIG. 46

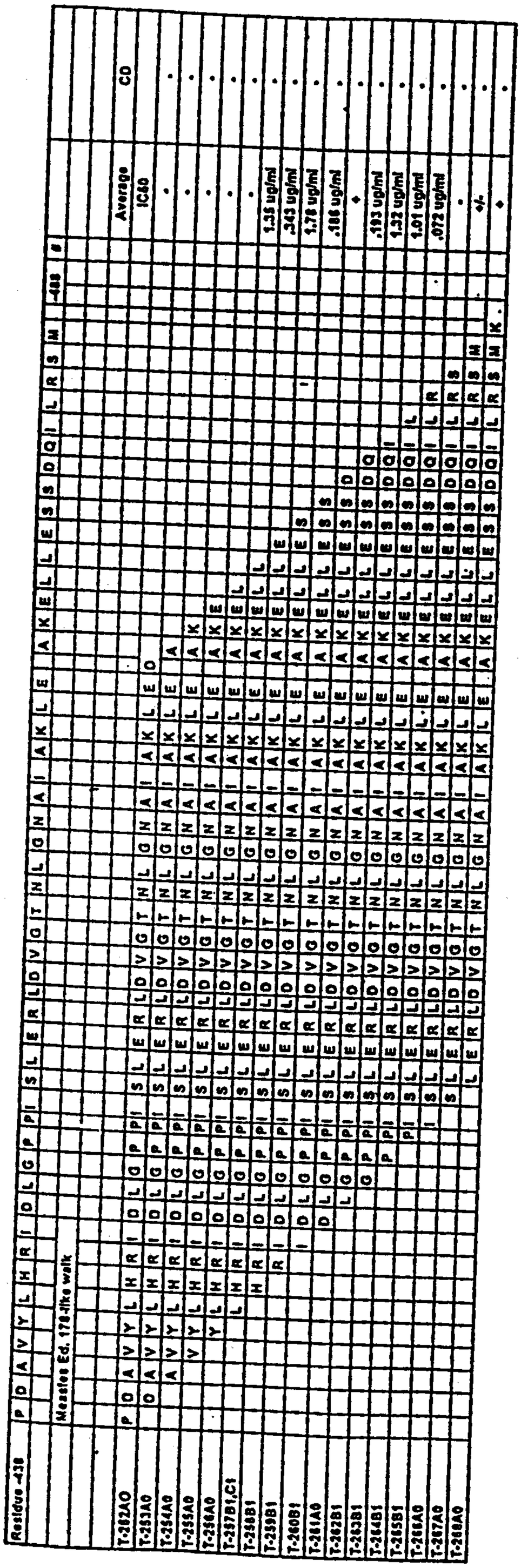

Residue -438
Average IC50
CO
Measles Ed. 178-like walk
1.35 ug/ml
.343 ug/ml
1.78 ug/ml
.186 ug/ml
.193 ug/ml
1.32 ug/ml
1.01 ug/ml
.072 ug/ml
+/-
T-252A0
T-253A0
T-254A0
T-255A0
T-256A0
T-257B1,C1
T-258B1
T-259B1
T-260B1
T-261A0
T-262B1
T-263B1
T-264B1
T-265B1
T-266A0
T-267A0
T-268A0

FIG. 47

| | | |

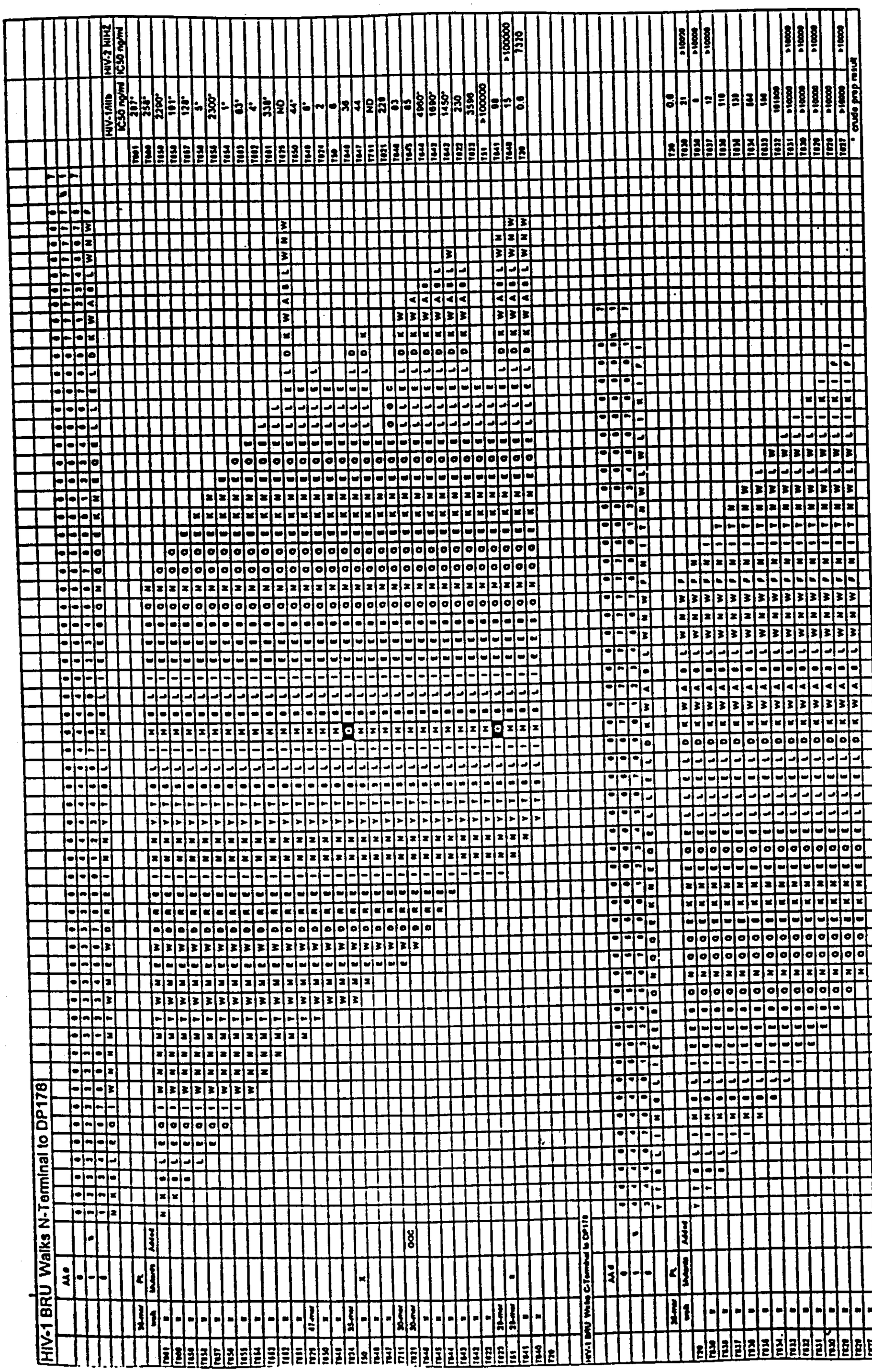

HIV-1 BRU Walks N-Terminal to DP178
HIV-1 BRU Walks C-Terminal to DP178
HIV-1IIIb IC50 ng/ml
HIV-2 NIH-Z IC50 ng/ml
* crude prep result

FIG. 49A

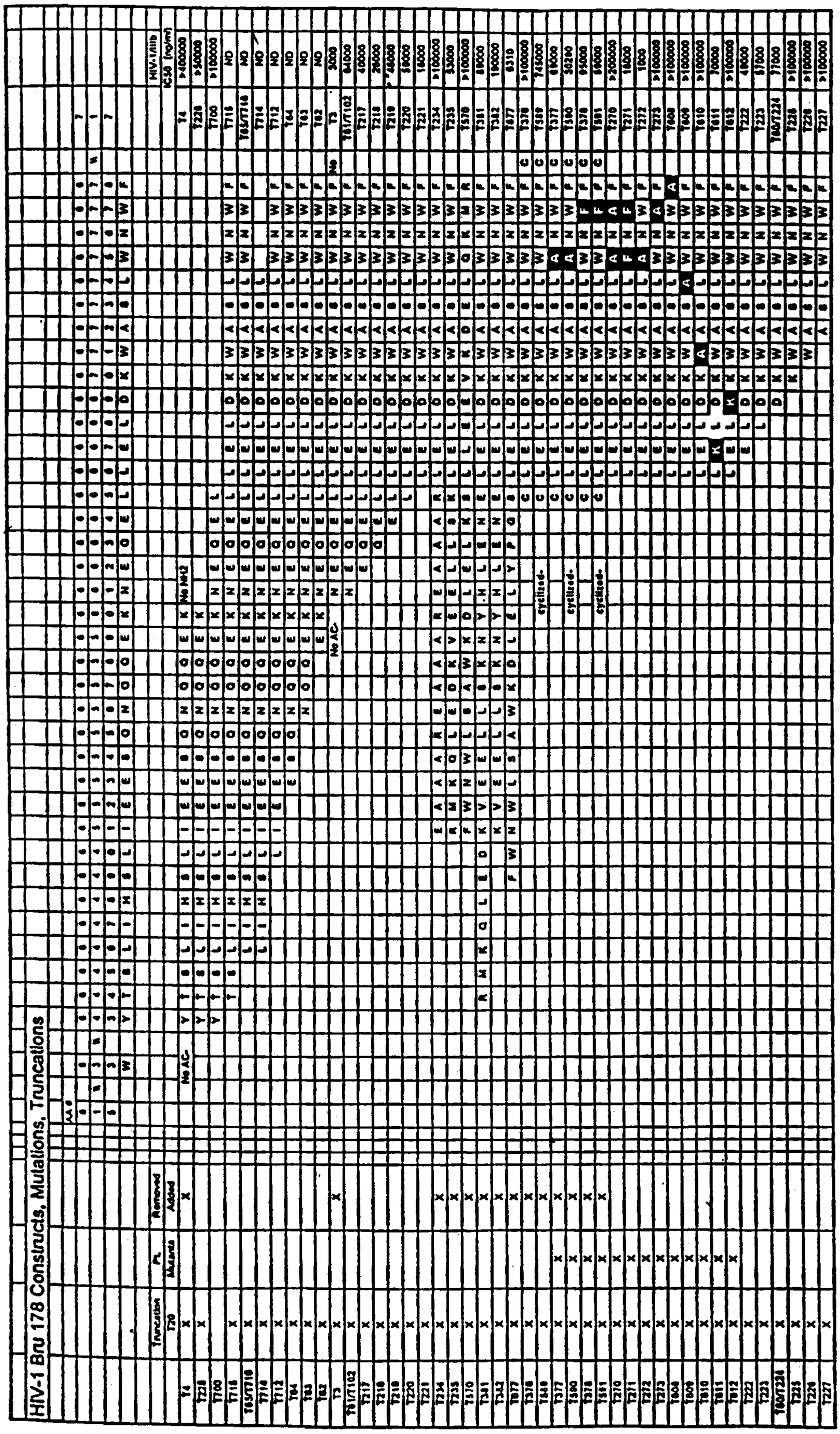


FIG. 49B

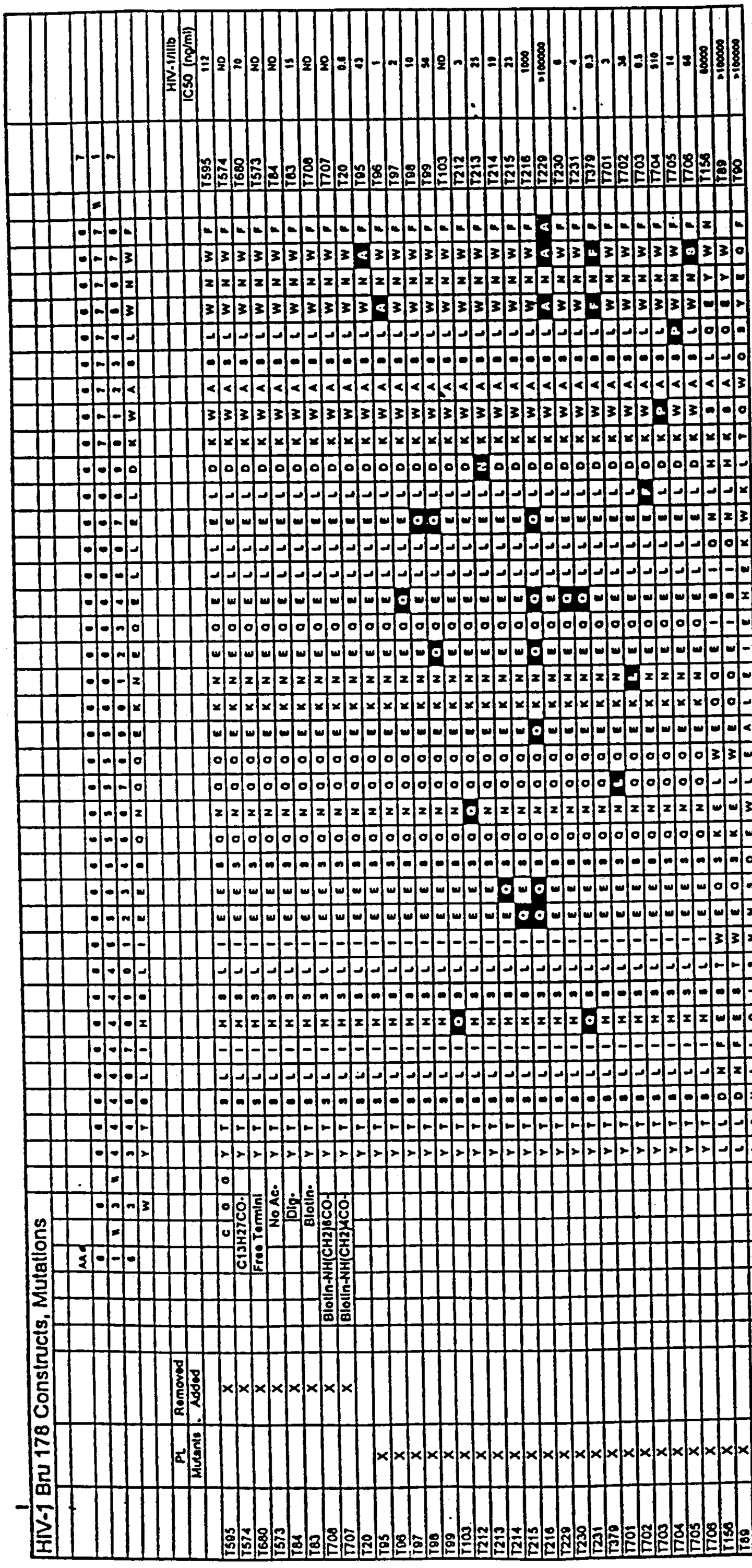


FIG. 49C

HIV-1 Bru 178 Constructs, Mutations

	PL Removed	Mutants Added	AA
T595	X		C O O
T574	X		C13H27CO-
T680	X		Free Termini
T573	X		No Ac-
T84	X		Dig-
T83	X		Biotin-
T706	X		Biotin-NH(CH2)6CO-
T707	X		Biotin-NH(CH2)4CO-
T20			
T95	X		
T96	X		
T97	X		
T98	X		
T99	X		
T103	X		
T212	X		
T213	X		
T214	X		
T215	X		
T216	X		
T229	X		
T230	X		
T231	X		
T379	X		
T701	X		
T702	X		
T703	X		
T704	X		
T705	X		
T706	X		
T156	X		
T89	X		
T80	X		

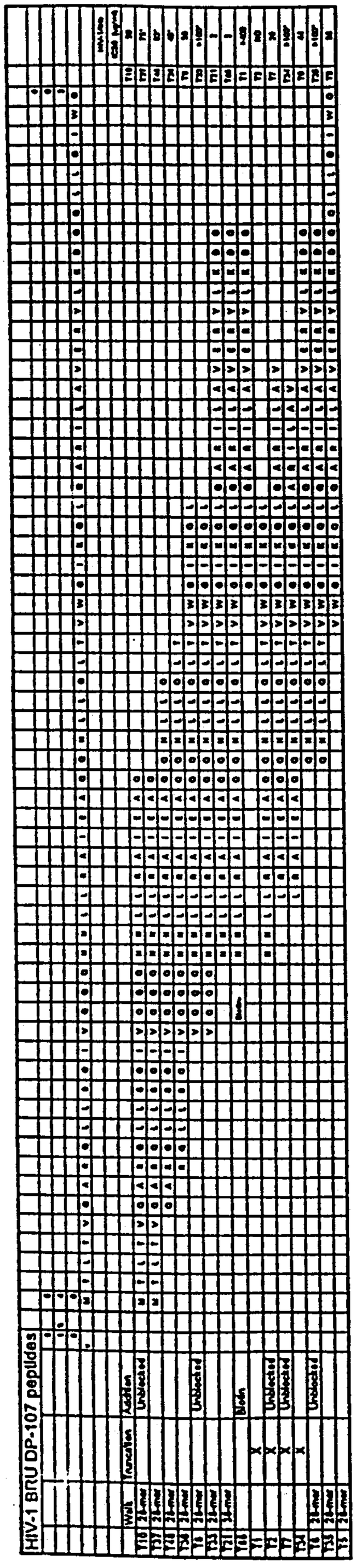


FIG. 50

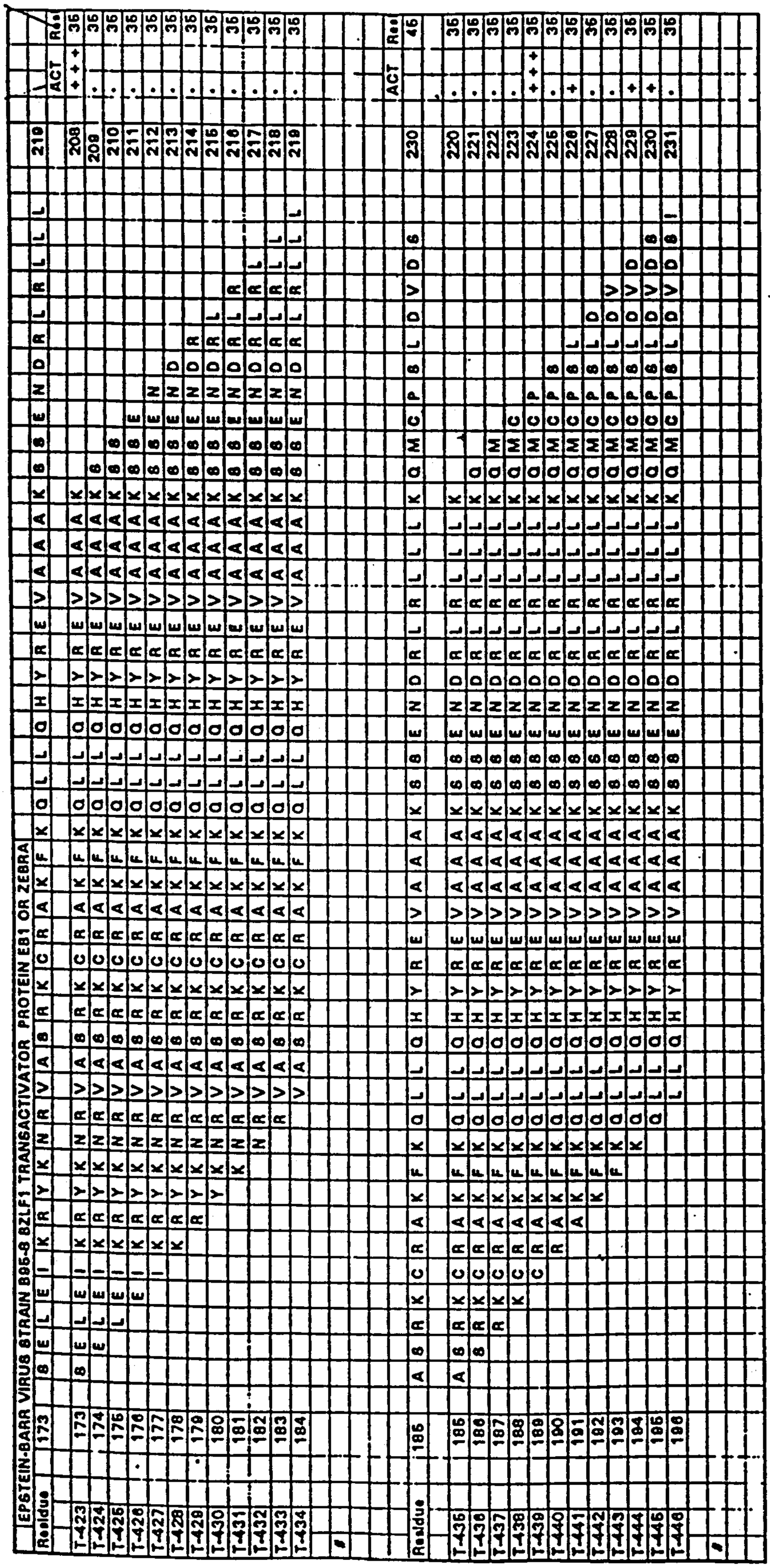


FIG. 51A

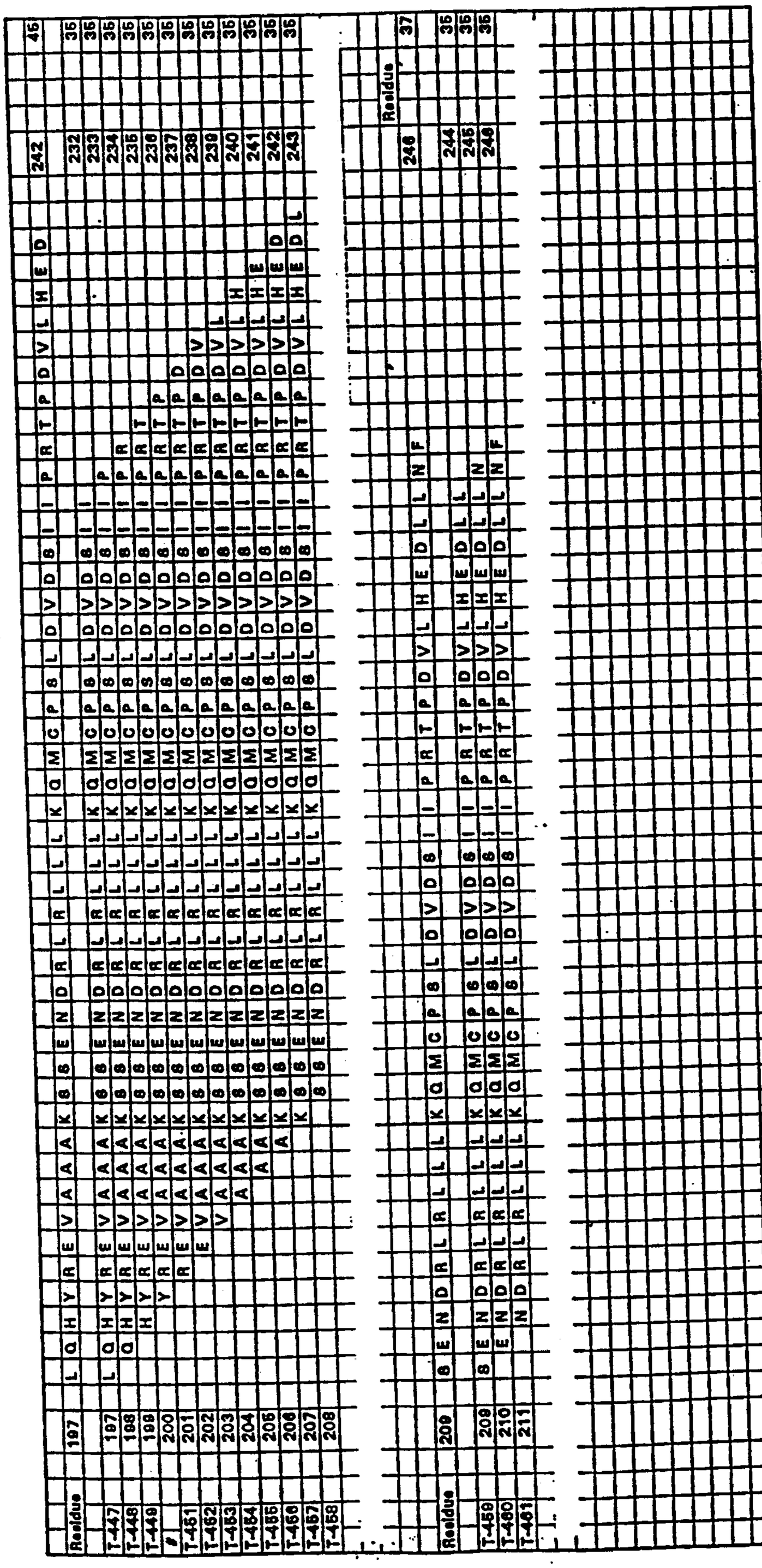

45
35
35
35
35
35
35
35
35
35
35
35
35
242
232
233
234
235
236
237
238
239
240
241
242
243
Residue
37
35
35
35
246
244
245
246
Residue
200
Residue
200
T-447
T-448
T-449
T-450
T-451
T-452
T-453
T-454
T-455
T-456
T-457
T-458
T-459
T-460
T-461

FIG. 51B

DOMAIN 1:

174 P-L-L-V-L-Q-A-G-F-F-L-L-T-R-I-L-T-I-L-P-Q-S-L-D-S-W-W-T-S-L-N-F-L-G-G-T-T-V-C-L-G-Q-N-S-Q-S-P 220

P-L-L-V-L-Q-A-G-F-F-L-L-T-R-I-L-T-I-L-P-Q-S-L-D-S-W-W-T-S-L-N-F-L-G-G-T-T-V-C-L-G-Q-N-S-Q-S-P

L-L-V-L-Q-A-G-F-F-L-L-T-R-I-L-T-I-L-P-Q-S-L-D-S-W-W-T-S-L-N-F-L-G-G-T-T-V-C-L-G-Q-N-S-Q-S-P

L-V-L-Q-A-G-F-F-L-L-T-R-I-L-T-I-L-P-Q-S-L-D-S-W-W-T-S-L-N-F-L-G-G-T-T-V-C-L-G-Q-N-S-Q-S-P

V-L-Q-A-G-F-F-L-L-T-R-I-L-T-I-L-P-Q-S-L-D-S-W-W-T-S-L-N-F-L-G-G-T-T-V-C-L-G-Q-N-S-Q-S-P

L-Q-A-G-F-F-L-L-T-R-I-L-T-I-L-P-Q-S-L-D-S-W-W-T-S-L-N-F-L-G-G-T-T-V-C-L-G-Q-N-S-Q-S-P

Q-A-G-F-F-L-L-T-R-I-L-T-I-L-P-Q-S-L-D-S-W-W-T-S-L-N-F-L-G-G-T-T-V-C-L-G-Q-N-S-Q-S-P

A-G-F-F-L-L-T-R-I-L-T-I-L-P-Q-S-L-D-S-W-W-T-S-L-N-F-L-G-G-T-T-V-C-L-G-Q-N-S-Q-S-P

G-F-F-L-L-T-R-I-L-T-I-L-P-Q-S-L-D-S-W-W-T-S-L-N-F-L-G-G-T-T-V-C-L-G-Q-N-S-Q-S-P

F-F-L-L-T-R-I-L-T-I-L-P-Q-S-L-D-S-W-W-T-S-L-N-F-L-G-G-T-T-V-C-L-G-Q-N-S-Q-S-P

F-L-L-T-R-I-L-T-I-L-P-Q-S-L-D-S-W-W-T-S-L-N-F-L-G-G-T-T-V-C-L-G-Q-N-S-Q-S-P

L-L-T-R-I-L-T-I-L-P-Q-S-L-D-S-W-W-T-S-L-N-F-L-G-G-T-T-V-C-L-G-Q-N-S-Q-S-P

FIG.52A

DOMAIN II:

223 P-G-Y-R-W-M-C-L-R-R-F-I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T-S-T-G-P-C-R-T-C-M-T-T 291

P-G-Y-R-W-M-C-L-R-R-F-I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L
G-Y-R-W-M-C-L-R-R-F-I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P
Y-R-W-M-C-L-R-R-F-I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V
R-W-M-C-L-R-R-F-I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C
W-M-C-L-R-R-F-I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P
M-C-L-R-R-F-I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L
C-L-R-R-F-I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I
L-R-R-F-I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P
R-R-F-I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G
R-F-I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S
F-I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-G-G-S-S
I-I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T

I-F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T-S
F-L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T-S-T
L-F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T-S-T-G
F-I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T-S-T-G-P
I-L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T-S-T-G-P-C
L-L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T-S-T-G-P-C-R
L-L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T-S-T-G-P-C-R-T
L-C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T-S-T-G-P-C-R-T-C
C-L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T-S-T-G-P-C-R-T-C-M
L-I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T-S-T-G-P-C-R-T-C-M-T
I-F-L-L-V-L-L-D-Y-Q-G-M-L-P-V-C-P-L-I-P-G-S-S-T-S-T-G-P-C-R-T-C-M-T-T

FIG.52B

RECTIFIED SHEET (RULE 91)

HIV1LAI	(DP-178; SEQ ID:1)	YTSLIHSLIEESQNQQEKNEQELLELDKWASLWNWF
HIV1SF2	(DP-185; SEQ ID:3)	YTNTIYNLLEESQNQQEKNEQELLELDKWASLWNWF
HIV1RF	(SEQ ID:4)	YTGIIYNLLEESQNQQEKNEQELLELDKWANLWNWF
HIV1MN	(SEQ ID:5)	YTSLIYSLLEKSQTQQEKNEQELLELDKWASLWNWF
HIV2ROD	(SEQ ID:6)	LEANISKSLEQAQIQQEKNMYELQKLNSWDIFGNWF
HIV2NIHZ	(SEQ ID:7)	LEANISQSLEQAQIQQEKNMYELQKLNSWDVFTNWL
DP180	(SEQ ID:2)	SSESFTLLEQWNNWKLQLAEQWLEQINEKHYLEDIS
DP118	(SEQ ID:10)	QQLLDVVKRQQEMLRLTVWGTKNLQARVTAIEKYLKDQ
DP125	(SEQ ID:8)	CGGNNLLRAIEAQQHLLQLTVWGIKQLQARILAVERYLKDQ
DP116	(SEQ ID:9)	LQARILAVERYLKDQQQ