

108095P502PC_SeqList_ST25.txt
SEQUENCE LISTING

<110> Merz Pharma GmbH & Co. KGaA
Grein, Sven
Hölscher, Kerstin
Eylenstein, Annett

<120> NOVEL METHOD FOR THE MANUFACTURING OF DI-CHAIN PROTEINS FOR USE
IN HUMANS

<130> 108095P502PC

<140> PCT/EP2012/xxxxxx
<141> 2012-12-21

<150> EP 11010178.9
<151> 2011-12-23

<150> US 61/579,771
<151> 2011-12-23

<160> 13

<170> PatentIn version 3.5

<210> 1
<211> 422
<212> PRT
<213> artificial

<220>
<223> modified BONT/E light chain

<400> 1

Met Pro Lys Ile Asn Ser Phe Asn Tyr Asn Asp Pro Val Asn Asp Arg
1 5 10 15

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

Phe Asn Ile Met Lys Asn Ile Trp Ile Ile Pro Glu Arg Asn Val Ile
35 40 45

Gly Thr Thr Pro Gln Asp Phe His Pro Pro Thr Ser Leu Lys Asn Gly
50 55 60

Asp Ser Ser Tyr Tyr Asp Pro Asn Tyr Leu Gln Ser Asp Glu Glu Lys
65 70 75 80

Asp Arg Phe Leu Lys Ile Val Thr Lys Ile Phe Asn Arg Ile Asn Asn
85 90 95

Asn Leu Ser Gly Gly Ile Leu Leu Glu Glu Leu Ser Lys Ala Asn Pro
100 105 110

Tyr Leu Gly Asn Asp Asn Thr Pro Asp Asn Gln Phe His Ile Gly Asp
115 120 125

Ala Ser Ala Val Glu Ile Lys Phe Ser Asn Gly Ser Gln Asp Ile Leu
Seite 1

130

135

140

Leu Pro Asn Val Ile Ile Met Gly Ala Glu Pro Asp Leu Phe Glu Thr
145 150 155 160

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

Gly Phe Gly Ser Ile Ala Ile Val Thr Phe Ser Pro Glu Tyr Ser Phe
180 185 190

Arg Phe Asn Asp Asn Ser Met Asn Glu Phe Ile Gln Asp Pro Ala Leu
195 200 205

Thr Leu Met His Glu Leu Ile His Ser Leu His Gly Leu Tyr Gly Ala
210 215 220

Lys Gly Ile Thr Thr Lys Tyr Thr Ile Thr Gln Lys Gln Asn Pro Leu
225 230 235 240

Ile Thr Asn Ile Arg Gly Thr Asn Ile Glu Glu Phe Leu Thr Phe Gly
245 250 255

Gly Thr Asp Leu Asn Ile Ile Thr Ser Ala Gln Ser Asn Asp Ile Tyr
260 265 270

Thr Asn Leu Leu Ala Asp Tyr Lys Lys Ile Ala Ser Lys Leu Ser Lys
275 280 285

Val Gln Val Ser Asn Pro Leu Leu Asn Pro Tyr Lys Asp Val Phe Glu
290 295 300

Ala Lys Tyr Gly Leu Asp Lys Asp Ala Ser Gly Ile Tyr Ser Val Asn
305 310 315 320

Ile Asn Lys Phe Asn Asp Ile Phe Lys Lys Leu Tyr Ser Phe Thr Glu
325 330 335

Phe Asp Leu Ala Thr Lys Phe Gln Val Lys Cys Arg Gln Thr Tyr Ile
340 345 350

Gly Gln Tyr Lys Tyr Phe Lys Leu Ser Asn Leu Leu Asn Asp Ser Ile
355 360 365

Tyr Asn Ile Ser Glu Gly Tyr Asn Ile Asn Asn Leu Lys Val Asn Phe
370 375 380

Arg Gly Gln Asn Ala Asn Leu Asn Pro Arg Ile Ile Thr Pro Ile Thr
385 390 395 400

Gly Arg Gly Leu Val Lys Lys Ile Ile Arg Phe Cys Lys Asn Ile Val

405

410

415

Ser Val Lys Gly Ile Arg
420

<210> 2
<211> 830
<212> PRT
<213> artificial

<220>
<223> modified BONT/E heavy chain

<400> 2

Lys Ser Ile Cys Ile Glu Ile Asn Asn Gly Glu Leu Phe Phe Val Ala
1 5 10 15

Ser Glu Asn Ser Tyr Asn Asp Asp Asn Ile Asn Thr Pro Lys Glu Ile
20 25 30

Asp Asp Thr Val Thr Ser Asn Asn Asn Tyr Glu Asn Asp Leu Asp Gln
35 40 45

Val Ile Leu Asn Phe Asn Ser Glu Ser Ala Pro Gly Leu Ser Asp Glu
50 55 60

Lys Leu Asn Leu Thr Ile Gln Asn Asp Ala Tyr Ile Pro Lys Tyr Asp
65 70 75 80

Ser Asn Gly Thr Ser Asp Ile Glu Gln His Asp Val Asn Glu Leu Asn
85 90 95

Val Phe Phe Tyr Leu Asp Ala Gln Lys Val Pro Glu Gly Glu Asn Asn
100 105 110

Val Asn Leu Thr Ser Ser Ile Asp Thr Ala Leu Leu Glu Gln Pro Lys
115 120 125

Ile Tyr Thr Phe Phe Ser Ser Glu Phe Ile Asn Asn Val Asn Lys Pro
130 135 140

Val Gln Ala Ala Leu Phe Val Ser Trp Ile Gln Gln Val Leu Val Asp
145 150 155 160

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

Ile Ser Ile Val Val Pro Tyr Ile Gly Leu Ala Leu Asn Ile Gly Asn
180 185 190

Glu Ala Gln Lys Gly Asn Phe Lys Asp Ala Leu Glu Leu Leu Gly Ala
195 200 205

108095P502PC_SeqList_ST25.txt

Gly Ile Leu Leu Glu Phe Glu Pro Glu Leu Leu Ile Pro Thr Ile Leu
210 215 220

Val Phe Thr Ile Lys Ser Phe Leu Gly Ser Ser Asp Asn Lys Asn Lys
225 230 235 240

Val Ile Lys Ala Ile Asn Asn Ala Leu Lys Glu Arg Asp Glu Lys Trp
245 250 255

Lys Glu Val Tyr Ser Phe Ile Val Ser Asn Trp Met Thr Lys Ile Asn
260 265 270

Thr Gln Phe Asn Lys Arg Lys Glu Gln Met Tyr Gln Ala Leu Gln Asn
275 280 285

Gln Val Asn Ala Ile Lys Thr Ile Ile Glu Ser Lys Tyr Asn Ser Tyr
290 295 300

Thr Leu Glu Glu Lys Asn Glu Leu Thr Asn Lys Tyr Asp Ile Lys Gln
305 310 315 320

Ile Glu Asn Glu Leu Asn Gln Lys Val Ser Ile Ala Met Asn Asn Ile
325 330 335

Asp Arg Phe Leu Thr Glu Ser Ser Ile Ser Tyr Leu Met Lys Leu Ile
340 345 350

Asn Glu Val Lys Ile Asn Lys Leu Arg Glu Tyr Asp Glu Asn Val Lys
355 360 365

Thr Tyr Leu Leu Asn Tyr Ile Ile Gln His Gly Ser Ile Leu Gly Glu
370 375 380

Ser Gln Gln Glu Leu Asn Ser Met Val Thr Asp Thr Leu Asn Asn Ser
385 390 395 400

Ile Pro Phe Lys Leu Ser Ser Tyr Thr Asp Asp Lys Ile Leu Ile Ser
405 410 415

Tyr Phe Asn Lys Phe Phe Lys Arg Ile Lys Ser Ser Ser Val Leu Asn
420 425 430

Met Arg Tyr Lys Asn Asp Lys Tyr Val Asp Thr Ser Gly Tyr Asp Ser
435 440 445

Asn Ile Asn Ile Asn Gly Asp Val Tyr Lys Tyr Pro Thr Asn Lys Asn
450 455 460

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

108095P502PC_SeqList_ST25.txt

Asn Asp Tyr Ile Ile Tyr Asp Asn Lys Tyr Lys Asn Phe Ser Ile Ser
 485 490 495
 Phe Trp Val Arg Ile Pro Asn Tyr Asp Asn Lys Ile Val Asn Val Asn
 500 505 510
 Asn Glu Tyr Thr Ile Ile Asn Cys Met Arg Asp Asn Asn Ser Gly Trp
 515 520 525
 Lys Val Ser Leu Asn His Asn Glu Ile Ile Trp Thr Leu Gln Asp Asn
 530 535 540
 Ala Gly Ile Asn Gln Lys Leu Ala Phe Asn Tyr Gly Asn Ala Asn Gly
 545 550 555 560
 Ile Ser Asp Tyr Ile Asn Lys Trp Ile Phe Val Thr Ile Thr Asn Asp
 565 570 575
 Arg Leu Gly Asp Ser Lys Leu Tyr Ile Asn Gly Asn Leu Ile Asp Gln
 580 585 590
 Lys Ser Ile Leu Asn Leu Gly Asn Ile His Val Ser Asp Asn Ile Leu
 595 600 605
 Phe Lys Ile Val Asn Cys Ser Tyr Thr Arg Tyr Ile Gly Ile Arg Tyr
 610 615 620
 Phe Asn Ile Phe Asp Lys Glu Leu Asp Glu Thr Glu Ile Gln Thr Leu
 625 630 635 640
 Tyr Ser Asn Glu Pro Asn Thr Asn Ile Leu Lys Asp Phe Trp Gly Asn
 645 650 655
 Tyr Leu Leu Tyr Asp Lys Glu Tyr Tyr Leu Leu Asn Val Leu Lys Pro
 660 665 670
 Asn Asn Phe Ile Asp Arg Arg Lys Asp Ser Thr Leu Ser Ile Asn Asn
 675 680 685
 Ile Arg Ser Thr Ile Leu Leu Ala Asn Arg Leu Tyr Ser Gly Ile Lys
 690 695 700
 Val Lys Ile Gln Arg Val Asn Asn Ser Ser Thr Asn Asp Asn Leu Val
 705 710 715 720
 Arg Lys Asn Asp Gln Val Tyr Ile Asn Phe Val Ala Ser Lys Thr His
 725 730 735
 Leu Phe Pro Leu Tyr Ala Asp Thr Ala Thr Thr Asn Lys Glu Lys Thr
 740 745 750

108095P502PC_SeqList_ST25.txt

Ile Lys Ile Ser Ser Ser Gly Asn Arg Phe Asn Gln Val Val Val Met
755 760 765

Asn Ser Val Gly Asn Asn Cys Thr Met Asn Phe Lys Asn Asn Asn Gly
770 775 780

Asn Asn Ile Gly Leu Leu Gly Phe Lys Ala Asp Thr Val Val Ala Ser
785 790 795 800

Thr Trp Tyr Tyr Thr His Met Arg Asp His Thr Asn Ser Asn Gly Cys
805 810 815

Phe Trp Asn Phe Ile Ser Glu Glu His Gly Trp Gln Glu Lys
820 825 830

<210> 3
<211> 427
<212> PRT
<213> artificial

<220>
<223> modified BONT/E light chain_2

<400> 3

Met Pro Lys Ile Asn Ser Phe Asn Tyr Asn Asp Pro Val Asn Asp Arg
1 5 10 15

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

Phe Asn Ile Met Lys Asn Ile Trp Ile Ile Pro Glu Arg Asn Val Ile
35 40 45

Gly Thr Thr Pro Gln Asp Phe His Pro Pro Thr Ser Leu Lys Asn Gly
50 55 60

Asp Ser Ser Tyr Tyr Asp Pro Asn Tyr Leu Gln Ser Asp Glu Glu Lys
65 70 75 80

Asp Arg Phe Leu Lys Ile Val Thr Lys Ile Phe Asn Arg Ile Asn Asn
85 90 95

Asn Leu Ser Gly Gly Ile Leu Leu Glu Glu Leu Ser Lys Ala Asn Pro
100 105 110

Tyr Leu Gly Asn Asp Asn Thr Pro Asp Asn Gln Phe His Ile Gly Asp
115 120 125

Ala Ser Ala Val Glu Ile Lys Phe Ser Asn Gly Ser Gln Asp Ile Leu
130 135 140

Leu Pro Asn Val Ile Ile Met Gly Ala Glu Pro Asp Leu Phe Glu Thr

145	1080955P302PC_Sequence125.txt														150	155														160
Asn	Ser	Ser	Asn	Ile 165	Ser	Leu	Arg	Asn	Asn 170	Tyr	Met	Pro	Ser	Asn 175	His															
Gly	Phe	Gly	Ser 180	Ile	Ala	Ile	Val	Thr 185	Phe	Ser	Pro	Glu	Tyr 190	Ser	Phe															
Arg	Phe	Asn 195	Asp	Asn	Ser	Met	Asn 200	Glu	Phe	Ile	Gln	Asp 205	Pro	Ala	Leu															
Thr	Leu 210	Met	His	Glu	Leu	Ile 215	His	Ser	Leu	His	Gly 220	Leu	Tyr	Gly	Ala															
Lys 225	Gly	Ile	Thr	Thr	Lys 230	Tyr	Thr	Ile	Thr	Gln 235	Lys	Gln	Asn	Pro	Leu 240															
Ile	Thr	Asn	Ile	Arg 245	Gly	Thr	Asn	Ile	Glu 250	Glu	Phe	Leu	Thr	Phe 255	Gly															
Gly	Thr	Asp	Leu 260	Asn	Ile	Ile	Thr	Ser 265	Ala	Gln	Ser	Asn	Asp 270	Ile	Tyr															
Thr	Asn	Leu 275	Leu	Ala	Asp	Tyr	Lys 280	Lys	Ile	Ala	Ser	Lys 285	Leu	Ser	Lys															
Val	Gln 290	Val	Ser	Asn	Pro	Leu 295	Leu	Asn	Pro	Tyr	Lys 300	Asp	Val	Phe	Glu															
Ala 305	Lys	Tyr	Gly	Leu	Asp 310	Lys	Asp	Ala	Ser	Gly 315	Ile	Tyr	Ser	Val	Asn 320															
Ile	Asn	Lys	Phe	Asn 325	Asp	Ile	Phe	Lys	Lys 330	Leu	Tyr	Ser	Phe	Thr 335	Glu															
Phe	Asp	Leu	Ala 340	Thr	Lys	Phe	Gln	Val 345	Lys	Cys	Arg	Gln	Thr 350	Tyr	Ile															
Gly	Gln	Tyr 355	Lys	Tyr	Phe	Lys	Leu 360	Ser	Asn	Leu	Leu	Asn 365	Asp	Ser	Ile															
Tyr	Asn 370	Ile	Ser	Glu	Gly	Tyr 375	Asn	Ile	Asn	Asn	Leu 380	Lys	Val	Asn	Phe															
Arg 385	Gly	Gln	Asn	Ala	Asn 390	Leu	Asn	Pro	Arg	Ile 395	Ile	Thr	Pro	Ile	Thr 400															
Gly	Arg	Gly	Leu	Val 405	Lys	Lys	Ile	Ile	Arg 410	Phe	Cys	Val	Arg	Gly 415	Ile															
Ile	Thr	Ser	Lys	Thr	Lys	Ser	Leu	Val	Pro	Arg																				

420

<210> 4
<211> 835
<212> PRT
<213> artificial

<220>
<223> modified BoNT/E heavy chain_2

<400> 4

Gly Ser Lys Ala Leu Asn Asp Leu Cys Ile Glu Ile Asn Asn Gly Glu
1 5 10 15

Leu Phe Phe Val Ala Ser Glu Asn Ser Tyr Asn Asp Asp Asn Ile Asn
20 25 30

Thr Pro Lys Glu Ile Asp Asp Thr Val Thr Ser Asn Asn Asn Tyr Glu
35 40 45

Asn Asp Leu Asp Gln Val Ile Leu Asn Phe Asn Ser Glu Ser Ala Pro
50 55 60

Gly Leu Ser Asp Glu Lys Leu Asn Leu Thr Ile Gln Asn Asp Ala Tyr
65 70 75 80

Ile Pro Lys Tyr Asp Ser Asn Gly Thr Ser Asp Ile Glu Gln His Asp
85 90 95

Val Asn Glu Leu Asn Val Phe Phe Tyr Leu Asp Ala Gln Lys Val Pro
100 105 110

Glu Gly Glu Asn Asn Val Asn Leu Thr Ser Ser Ile Asp Thr Ala Leu
115 120 125

Leu Glu Gln Pro Lys Ile Tyr Thr Phe Phe Ser Ser Glu Phe Ile Asn
130 135 140

Asn Val Asn Lys Pro Val Gln Ala Ala Leu Phe Val Ser Trp Ile Gln
145 150 155 160

Gln Val Leu Val Asp Phe Thr Thr Glu Ala Asn Gln Lys Ser Thr Val
165 170 175

Asp Lys Ile Ala Asp Ile Ser Ile Val Val Pro Tyr Ile Gly Leu Ala
180 185 190

Leu Asn Ile Gly Asn Glu Ala Gln Lys Gly Asn Phe Lys Asp Ala Leu
195 200 205

Glu Leu Leu Gly Ala Gly Ile Leu Leu Glu Phe Glu Pro Glu Leu Leu
210 215 220

108095P502PC_SeqList_ST25.txt

Ile Pro Thr Ile Leu Val Phe Thr Ile Lys Ser Phe Leu Gly Ser Ser
225 230 235 240

Asp Asn Lys Asn Lys Val Ile Lys Ala Ile Asn Asn Ala Leu Lys Glu
245 250 255

Arg Asp Glu Lys Trp Lys Glu Val Tyr Ser Phe Ile Val Ser Asn Trp
260 265 270

Met Thr Lys Ile Asn Thr Gln Phe Asn Lys Arg Lys Glu Gln Met Tyr
275 280 285

Gln Ala Leu Gln Asn Gln Val Asn Ala Ile Lys Thr Ile Ile Glu Ser
290 295 300

Lys Tyr Asn Ser Tyr Thr Leu Glu Glu Lys Asn Glu Leu Thr Asn Lys
305 310 315 320

Tyr Asp Ile Lys Gln Ile Glu Asn Glu Leu Asn Gln Lys Val Ser Ile
325 330 335

Ala Met Asn Asn Ile Asp Arg Phe Leu Thr Glu Ser Ser Ile Ser Tyr
340 345 350

Leu Met Lys Leu Ile Asn Glu Val Lys Ile Asn Lys Leu Arg Glu Tyr
355 360 365

Asp Glu Asn Val Lys Thr Tyr Leu Leu Asn Tyr Ile Ile Gln His Gly
370 375 380

Ser Ile Leu Gly Glu Ser Gln Gln Glu Leu Asn Ser Met Val Thr Asp
385 390 395 400

Thr Leu Asn Asn Ser Ile Pro Phe Lys Leu Ser Ser Tyr Thr Asp Asp
405 410 415

Lys Ile Leu Ile Ser Tyr Phe Asn Lys Phe Phe Lys Arg Ile Lys Ser
420 425 430

Ser Ser Val Leu Asn Met Arg Tyr Lys Asn Asp Lys Tyr Val Asp Thr
435 440 445

Ser Gly Tyr Asp Ser Asn Ile Asn Ile Asn Gly Asp Val Tyr Lys Tyr
450 455 460

Pro Thr Asn Lys Asn Gln Phe Gly Ile Tyr Asn Asp Lys Leu Ser Glu
465 470 475 480

Val Asn Ile Ser Gln Asn Asp Tyr Ile Ile Tyr Asp Asn Lys Tyr Lys
485 490 495

108095P502PC_SeqList_ST25.txt

Asn Phe Ser Ile Ser Phe Trp Val Arg Ile Pro Asn Tyr Asp Asn Lys
 500 505 510
 Ile Val Asn Val Asn Asn Glu Tyr Thr Ile Ile Asn Cys Met Arg Asp
 515 520 525
 Asn Asn Ser Gly Trp Lys Val Ser Leu Asn His Asn Glu Ile Ile Trp
 530 535 540
 Thr Leu Gln Asp Asn Ala Gly Ile Asn Gln Lys Leu Ala Phe Asn Tyr
 545 550 555 560
 Gly Asn Ala Asn Gly Ile Ser Asp Tyr Ile Asn Lys Trp Ile Phe Val
 565 570 575
 Thr Ile Thr Asn Asp Arg Leu Gly Asp Ser Lys Leu Tyr Ile Asn Gly
 580 585 590
 Asn Leu Ile Asp Gln Lys Ser Ile Leu Asn Leu Gly Asn Ile His Val
 595 600 605
 Ser Asp Asn Ile Leu Phe Lys Ile Val Asn Cys Ser Tyr Thr Arg Tyr
 610 615 620
 Ile Gly Ile Arg Tyr Phe Asn Ile Phe Asp Lys Glu Leu Asp Glu Thr
 625 630 635 640
 Glu Ile Gln Thr Leu Tyr Ser Asn Glu Pro Asn Thr Asn Ile Leu Lys
 645 650 655
 Asp Phe Trp Gly Asn Tyr Leu Leu Tyr Asp Lys Glu Tyr Tyr Leu Leu
 660 665 670
 Asn Val Leu Lys Pro Asn Asn Phe Ile Asp Arg Arg Lys Asp Ser Thr
 675 680 685
 Leu Ser Ile Asn Asn Ile Arg Ser Thr Ile Leu Leu Ala Asn Arg Leu
 690 695 700
 Tyr Ser Gly Ile Lys Val Lys Ile Gln Arg Val Asn Asn Ser Ser Thr
 705 710 715 720
 Asn Asp Asn Leu Val Arg Lys Asn Asp Gln Val Tyr Ile Asn Phe Val
 725 730 735
 Ala Ser Lys Thr His Leu Phe Pro Leu Tyr Ala Asp Thr Ala Thr Thr
 740 745 750
 Asn Lys Glu Lys Thr Ile Lys Ile Ser Ser Ser Gly Asn Arg Phe Asn
 755 760 765

108095P502PC_SeqList_ST25.txt

Gln Val Val Val Met Asn Ser Val Gly Asn Asn Cys Thr Met Asn Phe
770 775 780

Lys Asn Asn Asn Gly Asn Asn Ile Gly Leu Leu Gly Phe Lys Ala Asp
785 790 795 800

Thr Val Val Ala Ser Thr Trp Tyr Tyr Thr His Met Arg Asp His Thr
805 810 815

Asn Ser Asn Gly Cys Phe Trp Asn Phe Ile Ser Glu Glu His Gly Trp
820 825 830

Gln Glu Lys
835

<210> 5
<211> 3789
<212> DNA
<213> artificial

<220>
<223> modified BONT/E single chain precursor DNA

<400> 5
atgccgaaaa tcaacagctt caactataac gatccggtga acgatcgtac catcctgtat 60
attaaaccgg gcggttgcca ggaattttac aaaagcttca acatcatgaa aaacatctgg 120
attattccgg aacgtaacgt gattggcacc acccgcagg attttcatcc gccgaccagc 180
ctgaaaaacg gcgatagcag ctattatgat ccgaactatc tgcagtctga tgaagaaaaa 240
gatcgcttcc tgaaaatcgt gacaaaaatc ttcaaccgca tcaacaacaa cctgagcggc 300
ggcattctgc tggaagaact gagcaaagcg aatccgtatc tgggcaacga taacactcca 360
gataaccagt ttcatattgg tgatgcgagc gcggtggaaa ttaaatttag caacggctct 420
caggacattc tgctgccgaa cgtgattatt atgggcgcgg aaccggacct gtttgaaacc 480
aacagcagca acattagcct gcgtaacaac tatatgccga gcaaccatgg ttttggcagc 540
attgcgattg tgaccttag cccggaatat agctttcgct tcaacgataa cagcatgaac 600
gaatttattc aggaccggc gctgaccctg atgcacgaac tgattcatag cctgcatggc 660
ctgtatggcg cgaaaggcat taccaccaa tataccatca ccagaaaca gaatccgctg 720
attaccaaca ttcgtggcac caacattgaa gaatttctga cctttggcgg caccgatctg 780
aacattatta ccagcgcgca gagcaacgat atctatacca acctgctggc cgattataaa 840
aaaatcgcgt ctaaactgag caaagtgcag gtgagcaatc cgctgctgaa tccgtataaa 900
gatgtgtttg aagcgaaata tggcctggat aaagatgcta gcggcattta tagcgtgaac 960
atcaacaaat tcaacgacat cttcaaaaaa ctgtatagct ttaccgaatt tgatctggcc 1020
accaaatttc aggtgaaatg ccgccagacc tatattggcc agtataaata ttttaaacctg 1080
agcaacctgc tgaacgatag catttacaac atcagcgaag gctataacat caacaacctg 1140

108095P502PC_SeqList_ST25.txt

aaagtgaact	ttcgtggcca	gaacgcgaat	ttaaattccgc	gtattattac	cccgattacc	1200
ggccgtggac	tagtgaaaaa	aattatccgt	ttttgctg	gtggcattat	caccagcaaa	1260
accaaaaagcc	tgggtgccgcg	tggcagcaaa	gcgttaaagt	atttatgcat	cgaaatcaac	1320
aacggcgaac	tgttttttgt	ggcgcgcgaa	aacagctata	acgatgataa	catcaacacc	1380
ccgaaagaaa	ttgatgatac	cgtgaccagc	aataacaact	acgaaaacga	tctggatcag	1440
gtgatttctga	actttaacag	cgaaagcgca	ccgggcctgt	ctgatgaaaa	actgaacctg	1500
accattcaga	acgatgcgta	tatcccga	tatgatagca	acggcaccag	cgatattgaa	1560
cagcatgatg	tgaacgaact	gaacgtgttt	ttttatctgg	atgctgcagaa	agtgccggaa	1620
ggcgaataca	acgtgaatct	gaccagctca	attgataccg	cgctgctgga	acagccgaaa	1680
atctatacct	tttttagcag	cgaattcatc	aacaacgtga	acaaaccggt	gcaggcggcg	1740
ctgtttgtga	gctggattca	gcagggtgctg	gttgatttta	ccaccgaagc	gaaccagaaa	1800
agcaccgtgg	ataaaattgc	ggatattagc	attgtggtgc	cgtatattgg	cctggccctg	1860
aacattggca	acgaagcgca	gaaaggcaac	tttaaagatg	cgctggaact	gctgggtgctg	1920
ggcatttctgc	tgggaatttg	accggaactg	ctgattccga	ccattctggt	gtttaccatc	1980
aaaagctttc	tgggcagcag	cgataacaaa	aacaaagtga	tcaaagcgat	taacaacgcg	2040
ctgaaagaac	gtgatgaaaa	atggaaagaa	gtgtatagct	tcattgtgtc	taactggatg	2100
accaaaatca	acaccagtt	caacaaacgt	aaagaacaaa	tgtatcaggc	gctgcagaac	2160
cagggtgaacg	cgattaaaac	catcatcgaa	agcaaataca	acagctacac	cctggaagaa	2220
aaaaacgaac	tgaccaacaa	atatgacatc	aaacaaatcg	aaaatgaact	gaaccagaaa	2280
gtgagcattg	ccatgaacaa	cattgatcgc	tttctgaccg	aaagcagcat	tagctacctg	2340
atgaaactga	tcaacgaagt	gaaaatcaac	aaactgcgcg	aatatgatga	aaacgtgaaa	2400
acctacctgc	tgaactatat	tattcagcat	ggcagcattc	tgggcgaaaag	ccagcaagaa	2460
ctgaacagca	tggttaccga	taccctgaac	aacagcattc	cgtttaaact	gagcagctac	2520
accgatgata	aaatcctgat	cagctacttc	aacaaattct	tcaaacgcgt	caaaagcagc	2580
agcgtgctga	acatgcgtta	taaaaacgat	aaatacgtag	ataccagcgg	ctatgatagc	2640
aatatcaaca	ttaacggtga	tgtgtataaa	taccgcacca	acaaaaacca	gttcggcatc	2700
tacaacgata	aactgagcga	agtgaacatt	agccagaacg	attatatcat	ctacgataat	2760
aaatataaaa	acttcagcat	cagcttttgg	gtgcgtattc	cgaactacga	taacaaaatc	2820
gtgaacgtga	acaacgaata	caccatcatt	aactgcatgc	gtgataacaa	cagcggctgg	2880
aaagtgcgac	tgaaccataa	cgaaatcatc	tggaccctgc	aggataacgc	cggcattaac	2940
cagaaactgg	cttttaacta	tggcaacgcg	aacggcatta	gcgattacat	caacaaatgg	3000
atctttgtga	ccattaccaa	cgatcgtctg	ggcgatagca	aactgtatat	taacggcaac	3060
ctgatcgacc	agaaaagcat	tctgaacctg	ggcaacattc	atgtgagcga	taacatcctg	3120
ttcaaaattg	tgaactgcag	ctatacccgt	tatattggca	tccgctattt	caacatcttc	3180

108095P502PC_SeqList_ST25.txt

gataaagaac tggatgaaac cgaaattcag accctgtata gcaacgaacc gaacaccaac 3240
atcctgaaag atttctgggg caactatctg ctgtacgata aagaatatta tctgctgaac 3300
gtgctgaaac cgaacaactt tattgatcgc cgtaaagata gcaccctgag cattaacaac 3360
attcgtagca ccattctgct ggccaaccgt ctgtatagcg gcattaaagt gaaaattcag 3420
cgcgtgaaca atagcagcac caacgataac ctgggtgcgta aaaacgatca ggtgtatatc 3480
aactttgtgg ccagcaaaac ccacctgttt ccgctgtatg cggataccgc gaccaccaac 3540
aaagaaaaaa ccattaaaat cagcagcagc ggcaaccgtt ttaaccaggt ggtggtgatg 3600
aacagcgtgg gcaacaactg tacaatgaac ttcaaaaaca acaacggcaa caacattggc 3660
ctgctgggct ttaaagcgga taccgtgggtg gcgagcacct ggtattatac ccacatgcgt 3720
gatcatacca acagcaacgg ctgcttttgg aactttatta gcgaagaaca tggctggcag 3780
gaaaaataa 3789

<210> 6
<211> 1262
<212> PRT
<213> artificial

<220>
<223> modified BONT/E single chain precursor
<400> 6

Met Pro Lys Ile Asn Ser Phe Asn Tyr Asn Asp Pro Val Asn Asp Arg
1 5 10 15
Thr Ile Leu Tyr Ile Lys Pro Gly Gly Cys Gln Glu Phe Tyr Lys Ser
20 25 30
Phe Asn Ile Met Lys Asn Ile Trp Ile Ile Pro Glu Arg Asn Val Ile
35 40 45
Gly Thr Thr Pro Gln Asp Phe His Pro Pro Thr Ser Leu Lys Asn Gly
50 55 60
Asp Ser Ser Tyr Tyr Asp Pro Asn Tyr Leu Gln Ser Asp Glu Glu Lys
65 70 75 80
Asp Arg Phe Leu Lys Ile Val Thr Lys Ile Phe Asn Arg Ile Asn Asn
85 90 95
Asn Leu Ser Gly Gly Ile Leu Leu Glu Glu Leu Ser Lys Ala Asn Pro
100 105 110
Tyr Leu Gly Asn Asp Asn Thr Pro Asp Asn Gln Phe His Ile Gly Asp
115 120 125
Ala Ser Ala Val Glu Ile Lys Phe Ser Asn Gly Ser Gln Asp Ile Leu
130 135 140

108095P502PC_SeqList_ST25.txt

Leu Pro Asn Val Ile Ile Met Gly Ala Glu Pro Asp Leu Phe Glu Thr
 145 150 155 160
 Asn Ser Ser Asn Ile Ser Leu Arg Asn Asn Tyr Met Pro Ser Asn His
 165 170 175
 Gly Phe Gly Ser Ile Ala Ile Val Thr Phe Ser Pro Glu Tyr Ser Phe
 180 185 190
 Arg Phe Asn Asp Asn Ser Met Asn Glu Phe Ile Gln Asp Pro Ala Leu
 195 200 205
 Thr Leu Met His Glu Leu Ile His Ser Leu His Gly Leu Tyr Gly Ala
 210 215 220
 Lys Gly Ile Thr Thr Lys Tyr Thr Ile Thr Gln Lys Gln Asn Pro Leu
 225 230 235 240
 Ile Thr Asn Ile Arg Gly Thr Asn Ile Glu Glu Phe Leu Thr Phe Gly
 245 250 255
 Gly Thr Asp Leu Asn Ile Ile Thr Ser Ala Gln Ser Asn Asp Ile Tyr
 260 265 270
 Thr Asn Leu Leu Ala Asp Tyr Lys Lys Ile Ala Ser Lys Leu Ser Lys
 275 280 285
 Val Gln Val Ser Asn Pro Leu Leu Asn Pro Tyr Lys Asp Val Phe Glu
 290 295 300
 Ala Lys Tyr Gly Leu Asp Lys Asp Ala Ser Gly Ile Tyr Ser Val Asn
 305 310 315 320
 Ile Asn Lys Phe Asn Asp Ile Phe Lys Lys Leu Tyr Ser Phe Thr Glu
 325 330 335
 Phe Asp Leu Ala Thr Lys Phe Gln Val Lys Cys Arg Gln Thr Tyr Ile
 340 345 350
 Gly Gln Tyr Lys Tyr Phe Lys Leu Ser Asn Leu Leu Asn Asp Ser Ile
 355 360 365
 Tyr Asn Ile Ser Glu Gly Tyr Asn Ile Asn Asn Leu Lys Val Asn Phe
 370 375 380
 Arg Gly Gln Asn Ala Asn Leu Asn Pro Arg Ile Ile Thr Pro Ile Thr
 385 390 395 400
 Gly Arg Gly Leu Val Lys Lys Ile Ile Arg Phe Cys Val Arg Gly Ile
 405 410 415

108095P502PC_SeqList_ST25.txt

Ile Thr Ser Lys Thr Lys Ser Leu Val Pro Arg Gly Ser Lys Ala Leu
 420 425 430
 Asn Asp Leu Cys Ile Glu Ile Asn Asn Gly Glu Leu Phe Phe Val Ala
 435 440 445
 Ser Glu Asn Ser Tyr Asn Asp Asp Asn Ile Asn Thr Pro Lys Glu Ile
 450 455 460
 Asp Asp Thr Val Thr Ser Asn Asn Asn Tyr Glu Asn Asp Leu Asp Gln
 465 470 475 480
 Val Ile Leu Asn Phe Asn Ser Glu Ser Ala Pro Gly Leu Ser Asp Glu
 485 490 495
 Lys Leu Asn Leu Thr Ile Gln Asn Asp Ala Tyr Ile Pro Lys Tyr Asp
 500 505 510
 Ser Asn Gly Thr Ser Asp Ile Glu Gln His Asp Val Asn Glu Leu Asn
 515 520 525
 Val Phe Phe Tyr Leu Asp Ala Gln Lys Val Pro Glu Gly Glu Asn Asn
 530 535 540
 Val Asn Leu Thr Ser Ser Ile Asp Thr Ala Leu Leu Glu Gln Pro Lys
 545 550 555 560
 Ile Tyr Thr Phe Phe Ser Ser Glu Phe Ile Asn Asn Val Asn Lys Pro
 565 570 575
 Val Gln Ala Ala Leu Phe Val Ser Trp Ile Gln Gln Val Leu Val Asp
 580 585 590
 Phe Thr Thr Glu Ala Asn Gln Lys Ser Thr Val Asp Lys Ile Ala Asp
 595 600 605
 Ile Ser Ile Val Val Pro Tyr Ile Gly Leu Ala Leu Asn Ile Gly Asn
 610 615 620
 Glu Ala Gln Lys Gly Asn Phe Lys Asp Ala Leu Glu Leu Leu Gly Ala
 625 630 635 640
 Gly Ile Leu Leu Glu Phe Glu Pro Glu Leu Leu Ile Pro Thr Ile Leu
 645 650 655
 Val Phe Thr Ile Lys Ser Phe Leu Gly Ser Ser Asp Asn Lys Asn Lys
 660 665 670
 Val Ile Lys Ala Ile Asn Asn Ala Leu Lys Glu Arg Asp Glu Lys Trp
 675 680 685

108095P502PC_SeqList_ST25.txt

Lys Glu Val Tyr Ser Phe Ile Val Ser Asn Trp Met Thr Lys Ile Asn
 690 695 700
 Thr Gln Phe Asn Lys Arg Lys Glu Gln Met Tyr Gln Ala Leu Gln Asn
 705 710 715 720
 Gln Val Asn Ala Ile Lys Thr Ile Ile Glu Ser Lys Tyr Asn Ser Tyr
 725 730 735
 Thr Leu Glu Glu Lys Asn Glu Leu Thr Asn Lys Tyr Asp Ile Lys Gln
 740 745 750
 Ile Glu Asn Glu Leu Asn Gln Lys Val Ser Ile Ala Met Asn Asn Ile
 755 760 765
 Asp Arg Phe Leu Thr Glu Ser Ser Ile Ser Tyr Leu Met Lys Leu Ile
 770 775 780
 Asn Glu Val Lys Ile Asn Lys Leu Arg Glu Tyr Asp Glu Asn Val Lys
 785 790 795 800
 Thr Tyr Leu Leu Asn Tyr Ile Ile Gln His Gly Ser Ile Leu Gly Glu
 805 810 815
 Ser Gln Gln Glu Leu Asn Ser Met Val Thr Asp Thr Leu Asn Asn Ser
 820 825 830
 Ile Pro Phe Lys Leu Ser Ser Tyr Thr Asp Asp Lys Ile Leu Ile Ser
 835 840 845
 Tyr Phe Asn Lys Phe Phe Lys Arg Ile Lys Ser Ser Ser Val Leu Asn
 850 855 860
 Met Arg Tyr Lys Asn Asp Lys Tyr Val Asp Thr Ser Gly Tyr Asp Ser
 865 870 875 880
 Asn Ile Asn Ile Asn Gly Asp Val Tyr Lys Tyr Pro Thr Asn Lys Asn
 885 890 895
 Gln Phe Gly Ile Tyr Asn Asp Lys Leu Ser Glu Val Asn Ile Ser Gln
 900 905 910
 Asn Asp Tyr Ile Ile Tyr Asp Asn Lys Tyr Lys Asn Phe Ser Ile Ser
 915 920 925
 Phe Trp Val Arg Ile Pro Asn Tyr Asp Asn Lys Ile Val Asn Val Asn
 930 935 940
 Asn Glu Tyr Thr Ile Ile Asn Cys Met Arg Asp Asn Asn Ser Gly Trp
 945 950 955 960

108095P502PC_SeqList_ST25.txt

Lys Val Ser Leu Asn His Asn Glu Ile Ile Trp Thr Leu Gln Asp Asn
 965 970 975
 Ala Gly Ile Asn Gln Lys Leu Ala Phe Asn Tyr Gly Asn Ala Asn Gly
 980 985 990
 Ile Ser Asp Tyr Ile Asn Lys Trp Ile Phe Val Thr Ile Thr Asn Asp
 995 1000 1005
 Arg Leu Gly Asp Ser Lys Leu Tyr Ile Asn Gly Asn Leu Ile Asp
 1010 1015 1020
 Gln Lys Ser Ile Leu Asn Leu Gly Asn Ile His Val Ser Asp Asn
 1025 1030 1035
 Ile Leu Phe Lys Ile Val Asn Cys Ser Tyr Thr Arg Tyr Ile Gly
 1040 1045 1050
 Ile Arg Tyr Phe Asn Ile Phe Asp Lys Glu Leu Asp Glu Thr Glu
 1055 1060 1065
 Ile Gln Thr Leu Tyr Ser Asn Glu Pro Asn Thr Asn Ile Leu Lys
 1070 1075 1080
 Asp Phe Trp Gly Asn Tyr Leu Leu Tyr Asp Lys Glu Tyr Tyr Leu
 1085 1090 1095
 Leu Asn Val Leu Lys Pro Asn Asn Phe Ile Asp Arg Arg Lys Asp
 1100 1105 1110
 Ser Thr Leu Ser Ile Asn Asn Ile Arg Ser Thr Ile Leu Leu Ala
 1115 1120 1125
 Asn Arg Leu Tyr Ser Gly Ile Lys Val Lys Ile Gln Arg Val Asn
 1130 1135 1140
 Asn Ser Ser Thr Asn Asp Asn Leu Val Arg Lys Asn Asp Gln Val
 1145 1150 1155
 Tyr Ile Asn Phe Val Ala Ser Lys Thr His Leu Phe Pro Leu Tyr
 1160 1165 1170
 Ala Asp Thr Ala Thr Thr Asn Lys Glu Lys Thr Ile Lys Ile Ser
 1175 1180 1185
 Ser Ser Gly Asn Arg Phe Asn Gln Val Val Val Met Asn Ser Val
 1190 1195 1200
 Gly Asn Asn Cys Thr Met Asn Phe Lys Asn Asn Asn Gly Asn Asn
 1205 1210 1215

108095P502PC_SeqList_ST25.txt

Ile Gly Leu Leu Gly Phe Lys Ala Asp Thr Val Val Ala Ser Thr
1220 1225 1230

Trp Tyr Tyr Thr His Met Arg Asp His Thr Asn Ser Asn Gly Cys
1235 1240 1245

Phe Trp Asn Phe Ile Ser Glu Glu His Gly Trp Gln Glu Lys
1250 1255 1260

<210> 7
<211> 4
<212> PRT
<213> Homo sapiens

<400> 7

Pro Arg Gly Ser
1

<210> 8
<211> 6
<212> PRT
<213> Homo sapiens

<400> 8

Leu Val Pro Arg Gly Ser
1 5

<210> 9
<211> 8
<212> PRT
<213> Homo sapiens

<400> 9

Lys Ser Leu Val Pro Arg Gly Ser
1 5

<210> 10
<211> 9
<212> PRT
<213> Homo sapiens

<400> 10

Asn Lys Ser Leu Val Pro Arg Gly Ser
1 5

<210> 11
<211> 10
<212> PRT
<213> Homo sapiens

<400> 11

Glu Asn Lys Ser Leu Val Pro Arg Gly Ser
1 5 10

108095P502PC_SeqList_ST25.txt

<210> 12
<211> 8
<212> PRT
<213> Homo sapiens

<400> 12

Thr Ser Leu Val Pro Arg Gly Ser
1 5

<210> 13
<211> 8
<212> PRT
<213> Homo sapiens

<400> 13

Gly Gly Leu Val Pro Arg Gly Ser
1 5