GEC1_HUMAN Mass: 61641 Score: 4276 Matches: 166(1134) Sequences: 23(18) emPAI: 5.50

Glypican-1 OS=Homo sapiens GN=GPC1 PE=1 SV=2

Sequence Coverage: 46%

Matched peptides are underlined

1 MELRARGWWL LCAAAALVAC ARGDPASKSR SCGEVRQIYG AKGFSLSDVP
51 QAEISGEHLR ICPQGYTCCT SEMEENLAMR SHAELETALR DSSRVLQAML
101 ATQLRSFDDH FQHLLANDSER TLQATFPGAF GELYTQNARA FRDLYSELRL
151 YYRGANLHLE ETLAEFWARL LERLFKQLHP QLLLFDDYLD CLGRQAEALR
201 PFGEAPRELR LRATRAFVAA RSFVQGLGVA SDVVRKVAQV PLGPECSRAV
251 MKLVYCAHCL GVPGARPCPD YCRNVLKGCL ANQADLDAEW RNLLDSMVLI
301 TDKFWGTSGV ESVIGSVHTW LAEAINALQD NRDTLTAFVI QGCGNPKVNP
351 QGPGPEEKRR RGKLAPRERP PSGTLEKLVS EARAQLRIVQ DFWISLPGTL
401 CSEKMALSTA SDDRCWNGMA RGRYLPEVMG DGLANQINNP EVEVDITKPD
451 MTIRQQIMQL KIMTNRLRSA YNGNDVDFQD ASDDGSGSGS GDGCLDDLCS
501 RKVSRKSSSS RTPLTHALPG LSEQEGQKTS AASCPQPFTF LLPLLLFLAL
551 TVARPRWR (SEQ ID NO: 23)

Figure 1

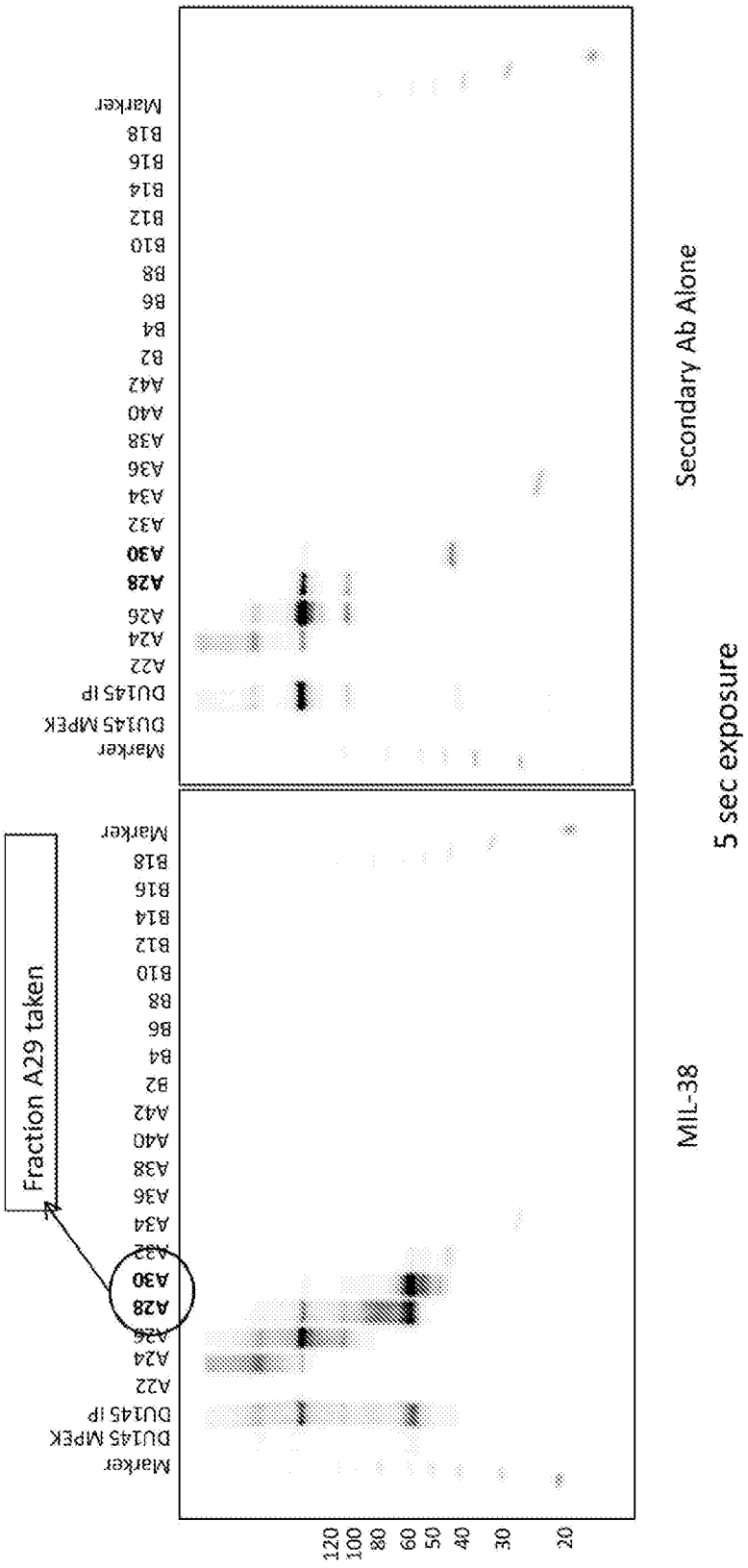

Secondary Ab Alone
5 sec exposure
MIL-38
Fraction A29 taken
Marker
DU145 MPEK
DU145 IP
A22
A24
A26
A28
A30
A32
A34
A36
A38
A40
A42
B2
B4
B6
B8
B10
B12
B14
B16
B18
Marker
120
100
80
60
50
40
30
20

Figure 2

Mass Spectrometry of Size Exclusion HPLC Fraction #29

GPC1_HUMAN Mass: 61641 Score: 290 Matches: 8(8) Sequences: 8(8) emPAI: 0.82
Glypican-1 OS=Homo sapiens GN=GPC1 PE=1 SV=2

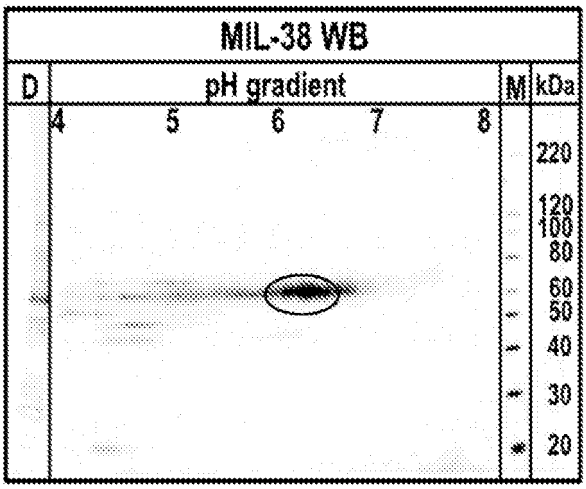
Sequence Coverage: 14%

Matched peptides are underlined

1 MELRARGWWL LCAAAALVAC ARGDFASKSR SCGEVRQIYG AKGFSLSDVP
51 QAEISGEHLR ICPQGYTCCT SEWEENLAMR SHAELETALR DSSRVLQAML
101 RTQLRSFTDH FQHLLMDSER TLQATFPGAF GELNTQWARA FRDLVSELRL
151 YYRGANLALE ETLAEFWARL LERLFKQLHP QLLIPDDYLD CLGNQAEALR
201 EFGEAFRELR LRATRAFVAA RSFVQGLGYA SDVVRKVAQV FLGFECSRAV
251 MELVYCARCL GVPGARFCPD YCRNVLKGCL ANQADLDAEW RNLLDSMVLI
301 TDKFWGTSGV ESVIGSYHTW LAFAINALQD MNDTLTAKVI QDCGNFKVMP
351 QGRGPEEWAR KGKLAPKERP PSFTLEKLVS EAKQQLAIVQ DFWISLPGTL
401 CEEKMALSTA SDDACWNCMA RGRYLPEVMG DGLANQIMMP EVEVDITKED
451 MTIRQQIMQL KIMTNRLRSA YMGNDVDFQD ASDDGSGSGS GDGCLDLLCS
501 RKVSRKSSSS RTPLITHALPG LSEQEGQKTS AASCPQFFTF LLFLLLFLAL
551 IVARRMWR (SEQ ID NO: 23)

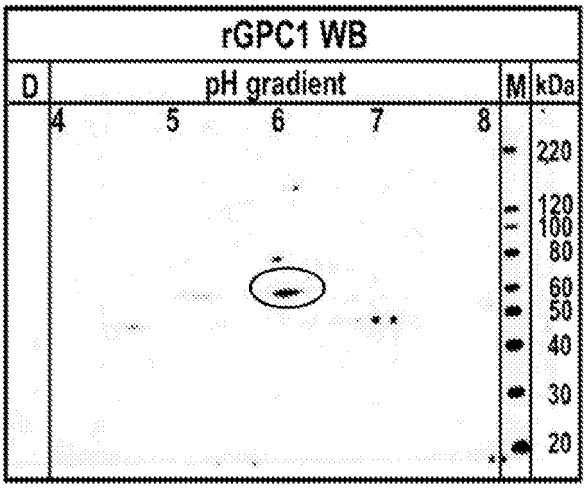
Figure 3

A



30 sec exposure

B



1 min exposure

Figure 4

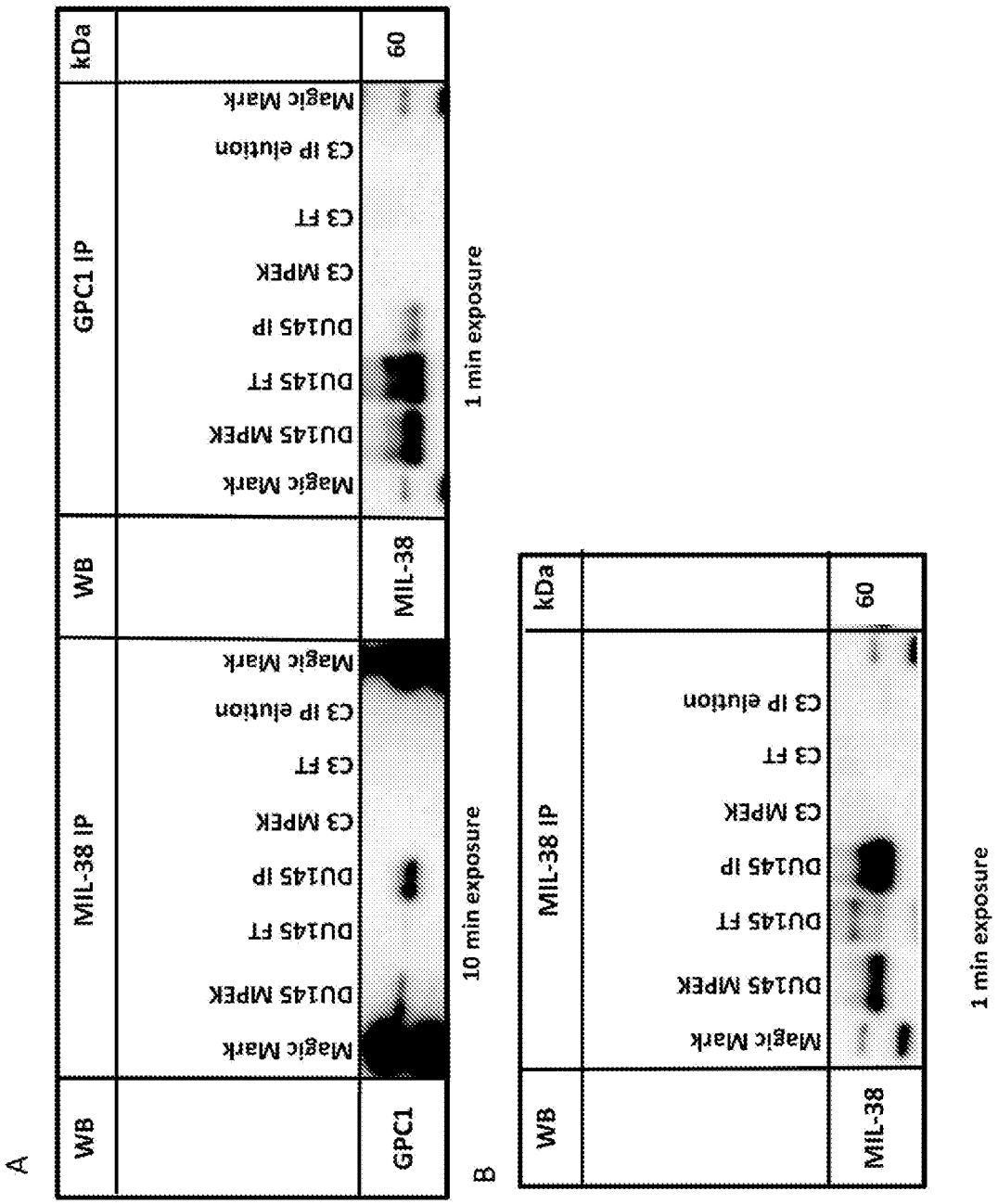

A
WB
MIL-38 IP
GPC1 IP
kDa
GPC1
MIL-38
60
Magic Mark
DU145 MPEK
DU145 FT
DU145 IP
C3 MPEK
C3 FT
C3 IP elution
Magic Mark
10 min exposure
1 min exposure
B
WB
MIL-38 IP
kDa
MIL-38
60
Magic Mark
DU145 MPEK
DU145 FT
DU145 IP
C3 MPEK
C3 FT
C3 IP elution
1 min exposure

Figure 5

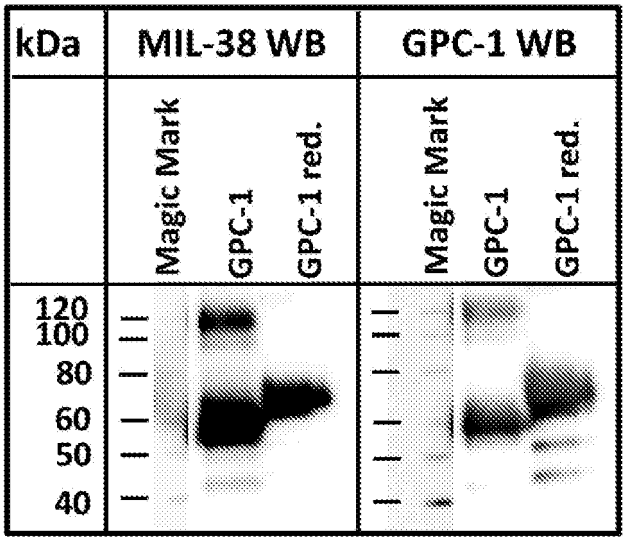
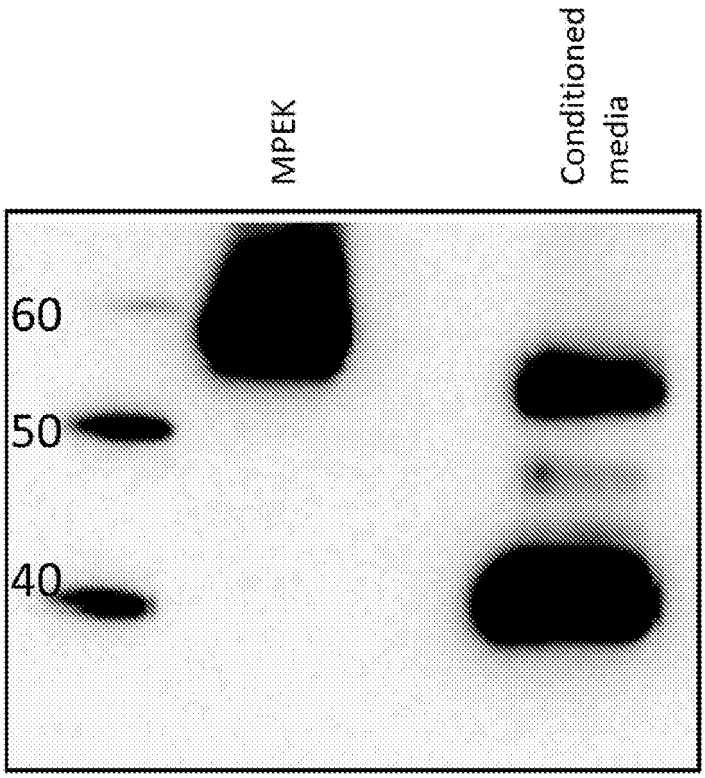

kDa
MIL-38 WB
GPC-1 WB
Magic Mark
GPC-1
GPC-1 red.
Magic Mark
GPC-1
GPC-1 red.
120
100
80
60
50
40

Figure 6



5 min exposure

Figure 7

A

GEC1_HUMAN Mass: 61641 Score: 882 Matches: 30(22) Sequences: 11(9) emPAI: 0.68

Glypican-1 OS=Homo sapiens GN=GPC1 PE=1 SV=2

Sequence Coverage: 16%

Matched peptides are underlined

1 MELRARGWWL LCAAAALVAC ARGDPASKSR SCGEVRQIYG AKGFSLSDVP
51 QAEISGEHLR ICPQGYTCCT SEMEENLANR SHAELETALR DSSRVLQAML
101 ATQLASFDDH FQHLLMDSER TLQATFPGAF GELYTQNARA FRDLYSELRL
151 YYRGANLHLE ETLAEFWARL LERLFKQLHF QLLLPDDYLD CLGKQAZAIR
201 PFGEAFRELR LRATRAFVAA RSPVQGLGYA SDVTRKVAQV FLGPEC3RAY
251 MRLYYCAHCL GVPGARPCPD YCANVLKGCL ANQADLDAEW RNLLDSMVLI
301 TDKFWGTSGY ESVIGSVHTW LAEAINALQD NRDTLTAKVI QGCGNPKVNP
351 QGPGFEEKRA RGKLAFREAP PSGTLEKLVS EAKAQLRDVQ DFWISLPGTL
401 CSEKMALSTA SDDRCWNGMA RGRYLPEYMG DGLANQIKNP EVEVDITKPD
451 MTIRCQIMQL KIMTNRLRSA YMGNDVDFQD ASDDGSGGGS GDGCLDMLCS
501 RKVSRKSSSS RTPLTHALES LSEQEGQKTS AASCPQPPTF LLPLLALFLA
551 TVARREAWR (SEQ ID NO: 23)

Figure 8

B

GEC1_HUMAN Mass: 61641 Score: 1551 Matches: 48(43) Sequences: 11(10) emPAI: 1.42

Glypican-1 OS=Homo sapiens GN=GPC1 PE=1 SV=3

Sequence Coverage: 18%

Matched peptides are underlined

1 MELRARGWWL LCLAAALVAC ARGDPASKSR SCGEVRQIYG AKGFSLSDVP
51 QAELSGEHLR ICPQGYTCCT SEMEENLAER SHAELETALR DSSRVLQAML
101 ATQLRSFTDH FQHLLMDSER TLQATTFGAF GELYTQWARA FRDLYSELRL
151 YYRGANLHIE ETLAEFWARL LERLFKQLHF QLLLPDDYLD CLGKQAEALR
201 PFGEAPRELR IRATRAFVAA RSFVQGLGTA SDVVRKVAQV PLGPECSRAV
251 MKLVYCARCL GVPGARECFD YCSNVLKGCL ANQADLDAEW RMLLDSMVLI
301 TDKFWGTSGV ESVIGSVHTM LAEAIMALQD NRDTLTAFVI QGCGMPKYMP
351 QGPGPEERSR RGKLAFRERP PSGTLEKLVS ZAKAQLRTVQ DFWISLPGTL
401 CSEKMALSTA SDDRCWNGMA RGRYLPEVMG DGLAMQTMWP EVEVDTIKFD
451 MTIRQQIMQL KIMTNRLRSA YNGMDVDPOD ASDNGSGSGS GNGCLDDLCS
501 RKVSRKSSSS RTPLITHALPS LSEQEGQKTS AASCPQPFTF LLPLLLFLAL
551 TVARPRWR (SEQ ID NO: 23)

Figure 8

C

GPLA_HUMAN Mass: 61641 Score: 1364 Matches: 39(33) Sequences: 13(9) emPAI: 1.18

Glycogen-1 OS=Homo sapiens GN=GPC1 PE=1 SV=2

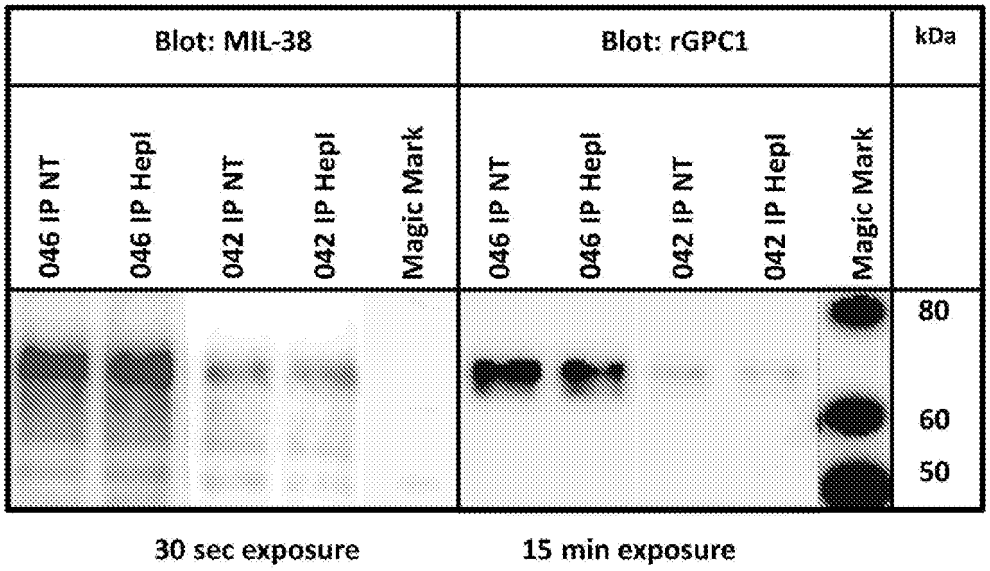
Sequence Coverage: 24%

Matched peptides are underlined

1	MELRARGWWL LCAAAALVAC ARGDPASKSR SCGEVRQIYG AKGFSLSDVP
51	QAEISGEHLR ICPQGYTCCT SEMEEMLAKW SHAELETALR DSSWTLQAWL
101	ATQLRSFDDH FQHLLNDSER TLQATFPGAF FRDLYSELRL
151	YRGANLHLE ETLAEFWARL LERLFKQLNP QLLLFDDYLD CLGKQAEALR
201	EFGEAFRELR LRATRAFVAA RSFVQGLGYA SDVVBNVAQV PLGPECSRAV
251	MKLVYCAHCL GVPGARPCFD YCRMVLKGCL ANQADLDAEW RWLLDSMVLI
301	IDKFWGTSGV ESVIGSVHTM LAEAINALQD NKDTLTAKVI QGCGNPKVNP
351	QGPGPEERRR RGKLAFRERF PSGTLEKLYS EANAQLRDVQ DFWISLPGTL
401	CSEKMALSTA SDDRCWNGMA RGRYLFEVMG DSLANQIMNP EVEVDITKFD
451	WTIRQQIMQL KIMTNRLRSA YMGNDWDFQD ASDDGSGSGS GDGCLDVLCS
501	RKVSRKSSSS RTPLTHALFG LSEQEGQRTS AASCFQFPTF LLFLLLFLAL
551	TVARPAWR (SEQ ID NO: 23)

Figure 8

A


Blot: MIL-38
Blot: rGPC1
kDa
046 IP NT
046 IP Hepl
042 IP NT
042 IP Hepl
Magic Mark
046 IP NT
046 IP Hepl
042 IP NT
042 IP Hepl
Magic Mark
80
60
50
30 sec exposure
15 min exposure

B

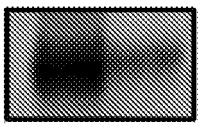

Prostate cancer
Normal prostate

Figure 9

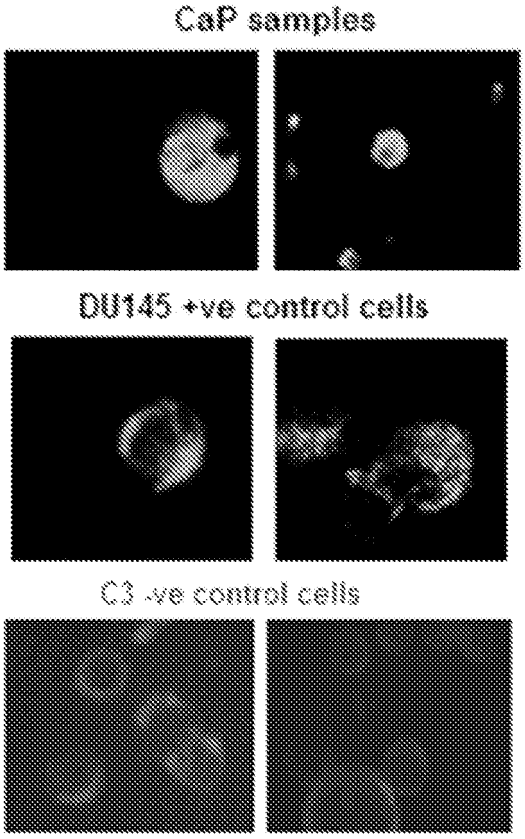


Figure 10

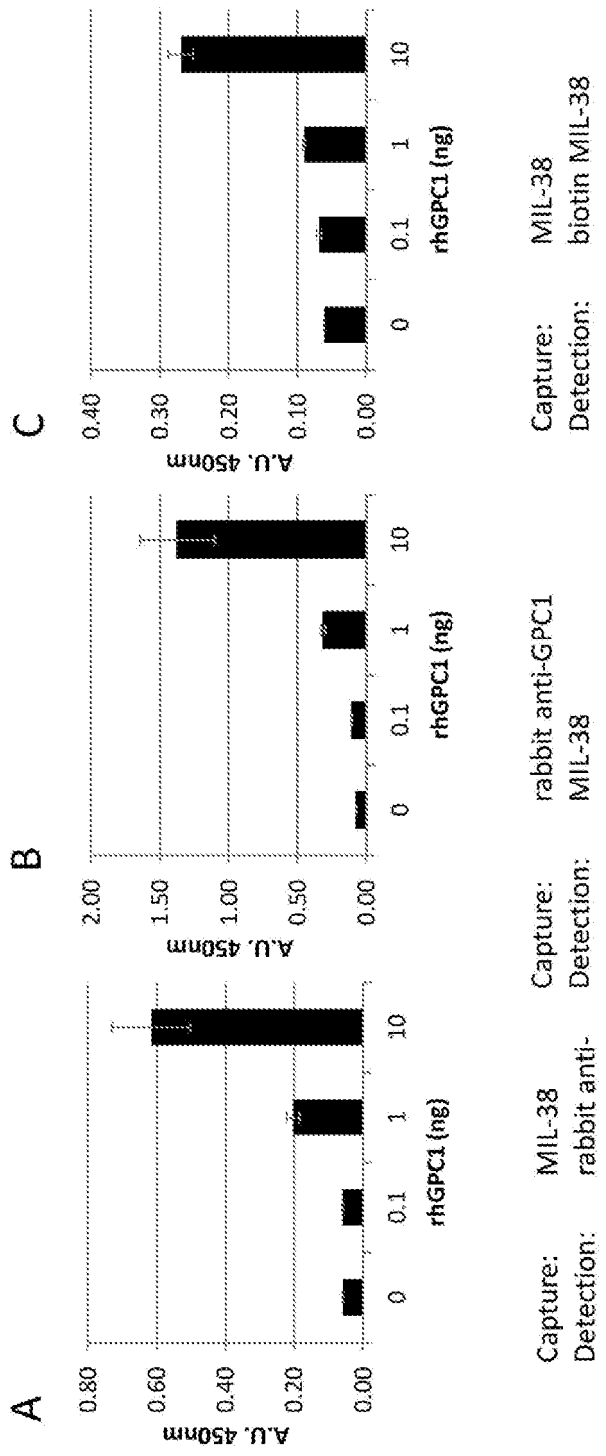

A
B
C
A.U. 450nm
rhGPC1 (ng)
0
0.1
1
10
0.00
0.20
0.40
0.60
0.80
0.50
1.00
1.50
2.00
0.10
0.30
0.40
Capture: MIL-38
Detection: rabbit anti-GPC1
Capture: rabbit anti-GPC1
Detection: MIL-38
Capture: MIL-38
Detection: biotin MIL-38

Figure 11

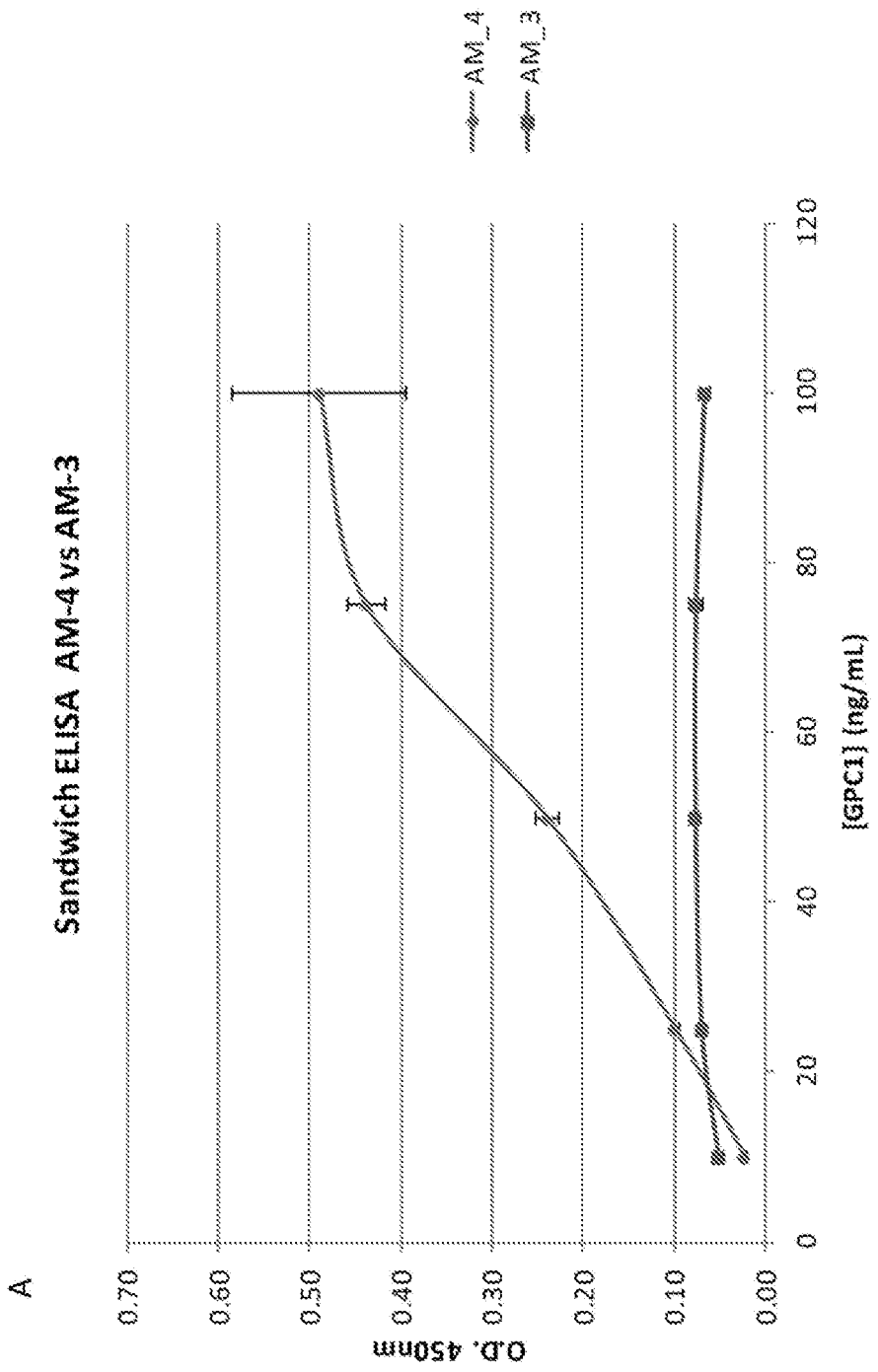

A
Sandwich ELISA AM-4 vs AM-3
AM_4
AM_3
O.D. 450nm
0.70
0.60
0.50
0.40
0.30
0.20
0.10
0.00
0
20
40
60
80
100
120
[GPC1] (ng/mL)

Figure 12

B

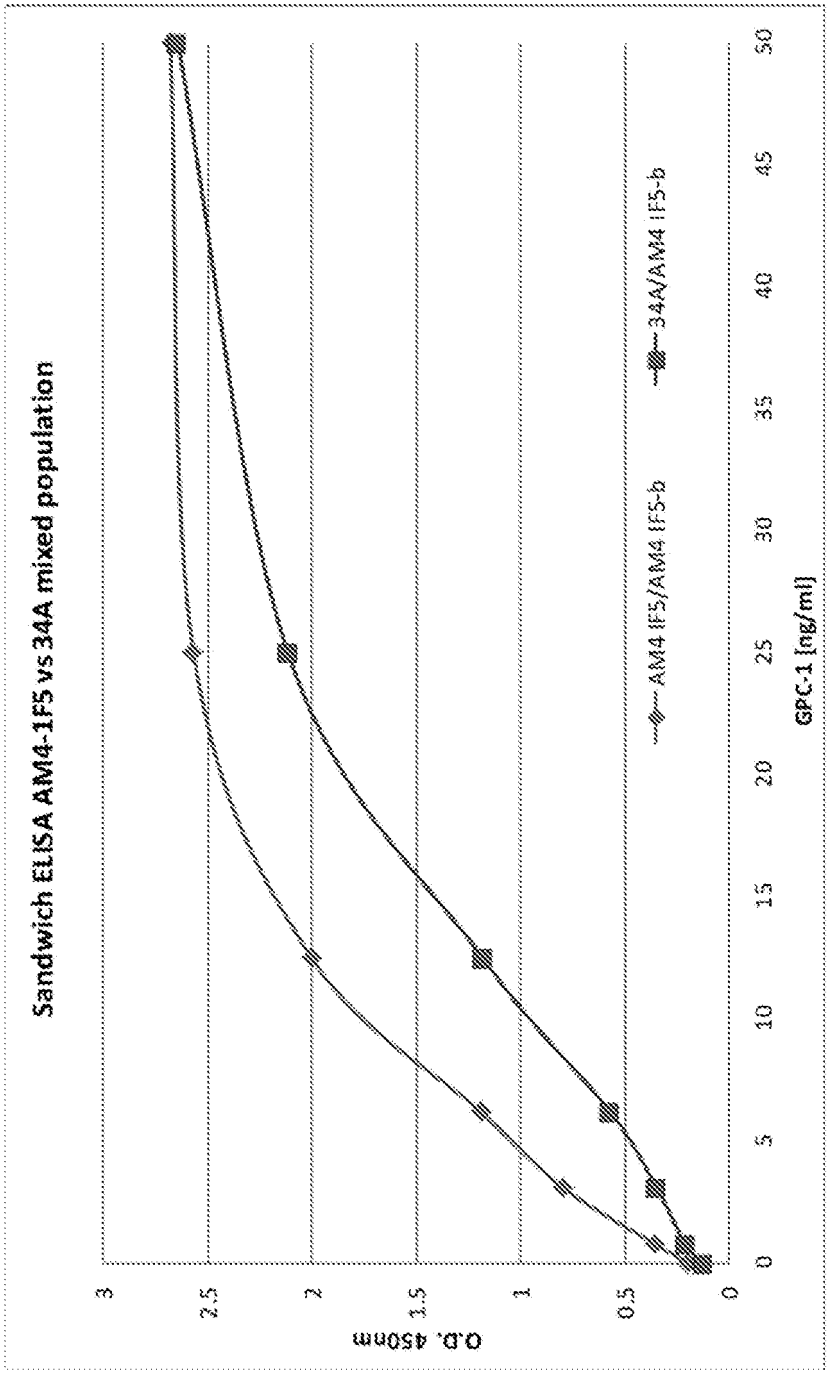

Sandwich ELISA AM4-1F5 vs 34A mixed population
O.D. 450nm
GPC-1 [ng/ml]
AM4 IF5/AM4 IF5-b
34A/AM4 IF5-b
0
0.5
1
1.5
2
2.5
3
0
5
10
15
20
25
30
35
40
45
50

Figure 12

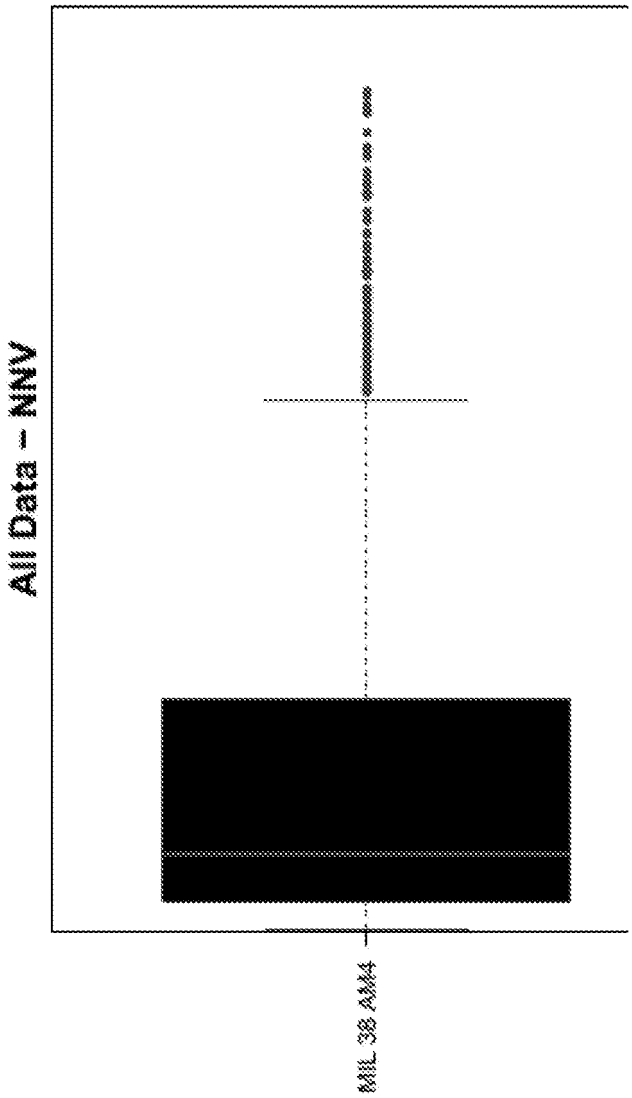


Figure 13

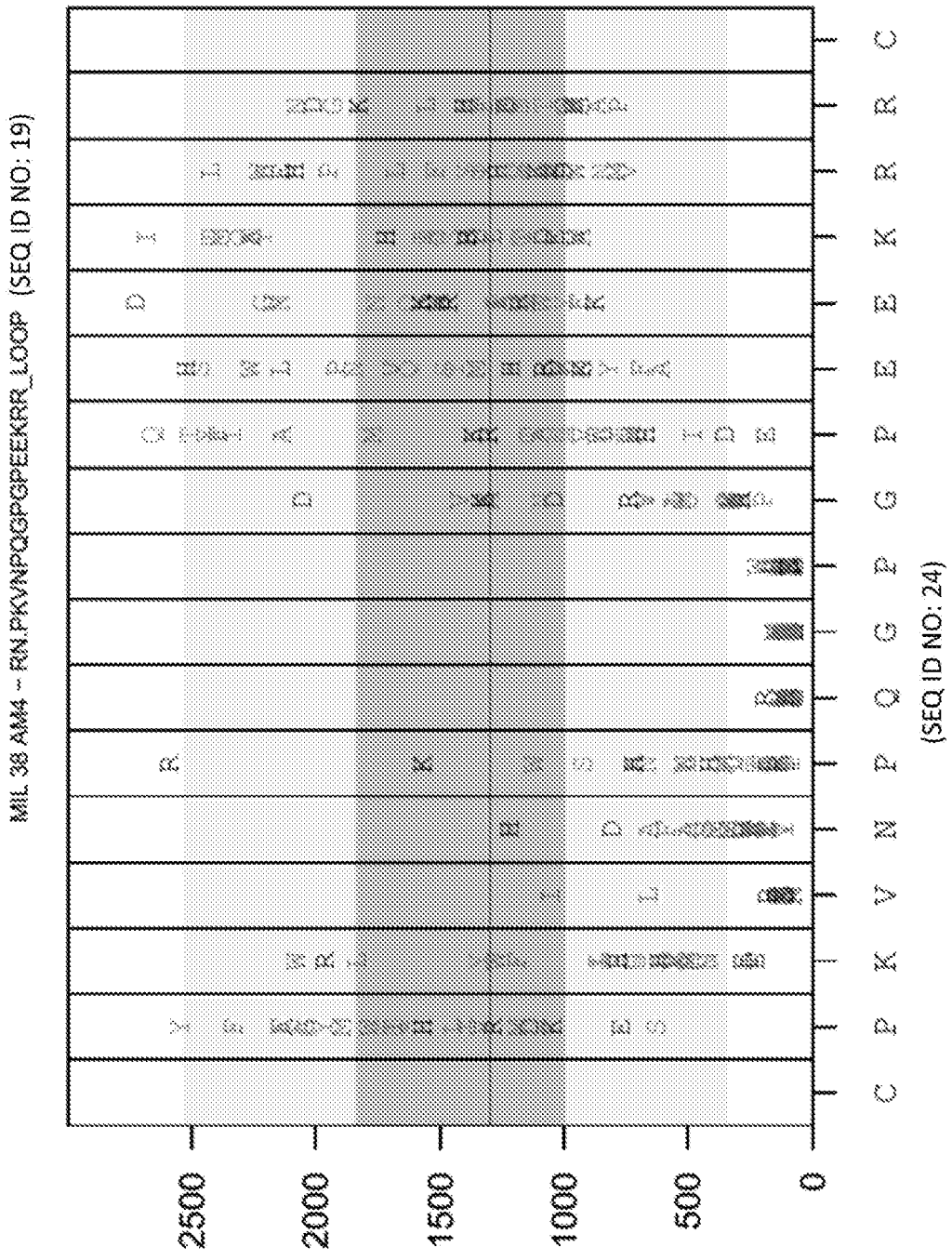

MIL 38 AM4 – RN.PKVNPQGPGPEEKRR_LOOP (SEQ ID NO: 19)
2500
2000
1500
1000
500
0
C P K V N P Q G P G P E E K R R C
(SEQ ID NO: 24)

Figure 14