

1
SEQUENCE LISTING

<110> Astellas Pharma Inc.
<120> NOVEL ANTI-HUMAN NGF ANTIBODY FAB FRAGMENT
<130> FA1535-16021
<150> JP2015-104806
<151> 2015-05-22
<160> 12
<170> PatentIn version 3.5
<210> 1
<211> 675
<212> DNA
<213> Artificial Sequence

<220>
<223> DNA encoding h1f.6 heavy chain fragment

<400> 1
cagggtgcagc tacagcagtg gggcgcagga ctgttgaagc cttcggagac cctgtccctc 60
acctgcgctg tctatgggtg gtccctcagt ggtaactact ggagctgggt ccgccagccc 120
ccagggaagg ggctggagtg gattggggaa atcgaccata gtggaagcac caacaacaac 180
ccgtccctca agagtgcagt caccatatca gtaggcacgt ccaagaacca gttctccctg 240
aagctgagct ctgtgaccgc cgcggacacg gctgtgtatt actgttcgag agatgggggc 300
cccgaatcgg ggatgggggc ttttgataac tggggccaag ggacaatggt caccgtctcc 360
tcagcctcca ccaaggggcc atcgggtctc cccctggcac cctcctccaa gaggacctct 420
gggggacacg cggccctggg ctgcctggtc aaggactact tccccgaacc ggtgacggtg 480
tcgtggaact caggcgcctt gaccagcggc gtgcacacct tcccggctgt cctacagtcc 540
tcaggactct actcccttag tagcgtggtg accgtgccct ccagcagctt gggcaccag 600
acctacatct gcaacgtgaa tcacaagccc agcaacacca aggtggacaa gaaagttgag 660
cccaaattct gtgac 675

<210> 2
<211> 672
<212> DNA
<213> Artificial Sequence

<220>
<223> DNA encoding h1f.7 heavy chain fragment

<400> 2
cagggtgcagc tacagcagtg gggcgcagga ctgttgaagc cttcggagac cctgtccctc 60
acctgcgctg totatggtgg gtccttcagt ggttactact ggagctgggt ccgccagccc 120
ccagggaagg ggctggagtg gattggggaa atcgaccata gtggaagcac caacaacaac 180
ccgtccctca agagtcgagt caccatatca gtaggcacgt ccaagaacca gttctccctg 240
aagctgagct ctgtgaccgc cgcggacacg gctgtgtatt actgttcgag agatgggggc 300
cccgaatcgg ggatgggggc ttttgatata tggggccaag ggacaatggt caccgtctcc 360
tcagcctcca ccaaggggcc atcggctctc cccctggcac cctcctccaa gagcacctct 420
gggggcacag cggccctggg ctgcctggtc aaggactact tccccgaacc ggtgacggtg 480
tcgtggaact caggcgcctt gaccagcggc gtgcacacct tcccggctgt cctacagtcc 540
tcaggactct actcccttag tagcgtggtg accgtgccct ccagcagctt gggcaccag 600
acctacatct gcaacgtgaa tcacaagccc agcaacacca aggtggacaa gaaagttgag 660
cccaaattct gt 672

<210> 3
<211> 678
<212> DNA
<213> Artificial Sequence

<220>
<223> DNA encoding hlf.8 heavy chain fragment

<400> 3
cagggtgcagc tacagcagtg gggcgcagga ctgttgaagc cttcggagac cctgtccctc 60
acctgcgctg totatggtgg gtccttcagt ggttactact ggagctgggt ccgccagccc 120
ccagggaagg ggctggagtg gattggggaa atcgaccata gtggaagcac caacaacaac 180
ccgtccctca agagtcgagt caccatatca gtaggcacgt ccaagaacca gttctccctg 240
aagctgagct ctgtgaccgc cgcggacacg gctgtgtatt actgttcgag agatgggggc 300
cccgaatcgg ggatgggggc ttttgatata tggggccaag ggacaatggt caccgtctcc 360
tcagcctcca ccaaggggcc atcggctctc cccctggcac cctcctccaa gagcacctct 420
gggggcacag cggccctggg ctgcctggtc aaggactact tccccgaacc ggtgacggtg 480
tcgtggaact caggcgcctt gaccagcggc gtgcacacct tcccggctgt cctacagtcc 540
tcaggactct actcccttag tagcgtggtg accgtgccct ccagcagctt gggcaccag 600
acctacatct gcaacgtgaa tcacaagccc agcaacacca aggtggacaa gaaagttgag 660

cccaaatott gtgcagcc

678

<210> 4
 <211> 657
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> DNA encoding 1-15 (N52D-A)-Fab' fragment light chain

<400> 4
 gatattgtga tgactcagtc tccactctcc ctgcccgtca cccctggaga gccggcctcc 60
 atctcctgca ggtctagtca gagcctcctg catagtaatg gattcaacta tttgggttgg 120
 tacctgcaga agccagggca gtctccacag ctctgatct atttgggttc taatcgggcc 180
 tccgggggtcc ctgacagggtt cagtggcagt ggatcaggca cagattttac tctgaaaatc 240
 agcagagtgg aggctgagga tgttgggggtt tattactgca tgcaagctct acaaactccg 300
 tacacttttg gccaggggac caagctggag atcaaacgga ctgtggctgc accatctgtc 360
 ttcatottcc cgccatctga tgagcagttg aaatctggaa ctgcctctgt tgtgtgcctg 420
 ctgaataact tctatcccag agaggccaaa gtacagtgga aggtggataa cgccctccaa 480
 tcgggtaact cccaggagag tgtcacagag caggacagca aggacagcac ctacagcctc 540
 agcagcacc ctagcgtgag caaagcagac tacgagaaac acaaagtcta cgctgcgaa 600
 gtcaccatc agggcctgag ctgcccgtc acaaagagct tcaacagggg agagtgt 657

<210> 5
 <211> 225
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> h1f.6 heavy chain fragment

<400> 5
 Gln Val Gln Leu Gln Gln Trp Gly Ala Gly Leu Leu Lys Pro Ser Glu
 1 5 10 15
 Thr Leu Ser Leu Thr Cys Ala Val Tyr Gly Gly Ser Phe Ser Gly Tyr
 20 25 30
 Tyr Trp Ser Trp Val Arg Gln Pro Pro Gly Lys Gly Leu Glu Trp Ile
 35 40 45

Gly Glu Ile Asp His Ser Gly Ser Thr Asn Asn⁴ Asn Pro Ser Leu Lys
 50 55 60
 Ser Arg Val Thr Ile Ser Val Gly Thr Ser Lys Asn Gln Phe Ser Leu
 65 70 75 80
 Lys Leu Ser Ser Val Thr Ala Ala Asp Thr Ala Val Tyr Tyr Cys Ser
 85 90 95
 Arg Asp Gly Gly Pro Glu Ser Gly Met Gly Ala Phe Asp Ile Trp Gly
 100 105 110
 Gln Gly Thr Met Val Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser
 115 120 125
 Val Phe Pro Leu Ala Pro Ser Ser Lys Ser Thr Ser Gly Gly Thr Ala
 130 135 140
 Ala Leu Gly Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val
 145 150 155 160
 Ser Trp Asn Ser Gly Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala
 165 170 175
 Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val
 180 185 190
 Pro Ser Ser Ser Leu Gly Thr Gln Thr Tyr Ile Cys Asn Val Asn His
 195 200 205
 Lys Pro Ser Asn Thr Lys Val Asp Lys Lys Val Glu Pro Lys Ser Cys
 210 215 220

Asp
 225

<210> 6
 <211> 224
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> h1f.7 heavy chain fragment
 <400> 6

Gln Val Gln Leu Gln Gln Trp Gly Ala Gly Leu Leu Lys Pro Ser Glu
1 5 10 15

Thr Leu Ser Leu Thr Cys Ala Val Tyr Gly Gly Ser Phe Ser Gly Tyr
20 25 30

Tyr Trp Ser Trp Val Arg Gln Pro Pro Gly Lys Gly Leu Glu Trp Ile
35 40 45

Gly Glu Ile Asp His Ser Gly Ser Thr Asn Asn Asn Pro Ser Leu Lys
50 55 60

Ser Arg Val Thr Ile Ser Val Gly Thr Ser Lys Asn Gln Phe Ser Leu
65 70 75 80

Lys Leu Ser Ser Val Thr Ala Ala Asp Thr Ala Val Tyr Tyr Cys Ser
85 90 95

Arg Asp Gly Gly Pro Glu Ser Gly Met Gly Ala Phe Asp Ile Trp Gly
100 105 110

Gln Gly Thr Met Val Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser
115 120 125

Val Phe Pro Leu Ala Pro Ser Ser Lys Ser Thr Ser Gly Gly Thr Ala
130 135 140

Ala Leu Gly Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val
145 150 155 160

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

Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val
180 185 190

Pro Ser Ser Ser Leu Gly Thr Gln Thr Tyr Ile Cys Asn Val Asn His
195 200 205

Lys Pro Ser Asn Thr Lys Val Asp Lys Lys Val Glu Pro Lys Ser Cys
210 215 220

<210> 7
 <211> 226
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> h1f.8 heavy chain fragment

<400> 7

Gln Val Gln Leu Gln Gln Trp Gly Ala Gly Leu Leu Lys Pro Ser Glu
 1 5 10 15

Thr Leu Ser Leu Thr Cys Ala Val Tyr Gly Gly Ser Phe Ser Gly Tyr
 20 25 30

Tyr Trp Ser Trp Val Arg Gln Pro Pro Gly Lys Gly Leu Glu Trp Ile
 35 40 45

Gly Glu Ile Asp His Ser Gly Ser Thr Asn Asn Asn Pro Ser Leu Lys
 50 55 60

Ser Arg Val Thr Ile Ser Val Gly Thr Ser Lys Asn Gln Phe Ser Leu
 65 70 75 80

Lys Leu Ser Ser Val Thr Ala Ala Asp Thr Ala Val Tyr Tyr Cys Ser
 85 90 95

Arg Asp Gly Gly Pro Glu Ser Gly Met Gly Ala Phe Asp Ile Trp Gly
 100 105 110

Gln Gly Thr Met Val Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser
 115 120 125

Val Phe Pro Leu Ala Pro Ser Ser Lys Ser Thr Ser Gly Gly Thr Ala
 130 135 140

Ala Leu Gly Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val
 145 150 155 160

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

Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val
 180 185 190

Pro Ser Ser Ser Leu Gly Thr Gln Thr Tyr Ile Cys Asn Val Asn His
 195 200 205

Lys Pro Ser Asn Thr Lys Val Asp Lys Lys Val Glu Pro Lys Ser Cys
 210 215 220

Ala Ala
 225

<210> 8
 <211> 219
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> 1-15(N52D-A)-Fab' fragment light chain

<400> 8

Asp Ile Val Met Thr Gln Ser Pro Leu Ser Leu Pro Val Thr Pro Gly
 1 5 10 15

Glu Pro Ala Ser Ile Ser Cys Arg Ser Ser Gln Ser Leu Leu His Ser
 20 25 30

Asn Gly Phe Asn Tyr Leu Gly Trp Tyr Leu Gln Lys Pro Gly Gln Ser
 35 40 45

Pro Gln Leu Leu Ile Tyr Leu Gly Ser Asn Arg Ala Ser Gly Val Pro
 50 55 60

Asp Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Lys Ile
 65 70 75 80

Ser Arg Val Glu Ala Glu Asp Val Gly Val Tyr Tyr Cys Met Gln Ala
 85 90 95

Leu Gln Thr Pro Tyr Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Lys
 100 105 110

Arg Thr Val Ala Ala Pro Ser Val Phe Ile Phe Pro Pro Ser Asp Glu
 115 120 125

Gln Leu Lys Ser Gly Thr Ala Ser Val Val Cys Leu Leu Asn Asn Phe
 130 135 140

Tyr Pro Arg Glu Ala Lys Val Gln Trp Lys Val Asp Asn Ala Leu Gln
145 150 155 160

Ser Gly Asn Ser Gln Glu Ser Val Thr Glu Gln Asp Ser Lys Asp Ser
165 170 175

Thr Tyr Ser Leu Ser Ser Thr Leu Thr Leu Ser Lys Ala Asp Tyr Glu
180 185 190

Lys His Lys Val Tyr Ala Cys Glu Val Thr His Gln Gly Leu Ser Ser
195 200 205

Pro Val Thr Lys Ser Phe Asn Arg Gly Glu Cys
210 215

<210> 9
<211> 363
<212> DNA
<213> Artificial Sequence

<220>
<223> VH gene of anti-human NGF antibody

<220>
<221> CDS
<222> (1).. (363)

<400> 9
cag gtg cag cta cag cag tgg ggc gca gga ctg ttg aag cct tcg gag 48
Gln Val Gln Leu Gln Gln Trp Gly Ala Gly Leu Leu Lys Pro Ser Glu
1 5 10 15
acc ctg tcc ctc acc tgc gct gtc tat ggt ggg tcc ttc agt ggt tac 96
Thr Leu Ser Leu Thr Cys Ala Val Tyr Gly Gly Ser Phe Ser Gly Tyr
20 25 30
tac tgg agc tgg gtc cgc cag ccc cca ggg aag ggg ctg gag tgg att 144
Tyr Trp Ser Trp Val Arg Gln Pro Pro Gly Lys Gly Leu Glu Trp Ile
35 40 45
ggg gaa atc gac cat agt gga agc acc aac aac aac ccg tcc ctc aag 192
Gly Glu Ile Asp His Ser Gly Ser Thr Asn Asn Asn Pro Ser Leu Lys
50 55 60
agt cga gtc acc ata tca gta ggc acg tcc aag aac cag ttc tcc ctg 240
Ser Arg Val Thr Ile Ser Val Gly Thr Ser Lys Asn Gln Phe Ser Leu
65 70 75 80
aag ctg agc tct gtg acc gcc gcg gac acg gct gtg tat tac tgt tcg 288

Lys Leu Ser Ser Val Thr Ala Ala Asp Thr⁹ Ala Val Tyr Tyr Cys Ser
85 90 95

aga gat ggg ggc ccc gaa tcg ggg atg ggg gct ttt gat atc tgg ggc 336
Arg Asp Gly Gly Pro Glu Ser Gly Met Gly Ala Phe Asp Ile Trp Gly
100 105 110

caa ggg aca atg gtc acc gtc tcc tca 363
Gln Gly Thr Met Val Thr Val Ser Ser
115 120

<210> 10
<211> 121
<212> PRT
<213> Artificial Sequence

<220>
<223> Synthetic Construct

<400> 10

Gln Val Gln Leu Gln Gln Trp Gly Ala Gly Leu Leu Lys Pro Ser Glu
1 5 10 15

Thr Leu Ser Leu Thr Cys Ala Val Tyr Gly Gly Ser Phe Ser Gly Tyr
20 25 30

Tyr Trp Ser Trp Val Arg Gln Pro Pro Gly Lys Gly Leu Glu Trp Ile
35 40 45

Gly Glu Ile Asp His Ser Gly Ser Thr Asn Asn Asn Pro Ser Leu Lys
50 55 60

Ser Arg Val Thr Ile Ser Val Gly Thr Ser Lys Asn Gln Phe Ser Leu
65 70 75 80

Lys Leu Ser Ser Val Thr Ala Ala Asp Thr Ala Val Tyr Tyr Cys Ser
85 90 95

Arg Asp Gly Gly Pro Glu Ser Gly Met Gly Ala Phe Asp Ile Trp Gly
100 105 110

Gln Gly Thr Met Val Thr Val Ser Ser
115 120

<210> 11
<211> 339
<212> DNA

<213> Artificial Sequence

<220>

<223> VL gene of anti-human NGF antibody

<220>

<221> CDS

<222> (1)..(339)

<400> 11

gat	att	gtg	atg	act	cag	tct	cca	ctc	tcc	ctg	ccc	gtc	acc	cct	gga	48
Asp	Ile	Val	Met	Thr	Gln	Ser	Pro	Leu	Ser	Leu	Pro	Val	Thr	Pro	Gly	
1				5					10					15		

gag	cgc	gcc	tcc	atc	tcc	tgc	agg	tct	agt	cag	agc	ctc	ctg	cat	agt	96
Glu	Pro	Ala	Ser	Ile	Ser	Cys	Arg	Ser	Ser	Gln	Ser	Leu	Leu	His	Ser	
			20					25					30			

aat	gga	ttc	aac	tat	ttg	ggt	tgg	tac	ctg	cag	aag	cca	ggg	cag	tct	144
Asn	Gly	Phe	Asn	Tyr	Leu	Gly	Trp	Tyr	Leu	Gln	Lys	Pro	Gly	Gln	Ser	
		35				40						45				

cca	cag	ctc	ctg	atc	tat	ttg	ggt	tct	aat	cgg	gcc	tcc	ggg	gtc	cct	192
Pro	Gln	Leu	Leu	Ile	Tyr	Leu	Gly	Ser	Asn	Arg	Ala	Ser	Gly	Val	Pro	
		50				55					60					

gac	agg	ttc	agt	ggc	agt	gga	tca	ggc	aca	gat	ttt	act	ctg	aaa	atc	240
Asp	Arg	Phe	Ser	Gly	Ser	Gly	Ser	Gly	Thr	Asp	Phe	Thr	Leu	Lys	Ile	
65				70					75						80	

agc	aga	gtg	gag	gct	gag	gat	gtt	ggg	gtt	tat	tac	tgc	atg	caa	gct	288
Ser	Arg	Val	Glu	Ala	Glu	Asp	Val	Gly	Val	Tyr	Tyr	Cys	Met	Gln	Ala	
			85					90						95		

cta	caa	act	cgc	tac	act	ttt	ggc	cag	ggg	acc	aag	ctg	gag	atc	aaa	336
Leu	Gln	Thr	Pro	Tyr	Thr	Phe	Gly	Gln	Gly	Thr	Lys	Leu	Glu	Ile	Lys	
			100					105					110			

cgg																339
Arg																

<210> 12

<211> 113

<212> PRT

<213> Artificial Sequence

<220>

<223> Synthetic Construct

<400> 12

Asp	Ile	Val	Met	Thr	Gln	Ser	Pro	Leu	Ser	Leu	Pro	Val	Thr	Pro	Gly
1				5					10					15	

Glu Pro Ala Ser Ile Ser Cys Arg Ser Ser Gln Ser Leu Leu His Ser
 20 25 30

Asn Gly Phe Asn Tyr Leu Gly Trp Tyr Leu Gln Lys Pro Gly Gln Ser
 35 40 45

Pro Gln Leu Leu Ile Tyr Leu Gly Ser Asn Arg Ala Ser Gly Val Pro
 50 55 60

Asp Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Lys Ile
 65 70 75 80

Ser Arg Val Glu Ala Glu Asp Val Gly Val Tyr Tyr Cys Met Gln Ala
 85 90 95

Leu Gln Thr Pro Tyr Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Lys
 100 105 110

Arg