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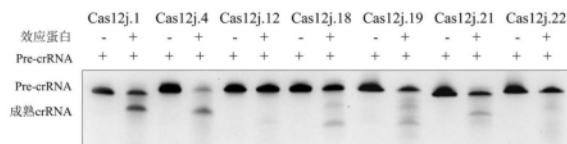
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序列表165页 附图4页

(54) 发明名称

CRISPR-Cas12j酶和系统

(57) 摘要

本发明提供了CRISPR-Cas12j酶和系统。具体而言,本发明提供一种Cas效应蛋白,包含此类蛋白的融合蛋白,以及其编码核酸分子。还提供用于核酸编辑的复合物和组合物,例如基因或基因组编辑的复合物和组合物,其包含Cas效应蛋白或融合蛋白,或其编码核酸分子。还提供用于核酸编辑的方法,例如基因或基因组编辑的方法,其使用Cas效应蛋白或融合蛋白。



1. 一种CRISPR/Cas系统中的效应蛋白,其由SEQ ID NO:2所示的序列组成。
2. 一种缀合物,其包含权利要求1所述的蛋白以及修饰部分;其中,所述修饰部分选自另外的蛋白或多肽、可检测的标记,及其任意组合。
3. 权利要求2所述的缀合物,其中,所述修饰部分任选地通过接头连接至所述蛋白的N端或C端。
4. 权利要求2所述的缀合物,其中,所述另外的蛋白或多肽选自表位标签、报告基因序列、核定位信号(NLS)序列、靶向部分、转录激活结构域、转录抑制结构域、核酸酶结构域、核苷酸脱氨酶结构域、甲基化酶结构域、去甲基化酶结构域、转录释放因子活性结构域、组蛋白修饰活性结构域和核酸结合活性结构域。
5. 权利要求2所述的缀合物,其中,所述另外的蛋白或多肽选自具有单链RNA切割活性的结构域、具有双链RNA切割活性的结构域、具有单链DNA切割活性的结构域、或具有双链DNA切割活性的结构域。
6. 权利要求2所述的缀合物,其中,所述缀合物包含表位标签。
7. 权利要求2所述的缀合物,其中,所述缀合物包含NLS序列。
8. 权利要求7所述的缀合物,其中,所述NLS序列如SEQ ID NO:81所示。
9. 权利要求7所述的缀合物,其中,所述NLS序列位于所述蛋白的N端或C端。
10. 一种融合蛋白,其包含权利要求1所述的蛋白以及另外的蛋白或多肽。
11. 权利要求10所述的融合蛋白,其中,所述另外的蛋白或多肽任选地通过接头连接至所述蛋白的N端或C端。
12. 权利要求10所述的融合蛋白,其中,所述另外的蛋白或多肽选自表位标签、报告基因序列、核定位信号(NLS)序列、靶向部分、转录激活结构域、转录抑制结构域、核酸酶结构域、核苷酸脱氨酶结构域、甲基化酶结构域、去甲基化酶结构域、转录释放因子活性结构域、组蛋白修饰活性结构域和核酸结合活性结构域。
13. 权利要求10所述的融合蛋白,其中,所述另外的蛋白或多肽选自具有单链RNA切割活性的结构域、具有双链RNA切割活性的结构域、具有单链DNA切割活性的结构域、或具有双链DNA切割活性的结构域。
14. 权利要求10所述的融合蛋白,其中,所述融合蛋白包含表位标签。
15. 权利要求10所述的融合蛋白,其中,所述融合蛋白包含NLS序列。
16. 权利要求15所述的融合蛋白,其中,所述NLS序列如SEQ ID NO:81所示。
17. 权利要求15所述的融合蛋白,其中,所述NLS序列位于所述蛋白的N端或C端。
18. 权利要求10所述的融合蛋白,其中,所述融合蛋白的氨基酸序列如SEQ ID NO:83所示。
19. 一种复合物,其包含:
 - (i) 蛋白组分,其选自:权利要求1所述的蛋白、权利要求2-9任一项所述的缀合物、权利要求10-18任一项所述的融合蛋白,及其任意组合;和
 - (ii) 核酸组分,其从5'至3'方向包含同向重复序列和能够与靶序列杂交的导向序列,所述同向重复序列选自SEQ ID NO:42所示的序列或其互补序列;
其中,所述蛋白组分与核酸组分相互结合形成复合物。
20. 权利要求19所述的复合物,其中,所述导向序列连接于所述核酸分子的3'端。

21. 权利要求19所述的复合物,其中,所述导向序列包含所述靶序列的互补序列。
22. 权利要求19所述的复合物,其中,所述核酸组分是CRISPR/Cas系统中的导向RNA。
23. 权利要求19所述的复合物,其中,所述复合物不包含反式作用crRNA(tracrRNA)。
24. 一种分离的核酸分子,其包含:
- (i) 编码权利要求1所述的蛋白、或权利要求2-9任一项所述的缀合物、或权利要求10-18任一项所述的融合蛋白的核苷酸序列;或,
- (ii) (i)所述的核苷酸序列以及编码同向重复序列的核苷酸序列,所述同向重复序列选自SEQ ID NO:42所示的序列或其互补序列。
25. 权利要求24所述的分离的核酸分子,其中,(i)或(ii)所述的核苷酸序列经密码子优化用于在原核细胞或真核细胞中进行表达。
26. 一种载体,其包含权利要求24或25所述的分离的核酸分子。
27. 一种宿主细胞,其包含权利要求24或25所述的分离的核酸分子或权利要求26所述的载体。
28. 一种组合物,其包含:
- (i) 第一组分,其选自:权利要求1所述的蛋白、权利要求2-9任一项所述的缀合物、权利要求10-18任一项所述的融合蛋白、编码所述蛋白或融合蛋白的核苷酸序列,以及其任意组合;和
- (ii) 第二组分,其为包含导向RNA的核苷酸序列,或者编码所述包含导向RNA的核苷酸序列的核苷酸序列;
- 其中,所述导向RNA从5'至3'方向包含同向重复序列和导向序列,所述导向序列能够与靶序列杂交;所述同向重复序列选自SEQ ID NO:42所示的序列或其互补序列;
- 所述导向RNA能够与(i)中所述的蛋白、缀合物或融合蛋白形成复合物。
29. 权利要求28所述的组合物,其中,所述导向序列连接至所述同向重复序列的3'端。
30. 权利要求28所述的组合物,其中,所述导向序列包含所述靶序列的互补序列。
31. 权利要求28所述的组合物,其中,所述组合物不包含反式作用crRNA(tracrRNA)。
32. 权利要求28所述的组合物,其中,所述组合物中的至少一个组分是非天然存在的或经修饰的。
33. 权利要求28所述的组合物,其中:当所述靶序列为DNA时,所述靶序列位于原间隔序列临近基序(PAM)的3'端,并且所述PAM具有5'-TTN所示的序列。
34. 一种组合物,其包含一种或多种载体,所述一种或多种载体包含:
- (i) 第一核酸,其为编码权利要求1所述的蛋白或权利要求10-18任一项所述的融合蛋白的核苷酸序列;任选地所述第一核酸可操作地连接至第一调节元件;以及
- (ii) 第二核酸,其编码包含导向RNA的核苷酸序列;任选地所述第二核酸可操作地连接至第二调节元件;
- 其中:
- 所述第一核酸与第二核酸存在于相同或不同的载体上;
- 所述导向RNA从5'至3'方向包含同向重复序列和导向序列,所述导向序列能够与靶序列杂交;所述同向重复序列选自SEQ ID NO:42所示的序列或其互补序列;
- 所述导向RNA能够与(i)中所述的效应蛋白或融合蛋白形成复合物。

35. 权利要求34所述的组合物,其中,所述导向序列连接至所述同向重复序列的3'端。
36. 权利要求34所述的组合物,其中,所述导向序列包含所述靶序列的互补序列。
37. 权利要求34所述的组合物,其中,所述组合物不包含反式作用crRNA(tracrRNA)。
38. 权利要求34所述的组合物,其中,所述组合物中的至少一个组分是非天然存在的或经修饰的。
39. 权利要求34所述的组合物,其中,所述第一调节元件是启动子。
40. 权利要求34所述的组合物,其中,所述第二调节元件是启动子。
41. 权利要求34所述的组合物,其中:当所述靶序列为DNA时,所述靶序列位于原间隔序列临近基序(PAM)的3'端,并且所述PAM具有5'-TTN所示的序列。
42. 权利要求34-41任一项所述的组合物,其中,当所述靶序列为RNA时,所述靶RNA序列不具有PAM结构域限制。
43. 权利要求34-41任一项所述的组合物,其中,所述靶序列是来自原核细胞或真核细胞的DNA或RNA序列;或者,所述靶序列是非天然存在的DNA或RNA序列。
44. 权利要求34-41任一项所述的组合物,其中,所述靶序列存在于细胞内。
45. 权利要求44所述的组合物,其中,所述靶序列存在于细胞核内、细胞质或细胞器内。
46. 权利要求44所述的组合物,其中,所述细胞是真核细胞或原核细胞。
47. 权利要求34-41任一项所述的组合物,其中,所述蛋白连接有一个或多个NLS序列,或者,所述缀合物或融合蛋白包含一个或多个NLS序列。
48. 权利要求47所述的组合物,其中,所述NLS序列连接至所述蛋白的N端或C端。
49. 一种试剂盒,其包括一种或多种选自下列的组分:权利要求1所述的蛋白、权利要求2-9任一项所述的缀合物、权利要求10-18任一项所述的融合蛋白、权利要求19-23任一项所述的复合物、权利要求24或25所述的分离的核酸分子、权利要求26所述的载体、权利要求28-48任一项所述的组合物。
50. 一种递送组合物,其包含递送载体,以及选自下列的一种或多种:权利要求1所述的蛋白、权利要求2-9任一项所述的缀合物、权利要求10-18任一项所述的融合蛋白、权利要求19-23任一项所述的复合物、权利要求24或25所述的分离的核酸分子、权利要求26所述的载体、权利要求28-48任一项所述的组合物。
51. 权利要求50所述的递送组合物,其中,所述递送载体选自脂质颗粒、糖颗粒、金属颗粒、蛋白颗粒、外泌体、微泡、基因枪或病毒载体。
52. 权利要求50所述的递送组合物,其中,所述递送载体选自脂质体。
53. 权利要求51所述的递送组合物,其中,所述病毒载体为复制缺陷型逆转录病毒、慢病毒、腺病毒或腺相关病毒。
54. 一种用于非治疗目的的修饰靶基因的方法,其包括:将权利要求19-23任一项所述的复合物或权利要求28-48任一项所述的组合物与所述靶基因接触,或者递送至包含所述靶基因的细胞中;所述靶序列存在于所述靶基因中。
55. 权利要求54所述的方法,其中,所述靶基因存在于细胞内。
56. 权利要求55所述的方法,其中,所述细胞是原核细胞或真核细胞。
57. 权利要求54所述的方法,其中,所述细胞选自哺乳动物细胞或植物细胞。
58. 权利要求54所述的方法,其中,所述靶基因存在于体外的核酸分子中。

59. 权利要求58所述的方法,其中,所述体外的核酸分子为质粒。
60. 权利要求54所述的方法,其中,所述修饰是指所述靶序列的断裂。
61. 权利要求60所述的方法,其中,所述修饰包括DNA的双链断裂或RNA的单链断裂。
62. 权利要求60所述的方法,其中,所述修饰还包括将外源核酸插入所述断裂中。
63. 一种用于非治疗目的的改变基因产物的表达的方法,其包括:将权利要求19-23任一项所述的复合物或权利要求28-48任一项所述的组合物与编码所述基因产物的核酸分子接触,或者递送至包含所述核酸分子的细胞中,所述靶序列存在于所述核酸分子中。
64. 权利要求63所述的方法,其中,所述核酸分子存在于细胞内。
65. 权利要求64所述的方法,其中,所述细胞是原核细胞或真核细胞。
66. 权利要求64所述的方法,其中,所述细胞选自哺乳动物细胞或植物细胞。
67. 权利要求63所述的方法,其中,所述核酸分子存在于体外的核酸分子中。
68. 权利要求67所述的方法,其中,所述体外的核酸分子为质粒。
69. 权利要求63所述的方法,其中,所述基因产物的表达被改变增强或降低。
70. 权利要求63所述的方法,其中,所述基因产物是蛋白。
71. 权利要求54-70任一项所述的方法,其中所述的蛋白、缀合物、融合蛋白、分离的核酸分子、复合物、载体或组合物包含于递送载体中。
72. 权利要求71所述的方法,其中,所述递送载体选自脂质颗粒、糖颗粒、金属颗粒、蛋白颗粒、外泌体、病毒载体。
73. 权利要求71所述的方法,其中,所述递送载体选自脂质体。
74. 权利要求72所述的方法,其中,所述病毒载体选自复制缺陷型逆转录病毒、慢病毒、腺病毒或腺相关病毒。
75. 权利要求54-70任一项所述的方法,其用于改变靶基因或编码靶基因产物的核酸分子中的一个或多个靶序列来修饰细胞、细胞系或非人生物体。
76. 一种体外的细胞或细胞系或它们的子代,所述细胞或细胞系或它们的子代包含:权利要求1所述的蛋白、权利要求2-9任一项所述的缀合物、权利要求10-18任一项所述的融合蛋白、权利要求19-23所述的复合物、权利要求24或25所述的分离的核酸分子、权利要求26所述的载体、权利要求28-48任一项所述的组合物。
77. 权利要求76所述的细胞或细胞系或它们的子代,其中,所述细胞是真核细胞。
78. 权利要求76所述的细胞或细胞系或它们的子代,其中,所述细胞是动物细胞或植物细胞。
79. 权利要求1所述的蛋白、权利要求2-9任一项所述的缀合物、权利要求10-18任一项所述的融合蛋白、权利要求19-23任一项所述的复合物、权利要求24或25所述的分离的核酸分子、权利要求26所述的载体、权利要求28-48任一项所述的组合物、权利要求49所述的试剂盒或权利要求50所述的递送组合物,用于非诊断治疗目的的核酸编辑的用途。
80. 权利要求79所述的用途,其中,所述核酸编辑包括基因或基因组编辑。
81. 权利要求80所述的用途,其中,所述基因或基因组编辑包括修饰基因、敲除基因、改变基因产物的表达、修复突变、和/或插入多核苷酸。
82. 权利要求1所述的蛋白、权利要求2-9任一项所述的缀合物、权利要求10-18任一项所述的融合蛋白、权利要求19-23任一项所述的复合物、权利要求24或25所述的分离的核酸

分子、权利要求26所述的载体、权利要求28-48任一项所述的组合物、权利要求49所述的试剂盒或权利要求50所述的递送组合物,在制备制剂中的用途,所述制剂用于:

- (i) 离体基因或基因组编辑;
- (ii) 离体单链DNA的检测;
- (iii) 编辑靶基因座中的靶序列来修饰非人类生物;
- (iv) 治疗由靶基因座中的靶序列的缺陷引起的病症。

CRISPR-Cas12j酶和系统

[0001] 本申请是申请号为201980014005.0、申请日为2019年11月15日、发明名称为“CRISPR-Cas12j酶和系统”的发明申请的分案申请。

技术领域

[0002] 本发明涉及核酸编辑领域,特别是规律成簇的间隔短回文重复(CRISPR)技术领域。具体而言,本发明涉及Cas效应蛋白,包含此类蛋白的融合蛋白,以及编码它们的核酸分子。本发明还涉及用于核酸编辑(例如,基因或基因组编辑)的复合物和组合物,其包含本发明的蛋白或融合蛋白,或编码它们的核酸分子。本发明还涉及用于核酸编辑(例如,基因或基因组编辑)的方法,其使用包含本发明的蛋白或融合蛋白。

背景技术

[0003] CRISPR/Cas技术是一种被广泛使用的基因编辑技术,它通过RNA引导对基因组上的靶序列进行特异性结合并切割DNA产生双链断裂,利用生物非同源末端连接或同源重组进行定点基因编辑。

[0004] CRISPR/Cas9系统是最常用的II型CRISPR系统,它识别3'-NGG的PAM基序,对靶标序列进行平末端切割。CRISPR/Cas Type V系统是一类近两年新发现的CRISPR系统,它具有5'-TTN的基序,对靶标序列进行粘性末端切割,例如Cpf1,C2c1,CasX,CasY。然而目前存在的不同的CRISPR/Cas各有不同的优点和缺陷。例如Cas9,C2c1和CasX均需要两条RNA进行导向RNA,而Cpf1只需要一条导向RNA而且可以用来进行多重基因编辑。CasX具有980个氨基酸的大小,而常见的Cas9,C2c1,CasY和Cpf1通常大小在1300个氨基酸左右。此外,Cas9,Cpf1,CasX,CasY的PAM序列都比较复杂多样,而C2c1识别严谨的5'-TTN,因此它的靶标位点比其他系统容易被预测从而降低了潜在的脱靶效应。

[0005] 总之,鉴于目前可获得的CRISPR/Cas系统都受限于一些缺陷,开发一种更稳健的、具有多方面良好性能的新型CRISPR/Cas系统对生物技术的发展具有重要意义。

发明内容

[0006] 本申请的发明人经过大量实验和反复摸索,出人意料地发现了一种新型RNA指导的核酸内切酶。基于这一发现,本发明人开发了新的CRISPR/Cas系统以及基于该系统的基因编辑方法。

[0007] Cas效应蛋白

[0008] 因此,在第一方面,本发明提供了多种蛋白,其具有SEQ ID NOs:1-20、107、108任一项所示的氨基酸序列或其直系同源物、同源物、变体或功能性片段;其中,所述直系同源物、同源物、变体或功能性片段基本保留了其所源自的序列的生物学功能。

[0009] 在本发明中,上述序列的生物学功能包括但不限于,与导向RNA结合的活性、核酸内切酶活性、在导向RNA引导下与靶序列特定位点结合并切割的活性。

[0010] 在某些实施方案中,所述直系同源物、同源物、变体与其所源自的序列相比具有至

少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性。

[0011] 在某些实施方案中,所述直系同源物、同源物、变体与SEQ ID NOs:1-20、107、108任一项所示的序列相比具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性,并且基本保留了其所源自的序列的生物学功能(例如,与导向RNA结合的活性、核酸内切酶活性、在导向RNA引导下与靶序列特定位点结合并切割的活性)。

[0012] 在某些实施方案中,所述蛋白是CRISPR/Cas系统中的效应蛋白。

[0013] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0014] (i) SEQ ID NOs:1-20、107、108任一项所示的序列;

[0015] (ii) 与SEQ ID NOs:1-20、107、108任一项所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;或

[0016] (iii) 与SEQ ID NOs:1-20、107、108任一项所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0017] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0018] (i) SEQ ID NO:1所示的序列;

[0019] (ii) 与SEQ ID NO:1所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;或

[0020] (iii) 与SEQ ID NO:1所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0021] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:2所示的氨基酸序列。

[0022] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0023] (i) SEQ ID NO:2所示的序列;

[0024] (ii) 与SEQ ID NO:2所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;或

[0025] (iii) 与SEQ ID NO:2所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0026] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:2所示的氨基酸序列。

[0027] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0028] (i) SEQ ID NO:3所示的序列;

[0029] (ii) 与SEQ ID NO:3所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;
或

[0030] (iii) 与SEQ ID NO:3所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0031] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:3所示的氨基酸序列。

[0032] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0033] (i) SEQ ID NO:4所示的序列;

[0034] (ii) 与SEQ ID NO:4所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;
或

[0035] (iii) 与SEQ ID NO:4所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0036] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:4所示的氨基酸序列。

[0037] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0038] (i) SEQ ID NO:5所示的序列;

[0039] (ii) 与SEQ ID NO:5所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;
或

[0040] (iii) 与SEQ ID NO:5所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0041] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:5所示的氨基酸序列。

[0042] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0043] (i) SEQ ID NO:6所示的序列;

[0044] (ii) 与SEQ ID NO:6所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;
或

[0045] (iii) 与SEQ ID NO:6所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0046] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:6所示的氨基酸序列。

[0047] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组

成：

[0048] (i) SEQ ID NO:7所示的序列；

[0049] (ii) 与SEQ ID NO:7所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列；
或

[0050] (iii) 与SEQ ID NO:7所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0051] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:7所示的氨基酸序列。

[0052] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成：

[0053] (i) SEQ ID NO:8所示的序列；

[0054] (ii) 与SEQ ID NO:8所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列；
或

[0055] (iii) 与SEQ ID NO:8所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0056] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:8所示的氨基酸序列。

[0057] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成：

[0058] (i) SEQ ID NO:9所示的序列；

[0059] (ii) 与SEQ ID NO:9所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列；
或

[0060] (iii) 与SEQ ID NO:9所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0061] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:9所示的氨基酸序列。

[0062] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成：

[0063] (i) SEQ ID NO:10所示的序列；

[0064] (ii) 与SEQ ID NO:10所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列；
或

[0065] (iii) 与SEQ ID NO:10所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0066] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:10所示的氨基酸序列。

[0067] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0068] (i) SEQ ID NO:11所示的序列;

[0069] (ii) 与SEQ ID NO:11所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;或

[0070] (iii) 与SEQ ID NO:11所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0071] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:11所示的氨基酸序列。

[0072] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0073] (i) SEQ ID NO:12所示的序列;

[0074] (ii) 与SEQ ID NO:12所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;或

[0075] (iii) 与SEQ ID NO:12所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0076] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:12所示的氨基酸序列。

[0077] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0078] (i) SEQ ID NO:13所示的序列;

[0079] (ii) 与SEQ ID NO:13所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;或

[0080] (iii) 与SEQ ID NO:13所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0081] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:13所示的氨基酸序列。

[0082] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0083] (i) SEQ ID NO:14所示的序列;

[0084] (ii) 与SEQ ID NO:14所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;或

[0085] (iii) 与SEQ ID NO:14所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

- [0086] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:14所示的氨基酸序列。
- [0087] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:
- [0088] (i)SEQ ID NO:15所示的序列;
- [0089] (ii)与SEQ ID NO:15所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;
- 或
- [0090] (iii)与SEQ ID NO:15所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。
- [0091] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:15所示的氨基酸序列。
- [0092] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:
- [0093] (i)SEQ ID NO:16所示的序列;
- [0094] (ii)与SEQ ID NO:16所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;
- 或
- [0095] (iii)与SEQ ID NO:16所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。
- [0096] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:16所示的氨基酸序列。
- [0097] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:
- [0098] (i)SEQ ID NO:17所示的序列;
- [0099] (ii)与SEQ ID NO:17所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;
- 或
- [0100] (iii)与SEQ ID NO:17所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。
- [0101] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:17所示的氨基酸序列。
- [0102] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:
- [0103] (i)SEQ ID NO:18所示的序列;
- [0104] (ii)与SEQ ID NO:18所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;
- 或
- [0105] (iii)与SEQ ID NO:18所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序

列同一性的序列。

[0106] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:18所示的氨基酸序列。

[0107] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0108] (i)SEQ ID NO:19所示的序列;

[0109] (ii)与SEQ ID NO:19所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;或

[0110] (iii)与SEQ ID NO:19所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0111] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:19所示的氨基酸序列。

[0112] 在某些实施方案中,本发明的蛋白包含选自下列的序列,或由选自下列的序列组成:

[0113] (i)SEQ ID NO:20所示的序列;

[0114] (ii)与SEQ ID NO:20所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;或

[0115] (iii)与SEQ ID NO:20所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0116] 在某些实施方案中,本发明的蛋白具有SEQ ID NO:20所示的氨基酸序列。

[0117] 衍生的蛋白

[0118] 本发明的蛋白可进行衍生化,例如被连接至另一个分子(例如另一个多肽或蛋白)。通常,蛋白的衍生化(例如,标记)不会不利影响该蛋白的期望活性(例如,与导向RNA结合的活性、核酸内切酶活性、在导向RNA引导下与靶序列特定位点结合并切割的活性)。因此,本发明的蛋白还意欲包括此类衍生化的形式。例如,可以将本发明的蛋白功能性连接(通过化学偶合、基因融合、非共价连接或其它方式)于一个或多个其它分子基团,例如另一个蛋白或多肽,检测试剂,药用试剂等。

[0119] 特别地,可以将本发明的蛋白连接其他功能性单元。例如,可以将其与核定位信号(NLS)序列连接,以提高本发明的蛋白进入细胞核的能力。例如,可以将其与靶向部分连接,以使得本发明的蛋白具有靶向性。例如,可以将其与可检测的标记连接,以便于对本发明的蛋白进行检测。例如,可以将其与表位标签连接,以便于本发明的蛋白的表达、检测、示踪和/或纯化。

[0120] 缀合物

[0121] 因此,在第二方面,本发明提供了一种缀合物,其包含如上所述的蛋白和修饰部分。

[0122] 在某些实施方案中,所述修饰部分选自另外的蛋白或多肽、可检测的标记或其任意组合。

[0123] 在某些实施方案中,所述另外的蛋白或多肽选自表位标签、报告基因序列、核定位信号(NLS)序列、靶向部分、转录激活结构域(例如,VP64)、转录抑制结构域(例如,KRAB结构域或SID结构域)、核酸酶结构域(例如,Fok1),具有选自下列的活性的结构域:核苷酸脱氨酶,甲基化酶活性,去甲基化酶,转录激活活性,转录抑制活性,转录释放因子活性,组蛋白修饰活性,核酸酶活性,单链RNA切割活性,双链RNA切割活性,单链DNA切割活性,双链DNA切割活性和核酸结合活性;以及其任意组合。

[0124] 在某些实施方案中,本发明的缀合物包含一个或多个NLS序列,例如SV40病毒大T抗原的NLS。在某些示例性实施方案中,所述NLS序列如SEQ ID NO:81所示。在某些实施方案中,所述NLS序列位于、靠近或接近本发明的蛋白的末端(例如,N端或C端)。在某些示例性实施方案中,所述NLS序列位于、靠近或接近本发明的蛋白的C端。

[0125] 在某些实施方案中,本发明的缀合物包含表位标签(epitope tag)。这类表位标签是本领域技术人员熟知的,其实例包括但不限于His、V5、FLAG、HA、Myc、VSV-G、Trx等,并且本领域技术人员已知如何根据期望目的(例如,纯化、检测或示踪)选择合适的表位标签。

[0126] 在某些实施方案中,本发明的缀合物包含报告基因序列。这类报告基因是本领域技术人员熟知的,其实例包括但不限于GST、HRP、CAT、GFP、HcRed、DsRed、CFP、YFP、BFP等。

[0127] 在某些实施方案中,本发明的缀合物包含能够与DNA分子或细胞内分子结合的结构域,例如麦芽糖结合蛋白(MBP)、Lex A的DNA结合结构域(DBD)、GAL4的DBD等。

[0128] 在某些实施方案中,本发明的缀合物包含可检测的标记,例如荧光染料,例如FITC或DAPI。

[0129] 在某些实施方案中,本发明的蛋白任选地通过接头与所述修饰部分偶联、缀合或融合。

[0130] 在某些实施方案中,所述修饰部分直接连接至本发明的蛋白的N端或C端。

[0131] 在某些实施方案中,所述修饰部分通过接头连接至本发明的蛋白的N端或C端。这类接头是本领域熟知的,其实例包括但不限于包含一个或多个(例如,1个,2个,3个,4个或5个)氨基酸(如,Glu或Ser)或氨基酸衍生物(如,Ahx、 β -Ala、GABA或Ava)的接头,或PEG等。

[0132] 融合蛋白

[0133] 在第三方面,本发明提供了一种融合蛋白,其包含本发明的蛋白以及另外的蛋白或多肽。

[0134] 在某些实施方案中,所述另外的蛋白或多肽选自表位标签、报告基因序列、核定位信号(NLS)序列、靶向部分、转录激活结构域(例如,VP64)、转录抑制结构域(例如,KRAB结构域或SID结构域)、核酸酶结构域(例如,Fok1),具有选自下列的活性的结构域:核苷酸脱氨酶,甲基化酶活性,去甲基化酶,转录激活活性,转录抑制活性,转录释放因子活性,组蛋白修饰活性,核酸酶活性,单链RNA切割活性,双链RNA切割活性,单链DNA切割活性,双链DNA切割活性和核酸结合活性;以及其任意组合。

[0135] 在某些实施方案中,本发明的融合蛋白包含一个或多个NLS序列,例如SV40病毒大T抗原的NLS。在某些实施方案中,所述NLS序列位于、靠近或接近本发明的蛋白的末端(例如,N端或C端)。在某些示例性实施方案中,所述NLS序列位于、靠近或接近本发明的蛋白的C端。

[0136] 在某些实施方案中,本发明的融合蛋白包含表位标签。

- [0137] 在某些实施方案中,本发明的融合蛋白包含报告基因序列。
- [0138] 在某些实施方案中,本发明的融合蛋白包含能够与DNA分子或细胞内分子结合的结构域。
- [0139] 在某些实施方案中,本发明的蛋白任选地通过接头与所述另外的蛋白或多肽融合。
- [0140] 在某些实施方案中,所述另外的蛋白或多肽直接连接至本发明的蛋白的N端或C端。
- [0141] 在某些实施方案中,所述另外的蛋白或多肽通过接头连接至本发明的蛋白的N端或C端。
- [0142] 在某些示例性实施方案中,本发明的融合蛋白具有选自下列的氨基酸序列:SEQ ID NOs:82-101。
- [0143] 本发明的蛋白、本发明的缀合物或本发明的融合蛋白不受其产生方式的限定,例如,其可以通过基因工程方法(重组技术)产生,也可以通过化学合成方法产生。
- [0144] 同向重复序列
- [0145] 在第四方面,本发明提供了一种分离的核酸分子,其包含选自下列的序列,或由选自下列的序列组成:
- [0146] (i)SEQ ID NOs:41-60任一项所示的序列;
- [0147] (ii)与SEQ ID NOs:41-60任一项所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;
- [0148] (iii)与SEQ ID NOs:41-60任一项所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;
- [0149] (iv)在严格条件下与(i)-(iii)任一项中所述的序列杂交的序列;或
- [0150] (v)(i)-(iii)任一项中所述的序列的互补序列;
- [0151] 并且,(ii)-(v)中任一项所述的序列基本保留了其所源自的序列的生物学功能,所述序列的生物学功能是指,作为CRISPR-Cas系统中的同向重复序列的活性。
- [0152] 在某些实施方案中,所述分离的核酸分子是CRISPR-Cas系统中的同向重复序列。
- [0153] 在某些实施方案中,所述核酸分子包含选自下列的序列,或由选自下列的序列组成:
- [0154] (a)SEQ ID NOs:41任一项所示的核苷酸序列;
- [0155] (b)在严格条件下与(a)中所述的序列杂交的序列;或
- [0156] (c)(a)中所述的序列的互补序列。
- [0157] 在某些实施方案中,所述分离的核酸分子是RNA。
- [0158] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:
- [0159] (i)SEQ ID NO:41所示的序列;
- [0160] (ii)与SEQ ID NO:41所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;
- [0161] (iii)与SEQ ID NO:41所示的序列具有至少20%、至少30%、至少40%、至少50%、

至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列；

[0162] (iv)在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列；或

[0163] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0164] 在某些实施方案中，所述分离的核酸分子包含选自下列的序列，或由选自下列的序列组成：

[0165] (a)SEQ ID NO:41所示的核苷酸序列；

[0166] (b)在严格条件下与(a)中所述的序列杂交的序列；或

[0167] (c)SEQ ID NO:41所示的核苷酸序列的互补序列。

[0168] 在某些实施方案中，所述分离的核酸分子是RNA。

[0169] 在某些实施方案中，所述分离的核酸分子包含选自下列的序列，或由选自下列的序列组成：

[0170] (i)SEQ ID NO:42所示的序列；

[0171] (ii)与SEQ ID NO:42所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列；

[0172] (iii)与SEQ ID NO:42所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列；

[0173] (iv)在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列；或

[0174] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0175] 在某些实施方案中，所述分离的核酸分子包含选自下列的序列，或由选自下列的序列组成：

[0176] (a)SEQ ID NO:42所示的核苷酸序列；

[0177] (b)在严格条件下与(a)中所述的序列杂交的序列；或

[0178] (c)SEQ ID NO:42所示的核苷酸序列的互补序列。

[0179] 在某些实施方案中，所述分离的核酸分子是RNA。

[0180] 在某些实施方案中，所述分离的核酸分子包含选自下列的序列，或由选自下列的序列组成：

[0181] (i)SEQ ID NO:43所示的序列；

[0182] (ii)与SEQ ID NO:43所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列；

[0183] (iii)与SEQ ID NO:43所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列；

[0184] (iv)在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列；或

[0185] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0186] 在某些实施方案中，所述分离的核酸分子包含选自下列的序列，或由选自下列的序列组成：

[0187] (a)SEQ ID NO:43所示的核苷酸序列；

[0188] (b)在严格条件下与(a)中所述的序列杂交的序列；或

[0189] (c)SEQ ID NO:43所示的核苷酸序列的互补序列。

[0190] 在某些实施方案中，所述分离的核酸分子是RNA。

[0191] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0192] (i) SEQ ID NO:44所示的序列;

[0193] (ii) 与SEQ ID NO:44所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;

[0194] (iii) 与SEQ ID NO:44所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;

[0195] (iv) 在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或

[0196] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0197] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0198] (a) SEQ ID NO:44所示的核苷酸序列;

[0199] (b) 在严格条件下与(a)中所述的序列杂交的序列;或

[0200] (c) SEQ ID NO:44所示的核苷酸序列的互补序列。

[0201] 在某些实施方案中,所述分离的核酸分子是RNA。

[0202] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0203] (i) SEQ ID NO:45所示的序列;

[0204] (ii) 与SEQ ID NO:45所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;

[0205] (iii) 与SEQ ID NO:45所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;

[0206] (iv) 在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或

[0207] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0208] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0209] (a) SEQ ID NO:45所示的核苷酸序列;

[0210] (b) 在严格条件下与(a)中所述的序列杂交的序列;或

[0211] (c) SEQ ID NO:45所示的核苷酸序列的互补序列。

[0212] 在某些实施方案中,所述分离的核酸分子是RNA。

[0213] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0214] (i) SEQ ID NO:46所示的序列;

[0215] (ii) 与SEQ ID NO:46所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;

[0216] (iii) 与SEQ ID NO:46所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;

[0217] (iv) 在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或

[0218] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0219] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0220] (a) SEQ ID NO:46所示的核苷酸序列;

[0221] (b) 在严格条件下与(a)中所述的序列杂交的序列;或

[0222] (c) SEQ ID NO:46所示的核苷酸序列的互补序列。

[0223] 在某些实施方案中,所述分离的核酸分子是RNA。

[0224] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0225] (i) SEQ ID NO:47所示的序列;

[0226] (ii) 与SEQ ID NO:47所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;

[0227] (iii) 与SEQ ID NO:47所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;

[0228] (iv) 在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或

[0229] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0230] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0231] (a) SEQ ID NO:47所示的核苷酸序列;

[0232] (b) 在严格条件下与(a)中所述的序列杂交的序列;或

[0233] (c) SEQ ID NO:47所示的核苷酸序列的互补序列。

[0234] 在某些实施方案中,所述分离的核酸分子是RNA。

[0235] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0236] (i) SEQ ID NO:48所示的序列;

[0237] (ii) 与SEQ ID NO:48所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;

[0238] (iii) 与SEQ ID NO:48所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;

[0239] (iv) 在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或

[0240] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0241] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0242] (a) SEQ ID NO:48所示的核苷酸序列;

[0243] (b) 在严格条件下与(a)中所述的序列杂交的序列;或

[0244] (c) SEQ ID NO:48所示的核苷酸序列的互补序列。

[0245] 在某些实施方案中,所述分离的核酸分子是RNA。

[0246] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0247] (i) SEQ ID NO:49所示的序列;

[0248] (ii) 与SEQ ID NO:49所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;

[0249] (iii) 与SEQ ID NO:49所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;

[0250] (iv) 在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或

[0251] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0252] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0253] (a) SEQ ID NO:49所示的核苷酸序列;

[0254] (b) 在严格条件下与(a)中所述的序列杂交的序列;或

[0255] (c) SEQ ID NO:49所示的核苷酸序列的互补序列。

[0256] 在某些实施方案中,所述分离的核酸分子是RNA。

[0257] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0258] (i) SEQ ID NO:50所示的序列;

[0259] (ii) 与SEQ ID NO:50所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;

[0260] (iii) 与SEQ ID NO:50所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;

[0261] (iv) 在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或

[0262] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0263] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0264] (a) SEQ ID NO:50所示的核苷酸序列;

[0265] (b) 在严格条件下与(a)中所述的序列杂交的序列;或

[0266] (c) SEQ ID NO:50所示的核苷酸序列的互补序列。

[0267] 在某些实施方案中,所述分离的核酸分子是RNA。

[0268] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0269] (i) SEQ ID NO:51所示的序列;

[0270] (ii) 与SEQ ID NO:51所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;

[0271] (iii) 与SEQ ID NO:51所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;

[0272] (iv) 在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或

[0273] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0274] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0275] (a) SEQ ID NO:51所示的核苷酸序列;

- [0276] (b)在严格条件下与(a)中所述的序列杂交的序列;或
- [0277] (c)SEQ ID NO:51所示的核苷酸序列的互补序列。
- [0278] 在某些实施方案中,所述分离的核酸分子是RNA。
- [0279] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:
- [0280] (i)SEQ ID NO:52所示的序列;
- [0281] (ii)与SEQ ID NO:52所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;
- [0282] (iii)与SEQ ID NO:52所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;
- [0283] (iv)在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或
- [0284] (v) (i) - (iii)任一项中所述的序列的互补序列。
- [0285] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:
- [0286] (a)SEQ ID NO:52所示的核苷酸序列;
- [0287] (b)在严格条件下与(a)中所述的序列杂交的序列;或
- [0288] (c)SEQ ID NO:52所示的核苷酸序列的互补序列。
- [0289] 在某些实施方案中,所述分离的核酸分子是RNA。
- [0290] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:
- [0291] (i)SEQ ID NO:53所示的序列;
- [0292] (ii)与SEQ ID NO:53所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;
- [0293] (iii)与SEQ ID NO:53所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;
- [0294] (iv)在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或
- [0295] (v) (i) - (iii)任一项中所述的序列的互补序列。
- [0296] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:
- [0297] (a)SEQ ID NO:53所示的核苷酸序列;
- [0298] (b)在严格条件下与(a)中所述的序列杂交的序列;或
- [0299] (c)SEQ ID NO:53所示的核苷酸序列的互补序列。
- [0300] 在某些实施方案中,所述分离的核酸分子是RNA。
- [0301] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:
- [0302] (i)SEQ ID NO:54所示的序列;
- [0303] (ii)与SEQ ID NO:54所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;
- [0304] (iii)与SEQ ID NO:54所示的序列具有至少20%、至少30%、至少40%、至少50%、

至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列；

[0305] (iv)在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列；或

[0306] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0307] 在某些实施方案中，所述分离的核酸分子包含选自下列的序列，或由选自下列的序列组成：

[0308] (a)SEQ ID NO:54所示的核苷酸序列；

[0309] (b)在严格条件下与(a)中所述的序列杂交的序列；或

[0310] (c)SEQ ID NO:54所示的核苷酸序列的互补序列。

[0311] 在某些实施方案中，所述分离的核酸分子是RNA。

[0312] 在某些实施方案中，所述分离的核酸分子包含选自下列的序列，或由选自下列的序列组成：

[0313] (i)SEQ ID NO:55所示的序列；

[0314] (ii)与SEQ ID NO:55所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列；

[0315] (iii)与SEQ ID NO:55所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列；

[0316] (iv)在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列；或

[0317] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0318] 在某些实施方案中，所述分离的核酸分子包含选自下列的序列，或由选自下列的序列组成：

[0319] (a)SEQ ID NO:55所示的核苷酸序列；

[0320] (b)在严格条件下与(a)中所述的序列杂交的序列；或

[0321] (c)SEQ ID NO:55所示的核苷酸序列的互补序列。

[0322] 在某些实施方案中，所述分离的核酸分子是RNA。

[0323] 在某些实施方案中，所述分离的核酸分子包含选自下列的序列，或由选自下列的序列组成：

[0324] (i)SEQ ID NO:56所示的序列；

[0325] (ii)与SEQ ID NO:56所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列；

[0326] (iii)与SEQ ID NO:56所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列；

[0327] (iv)在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列；或

[0328] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0329] 在某些实施方案中，所述分离的核酸分子包含选自下列的序列，或由选自下列的序列组成：

[0330] (a)SEQ ID NO:56所示的核苷酸序列；

[0331] (b)在严格条件下与(a)中所述的序列杂交的序列；或

[0332] (c)SEQ ID NO:56所示的核苷酸序列的互补序列。

[0333] 在某些实施方案中，所述分离的核酸分子是RNA。

[0334] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0335] (i) SEQ ID NO:57所示的序列;

[0336] (ii) 与SEQ ID NO:57所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;

[0337] (iii) 与SEQ ID NO:57所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;

[0338] (iv) 在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或

[0339] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0340] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0341] (a) SEQ ID NO:57所示的核苷酸序列;

[0342] (b) 在严格条件下与(a)中所述的序列杂交的序列;或

[0343] (c) SEQ ID NO:57所示的核苷酸序列的互补序列。

[0344] 在某些实施方案中,所述分离的核酸分子是RNA。

[0345] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0346] (i) SEQ ID NO:58所示的序列;

[0347] (ii) 与SEQ ID NO:58所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;

[0348] (iii) 与SEQ ID NO:58所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;

[0349] (iv) 在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或

[0350] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0351] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0352] (a) SEQ ID NO:58所示的核苷酸序列;

[0353] (b) 在严格条件下与(a)中所述的序列杂交的序列;或

[0354] (c) SEQ ID NO:58所示的核苷酸序列的互补序列。

[0355] 在某些实施方案中,所述分离的核酸分子是RNA。

[0356] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0357] (i) SEQ ID NO:59所示的序列;

[0358] (ii) 与SEQ ID NO:59所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;

[0359] (iii) 与SEQ ID NO:59所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;

[0360] (iv) 在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或

[0361] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0362] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0363] (a) SEQ ID NO:59所示的核苷酸序列;

[0364] (b) 在严格条件下与(a)中所述的序列杂交的序列;或

[0365] (c) SEQ ID NO:59所示的核苷酸序列的互补序列。

[0366] 在某些实施方案中,所述分离的核酸分子是RNA。

[0367] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0368] (i) SEQ ID NO:60所示的序列;

[0369] (ii) 与SEQ ID NO:60所示的序列相比具有一个或多个碱基的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个碱基的置换、缺失或添加)的序列;

[0370] (iii) 与SEQ ID NO:60所示的序列具有至少20%、至少30%、至少40%、至少50%、至少60%、至少70%、至少80%、至少90%、至少95%的序列同一性的序列;

[0371] (iv) 在严格条件下与(i) - (iii)任一项中所述的序列杂交的序列;或

[0372] (v) (i) - (iii)任一项中所述的序列的互补序列。

[0373] 在某些实施方案中,所述分离的核酸分子包含选自下列的序列,或由选自下列的序列组成:

[0374] (a) SEQ ID NO:60所示的核苷酸序列;

[0375] (b) 在严格条件下与(a)中所述的序列杂交的序列;或

[0376] (c) SEQ ID NO:60所示的核苷酸序列的互补序列。

[0377] CRISPR/Cas复合物

[0378] 在第五方面,本发明提供了一种复合物,其包含:

[0379] (i) 蛋白组分,其选自:本发明的蛋白、缀合物或融合蛋白,及其任意组合;和

[0380] (ii) 核酸组分,其从5'至3'方向包含如上文所述的分离的核酸分子和能够与靶序列杂交的导向序列,

[0381] 其中,所述蛋白组分与核酸组分相互结合形成复合物。

[0382] 在某些实施方案中,所述导向序列连接于所述核酸分子的3'端。

[0383] 在某些实施方案中,所述导向序列包含所述靶序列的互补序列。

[0384] 在某些实施方案中,所述核酸组分是CRISPR-Cas系统中的导向RNA。

[0385] 在某些实施方案中,所述核酸分子是RNA。

[0386] 在某些实施方案中,所述复合物不包含反式作用crRNA(tracrRNA)。

[0387] 在某些实施方案中,所述导向序列在长度上为至少5个、至少10个、至少14个。在某些实施方案中,所述导向序列在长度上为10-30个、或15-25个、或15-22个、或19-25个、19-22个核苷酸或14-28个核苷酸。

[0388] 在某些实施方案中,所述分离的核酸分子在长度上为55-70个核苷酸,例如55-65个核苷酸,例如60-65个核苷酸,例如62-65个核苷酸,例如63-64个核苷酸。在某些实施方案中,所述分离的核酸分子在长度上为15-30个核苷酸,例如15-25个核苷酸,例如20-25个核苷酸,例如22-24个核苷酸,例如23个核苷酸。

[0389] 编码核酸、载体及宿主细胞

[0390] 在第六方面,本发明提供了一种分离的核酸分子,其包含:

[0391] (i)编码本发明的蛋白或融合蛋白的核苷酸序列;

[0392] (ii)编码如第四方面所述的分离的核酸分子;或

[0393] (iii)包含(i)和(ii)的核苷酸序列。

[0394] 在某些实施方案中,(i)-(iii)任一项中所述的核苷酸序列经密码子优化用于在原核细胞中进行表达。在某些实施方案中,(i)-(iii)任一项中所述的核苷酸序列经密码子优化用于在真核细胞中进行表达。

[0395] 在第七方面,本发明还提供了一种载体,其包含如第六方面所述的分离的核酸分子。本发明的载体可以是克隆载体,也可以是表达载体。在某些实施方案中,本发明的载体是例如质粒,粘粒,噬菌体,柯斯质粒等等。在某些实施方案中,所述载体能够在受试者(例如哺乳动物,例如人)体内表达本发明的蛋白、融合蛋白、如第四方面所述的分离的核酸分子或如第五方面所述的复合物。

[0396] 在第八方面,本发明还提供了包含如上所述的分离的核酸分子或载体的宿主细胞。此类宿主细胞包括但不限于,原核细胞例如大肠杆菌细胞,以及真核细胞例如酵母细胞,昆虫细胞,植物细胞和动物细胞(如哺乳动物细胞,例如小鼠细胞、人细胞等)。本发明的细胞还可以是细胞系,例如293T细胞。

[0397] 组合物及载体组合物

[0398] 在第九方面,本发明还提供了一种组合物,其包含:

[0399] (i)第一组分,其选自:本发明的蛋白、缀合物、融合蛋白、编码所述蛋白或融合蛋白的核苷酸序列,以及其任意组合;和

[0400] (ii)第二组分,其为包含导向RNA的核苷酸序列,或者编码所述包含导向RNA的核苷酸序列的核苷酸序列;

[0401] 其中,所述导向RNA从5'至3'方向包含同向重复序列和导向序列,所述导向序列能够与靶序列杂交;

[0402] 所述导向RNA能够与(i)中所述的蛋白、缀合物或融合蛋白形成复合物。

[0403] 在某些实施方案中,所述同向重复序列是如第四方面所定义的分离的核酸分子。

[0404] 在某些实施方案中,所述导向序列连接至所述同向重复序列的3'端。在某些实施方案中,所述导向序列包含所述靶序列的互补序列。

[0405] 在某些实施方案中,所述组合物不包含tracrRNA。

[0406] 在某些实施方案中,所述组合物是非天然存在的或经修饰的。在某些实施方案中,所述组合物中的至少一个组分是非天然存在的或经修饰的。在某些实施方案中,所述第一组分是非天然存在的或经修饰的;和/或,所述第二组分是非天然存在的或经修饰的。

[0407] 在某些实施方案中,当所述靶标序列为DNA时,所述靶序列位于原间隔序列临近基序(PAM)的3'端,并且所述PAM具有5'-ATG所示的序列。

[0408] 在某些实施方案中,当所述靶标序列为DNA时,所述靶序列位于原间隔序列临近基序(PAM)的3'端,并且所述PAM具有5'-TTN所示的序列,其中,N选自A、G、T、C。

[0409] 在某些实施方案中,当所述靶标序列为DNA时,所述靶序列位于原间隔序列临近基序(PAM)的3'端,并且所述PAM具有5'-KTR所示的序列。

[0410] 在某些实施方案中,当所述靶序列为RNA时,所述靶序列不具有PAM结构域限制。

[0411] 在某些实施方案中,所述靶序列是来自原核细胞或真核细胞的DNA或RNA序列。在某些实施方案中,所述靶序列是非天然存在的DNA或RNA序列。

[0412] 在某些实施方案中,所述靶序列存在于细胞内。在某些实施方案中,所述靶序列存在于细胞核内或细胞质(例如,细胞器)内。在某些实施方案中,所述细胞是真核细胞。在某些实施方案中,所述细胞是原核细胞。

[0413] 在某些实施方案中,所述蛋白连接有一个或多个NLS序列。在某些实施方案中,所述缀合物或融合蛋白包含一个或多个NLS序列。在某些实施方案中,所述NLS序列连接至所述蛋白的N端或C端。在某些实施方案中,所述NLS序列融合至所述蛋白的N端或C端。

[0414] 在第十方面,本发明还提供了一种组合物,其包含一种或多种载体,所述一种或多种载体包含:

[0415] (i) 第一核酸,其为编码本发明的蛋白或融合蛋白的核苷酸序列;任选地所述第一核酸可操作地连接至第一调节元件;以及

[0416] (ii) 第二核酸,其编码包含导向RNA的核苷酸序列;任选地所述第二核酸可操作地连接至第二调节元件;

[0417] 其中:

[0418] 所述第一核酸与第二核酸存在于相同或不同的载体上;

[0419] 所述导向RNA从5'至3'方向包含同向重复序列和导向序列,所述导向序列能够与靶序列杂交;

[0420] 所述导向RNA能够与(i)中所述的效应蛋白或融合蛋白形成复合物。

[0421] 在某些实施方案中,所述同向重复序列是如第四方面所定义的分离的核酸分子。

[0422] 在某些实施方案中,所述导向序列连接至所述同向重复序列的3'端。在某些实施方案中,所述导向序列包含所述靶序列的互补序列。

[0423] 在某些实施方案中,所述组合物不包含tracrRNA。

[0424] 在某些实施方案中,所述组合物是非天然存在的或经修饰的。在某些实施方案中,所述组合物中的至少一个组分是非天然存在的或经修饰的。

[0425] 在某些实施方案中,所述第一调节元件是启动子,例如诱导型启动子。

[0426] 在某些实施方案中,所述第二调节元件是启动子,例如诱导型启动子。

[0427] 在某些实施方案中,当所述靶标序列为DNA时,所述靶序列位于原间隔序列临近基序(PAM)的3'端,并且所述PAM具有5'-ATG所示的序列。

[0428] 在某些实施方案中,当所述靶标序列为DNA时,所述靶序列位于原间隔序列临近基序(PAM)的3'端,并且所述PAM具有5'-TTN所示的序列,其中,N选自A、G、T、C。

[0429] 在某些实施方案中,当所述靶标序列为DNA时,所述靶序列位于原间隔序列临近基序(PAM)的3'端,并且所述PAM具有5'-KTR所示的序列。

[0430] 在某些实施方案中,当所述靶序列为RNA时,所述靶序列不具有PAM结构域限制。

[0431] 在某些实施方案中,所述靶序列是来自原核细胞或真核细胞的DNA或RNA序列。在某些实施方案中,所述靶序列是非天然存在的DNA或RNA序列。

[0432] 在某些实施方案中,所述靶序列存在于细胞内。在某些实施方案中,所述靶序列存在于细胞核内或细胞质(例如,细胞器)内。在某些实施方案中,所述细胞是真核细胞。在某些实施方案中,所述细胞是原核细胞。

[0433] 在某些实施方案中,所述蛋白连接有一个或多个NLS序列。在某些实施方案中,所述缀合物或融合蛋白包含一个或多个NLS序列。在某些实施方案中,所述NLS序列连接至所述蛋白的N端或C端。在某些实施方案中,所述NLS序列融合至所述蛋白的N端或C端。

[0434] 在某些实施方案中,一种类型的载体是质粒,其是指其中可以例如通过标准分子克隆技术插入另外的DNA片段的环状双链DNA环。另一种类型的载体是病毒载体,其中病毒衍生的DNA或RNA序列存在于用于包装病毒(例如,逆转录病毒、复制缺陷型逆转录病毒、腺病毒、复制缺陷型腺病毒、以及腺相关病毒)的载体中。病毒载体还包含由用于转染到一种宿主细胞中的病毒携带的多核苷酸。某些载体(例如,具有细菌复制起点的细菌载体和附加型哺乳动物载体)能够在它们被导入的宿主细胞中自主复制。其他载体(例如,非附加型哺乳动物载体)在引入宿主细胞后整合到该宿主细胞的基因组中,并且由此与该宿主基因组一起复制。而且,某些载体能够指导它们可操作连接的基因的表达。这样的载体在此被称为“表达载体”。在重组DNA技术中使用的普通表达载体通常是质粒形式。

[0435] 重组表达载体可包含处于适合于在宿主细胞中的核酸表达的形式本发明的核酸分子,这意味着这些重组表达载体包含基于待用于表达的宿主细胞而选择的一种或多种调节元件,所述调节元件可操作地连接至待表达的核酸序列。

[0436] 递送及递送组合物

[0437] 本发明的蛋白、缀合物、融合蛋白、如第四方面所述的分离的核酸分子、本发明的复合物、如第六方面所述的分离的核酸分子、如第七方面所述的载体、如第九方面及第十方面所述的组合物,可以通过本领域已知的任何方法进行递送。此类方法包括但不限于,电穿孔、脂转染、核转染、显微注射、声孔效应、基因枪、磷酸钙介导的转染、阳离子转染、脂质体转染、树枝状转染、热激转染、核转染、磁转染、脂转染、穿刺转染、光学转染、试剂增强性核酸摄取、以及经由脂质体、免疫脂质体、病毒颗粒、人工病毒体等的递送。

[0438] 因此,在另一个方面,本发明提供了一种递送组合物,其包含递送载体,以及选自下列的一种或多种:本发明的蛋白、缀合物、融合蛋白、如第四方面所述的分离的核酸分子、本发明的复合物、如第六方面所述的分离的核酸分子、如第七方面所述的载体、如第九方面及第十方面所述的组合物。

[0439] 在某些实施方案中,所述递送载体是粒子。

[0440] 在某些实施方案中,所述递送载体选自脂质颗粒、糖颗粒、金属颗粒、蛋白颗粒、脂质体、外泌体、微泡、基因枪或病毒载体(例如,复制缺陷型逆转录病毒、慢病毒、腺病毒或腺相关病毒)。

[0441] 试剂盒

[0442] 在另一个方面,本发明提供了一种试剂盒,其包含如上所述的组分中的一种或多种。在某些实施方案中,所述试剂盒包含一种或多种选自下列的组分:本发明的蛋白、缀合物、融合蛋白、如第四方面所述的分离的核酸分子、本发明的复合物、如第六方面所述的分离的核酸分子、如第七方面所述的载体、如第九方面及第十方面所述的组合物。

[0443] 在某些实施方案中,本发明的试剂盒包含如第九方面所述的组合物。在某些实施方案中,所述试剂盒还包含使用所述组合物的说明书。

[0444] 在某些实施方案中,本发明的试剂盒包含如第十方面所述的组合物。在某些实施方案中,所述试剂盒还包含使用所述组合物的说明书。

[0445] 在某些实施方案中,本发明的试剂盒中包含的组分可以被提供于任何适合的容器中。

[0446] 在某些实施方案中,所述试剂盒还包含一种或多种缓冲液。缓冲液可以是任何缓冲液,包括但不限于碳酸钠缓冲液、碳酸氢钠缓冲液、硼酸盐缓冲液、Tris缓冲液、MOPS缓冲液、HEPES缓冲液及其组合。在某些实施方案中,该缓冲液是碱性的。在某些实施方案中,该缓冲液具有从约7至约10的pH。

[0447] 在某些实施方案中,该试剂盒还包括一个或多个寡核苷酸,该一个或多个寡核苷酸对应于一个用于插入进载体中的导向序列,以便可操作地连接该导向序列和调节元件。在某些实施方案中,该试剂盒包括同源重组模板多核苷酸。

[0448] 方法及用途

[0449] 在另一个方面,本发明提供了一种修饰靶基因的方法,其包括:将如第五方面所述的复合物、如第九方面所述的组合物或如第十方面所述的组合物与所述靶基因接触,或者递送至包含所述靶基因的细胞中;所述靶序列存在于所述靶基因中。

[0450] 在某些实施方案中,所述靶基因存在于细胞内。在某些实施方案中,所述细胞是原核细胞。在某些实施方案中,所述细胞是真核细胞。在某些实施方案中,所述细胞是哺乳动物细胞。在某些实施方案中,所述细胞是人类细胞。在某些实施方案中,所述细胞选自非人灵长类动物、牛、猪或啮齿类动物细胞。在某些实施方案中,所述细胞是非哺乳动物真核细胞,例如家禽或鱼等。在某些实施方案中,所述细胞是植物细胞,例如栽培植物(如木薯、玉米、高粱、小麦或水稻)、藻类、树或蔬菜具有的细胞。

[0451] 在某些实施方案中,所述靶基因存在于体外的核酸分子(例如,质粒)中。在某些实施方案中,所述靶基因存在于质粒中。

[0452] 在某些实施方案中,所述修饰是指所述靶序列的断裂,如DNA的双链断裂或RNA的单链断裂。

[0453] 在某些实施方案中,所述断裂导致靶基因的转录降低。

[0454] 在某些实施方案中,所述方法还包括:将编辑模板与所述靶基因接触,或者递送至包含所述靶基因的细胞中。在此类实施方案中,所述方法通过与外源模板多核苷酸同源重组修复所述断裂的靶基因,其中所述修复导致一种突变,包括所述靶基因的一个或多个核苷酸的插入、缺失、或取代。在某些实施方案中,所述突变导致在从包含该靶序列的基因表达的蛋白质中的一个或多个氨基酸改变。

[0455] 因此,在某些实施方案中,所述修饰还包括将编辑模板(例如外源核酸)插入所述断裂中。

[0456] 在某些实施方案中,所述的蛋白、缀合物、融合蛋白、分离的核酸分子、复合物、载体或组合物包含于递送载体中。

[0457] 在某些实施方案中,所述递送载体选自脂质颗粒、糖颗粒、金属颗粒、蛋白颗粒、脂质体、外泌体、病毒载体(如复制缺陷型逆转录病毒、慢病毒、腺病毒或腺相关病毒)。

[0458] 在某些实施方案中,所述方法其用于改变靶基因或编码靶基因产物的核酸分子中的一个或多个靶序列来修饰细胞、细胞系或生物体。

[0459] 在另一个方面,本发明提供了一种改变基因产物的表达的方法,其包括:将如第五方面所述的复合物、如第九方面所述的组合物或如第十方面所述的组合物与编码所述基因

产物的核酸分子接触,或者递送至包含所述核酸分子的细胞中,所述靶序列存在于所述核酸分子中。

[0460] 在某些实施方案中,所述核酸分子存在于细胞内。在某些实施方案中,所述细胞是原核细胞。在某些实施方案中,所述细胞是真核细胞。在某些实施方案中,所述细胞是哺乳动物细胞。在某些实施方案中,所述细胞是人类细胞。在某些实施方案中,所述细胞选自非人灵长类动物、牛、猪或啮齿类动物细胞。在某些实施方案中,所述细胞是非哺乳动物真核细胞,例如家禽或鱼等。在某些实施方案中,所述细胞是植物细胞,例如栽培植物(如木薯、玉米、高粱、小麦或水稻)、藻类、树或蔬菜具有的细胞。

[0461] 在某些实施方案中,所述核酸分子存在于体外的核酸分子(例如,质粒)中。在某些实施方案中,所述核酸分子存在于质粒中。

[0462] 在某些实施方案中,所述基因产物的表达被改变(例如,增强或降低)。在某些实施方案中,所述基因产物的表达被增强。在某些实施方案中,所述基因产物的表达被降低。

[0463] 在某些实施方案中,所述基因产物是蛋白。

[0464] 在某些实施方案中,所述的蛋白、缀合物、融合蛋白、分离的核酸分子、复合物、载体或组合物包含于递送载体中。

[0465] 在某些实施方案中,所述递送载体选自脂质颗粒、糖颗粒、金属颗粒、蛋白颗粒、脂质体、外泌体、病毒载体(如复制缺陷型逆转录病毒、慢病毒、腺病毒或腺相关病毒)。

[0466] 在某些实施方案中,所述方法其用于改变靶基因或编码靶基因产物的核酸分子中的一个或多个靶序列来修饰细胞、细胞系或生物体。

[0467] 在另一个方面,本发明涉及如第一方面所述的蛋白、如第二方面所述的缀合物、如第三方面所述的融合蛋白、如第四方面所述的分离的核酸分子、如第五方面所述的复合物、如第六方面所述的分离的核酸分子、如第七方面所述的载体、如第九方面所述的组合物、如第十方面所述的组合物、本发明的试剂盒或递送组合物,用于核酸编辑的用途。

[0468] 在某些实施方案中,所述核酸编辑包括基因或基因组编辑,例如修饰基因、敲除基因、改变基因产物的表达、修复突变、和/或插入多核苷酸。

[0469] 在另一个方面,本发明涉及如第一方面所述的蛋白、如第二方面所述的缀合物、如第三方面所述的融合蛋白、如第四方面所述的分离的核酸分子、如第五方面所述的复合物、如第六方面所述的分离的核酸分子、如第七方面所述的载体、如第九方面所述的组合物、如第十方面所述的组合物、本发明的试剂盒或递送组合物,在制备制剂中的用途,所述制剂用于:

[0470] (i) 离体基因或基因组编辑;

[0471] (ii) 离体单链DNA的检测;

[0472] (iii) 编辑靶基因座中的靶序列来修饰生物或非人类生物;

[0473] (iv) 治疗由靶基因座中的靶序列的缺陷引起的病症。

[0474] 细胞及细胞子代

[0475] 在某些情况下,由本发明的方法引入到细胞的修饰可以使得细胞和其子代被改变以改进其生物产物(如抗体、淀粉、乙醇或其他期望的细胞输出物)的产生。在某些情况下,由本发明的方法引入到细胞的修饰可以使得细胞和其子代包括使所生产物发生变化的改变。

[0476] 因此,在另一方面,本发明还涉及如上所述的方法获得的细胞或其子代,其中所述细胞含有在其野生型中不存在的修饰。

[0477] 本发明还涉及如上所述的细胞或其子代的细胞产物。

[0478] 本发明还涉及一种体外的、离体的或体内的细胞或细胞系或它们的子代,所述细胞或细胞系或它们的子代包含:如第一方面所述的蛋白、如第二方面所述的缀合物、如第三方面所述的融合蛋白、如第四方面所述的分离的核酸分子、如第五方面所述的复合物、如第六方面所述的分离的核酸分子、如第七方面所述的载体、如第九方面所述的组合物、如第十方面所述的组合物、本发明的试剂盒或递送组合物。

[0479] 在某些实施方案中,所述细胞是原核细胞。

[0480] 在某些实施方案中,所述细胞是真核细胞。在某些实施方案中,所述细胞是哺乳动物细胞。在某些实施方案中,所述细胞是人类细胞。某些实施方案中,所述细胞是非人哺乳动物细胞,例如非人灵长类动物、牛、羊、猪、犬、猴、兔、啮齿类(如大鼠或小鼠)的细胞。在某些实施方案中,所述细胞是非哺乳动物真核细胞,例如家禽鸟类(如鸡)、鱼类或甲壳动物(如蛤蜊、虾)的细胞。在某些实施方案中,所述细胞是植物细胞,例如单子叶植物或双子叶植物具有的细胞或栽培植物或粮食作物如木薯、玉米、高粱、大豆、小麦、燕麦或水稻具有的细胞,例如藻类、树或生产植物、果实或蔬菜(例如,树类如柑橘树、坚果树;茄属植物、棉花、烟草、番茄、葡萄、咖啡、可可等)。

[0481] 在某些实施方案中,所述细胞是干细胞或干细胞系。

[0482] 术语定义

[0483] 在本发明中,除非另有说明,否则本文中使用的科学和技术名词具有本领域技术人员所通常理解的含义。并且,本文中所用的分子遗传学、核酸化学、化学、分子生物学、生物化学、细胞培养、微生物学、细胞生物学、基因组学和重组DNA等操作步骤均为相应领域内广泛使用的常规步骤。同时,为了更好地理解本发明,下面提供相关术语的定义和解释。

[0484] 在本发明中,表述“Cas12j”是指,本发明人首次发现并鉴定的一种Cas效应蛋白,其具有选自下列的氨基酸序列:

[0485] (i) SEQ ID NOs:1-20、107、108任一项所示的序列;

[0486] (ii) 与SEQ ID NOs:1-20、107、108任一项所示的序列相比具有一个或多个氨基酸的置换、缺失或添加(例如1个,2个,3个,4个,5个,6个,7个,8个,9个或10个氨基酸的置换、缺失或添加)的序列;或

[0487] (iii) 与SEQ ID NOs:1-20、107、108任一项所示的序列具有至少80%、至少85%、至少90%、至少91%、至少92%、至少93%、至少94%、至少95%、至少96%、至少97%、至少98%、或至少99%的序列同一性的序列。

[0488] 本发明的Cas12j是一种在导向RNA引导下与靶序列特定位点结合并切割的核酸内切酶,同时具有DNA和RNA内切酶活性。

[0489] 如本文中所使用的,术语“规律成簇的间隔短回文重复(CRISPR)-CRISPR-相关(Cas)(CRISPR-Cas)系统”或“CRISPR系统”可互换地使用并且具有本领域技术人员通常理解的含义,其通常包含与CRISPR相关(“Cas”)基因的表达有关的转录产物或其他元件,或者能够指导所述Cas基因活性的转录产物或其他元件。此类转录产物或其他元件可以包含编码Cas效应蛋白的序列和包含CRISPR RNA(crRNA)的导向RNA,以及在CRISPR-Cas9系统中所

含有的反式作用crRNA(tracrRNA)序列,或来自CRISPR基因座的其他序列或转录产物。

[0490] 如本文中所使用的,术语“Cas效应蛋白”、“Cas效应酶”可互换地使用并且是指,CRISPR-Cas系统中呈现的任一种大于长度800个氨基酸的蛋白质。在某些情况下,这类蛋白是指从Cas基因座中鉴定的蛋白。

[0491] 如本文中所使用的,术语“导向RNA(guide RNA)”、“成熟crRNA”可互换地使用并且具有本领域技术人员通常理解的含义。一般而言,导向RNA可以包含同向(direct)重复序列和导向序列(guide sequence),或者基本上由或由同向重复序列和导向序列(在内源性CRISPR系统背景下也称为间隔序列(spacer))组成。在某些情况下,导向序列是与靶序列具有足够互补性从而与所述靶序列杂交并引导CRISPR/Cas复合物与所述靶序列的特异性结合的任何多核苷酸序列。在某些实施方案中,当最佳比对时,导向序列与其相应靶序列之间的互补程度为至少50%、至少60%、至少70%、至少80%、至少90%、至少95%、或至少99%。确定最佳比对在本领域的普通技术人员的能力范围内。例如,存在公开和可商购的比对算法和程序,诸如但不限于ClustalW、matlab中的史密斯-沃特曼算法(Smith-Waterman)、Bowtie、Geneious、Biopython以及SeqMan。

[0492] 在某些情况下,所述导向序列在长度上为至少5个、至少10个、至少15个、至少16个、至少17个、至少18个、至少19个、至少20个、至少21个、至少22个、至少23个、至少24个、至少25个、至少26个、至少27个、至少28个、至少29个、至少30个、至少35个、至少40个、至少45个或至少50个核苷酸。在某些情况下,所述导向序列在长度上为不超过50个、45个、40个、35个、30个、25个、24个、23个、22个、21个、20个、15个、10个或更少个核苷酸。在某些实施方案中,所述导向序列在长度上为10-30个、或15-25个、或15-22个、或19-25个或19-22个核苷酸。

[0493] 在某些情况下,所述同向重复序列在长度上为至少10个、至少15个、至少16个、至少17个、至少18个、至少19个、至少20个、至少21个、至少22个、至少23个、至少24个、至少25个、至少26个、至少27个、至少28个、至少29个、至少30个、至少35个、至少40个、至少45个、至少50个、至少55个、至少56个、至少57个、至少58个、至少59个、至少60个、至少61个、至少62个、至少63个、至少64个、至少65个或至少70个核苷酸。在某些情况下,所述同向重复序列在长度上为不超过70个、65个、64个、63个、62个、61个、60个、59个、58个、57个、56个、55个、50个、45个、40个、35个、30个、29个、28个、27个、26个、25个、24个、23个、22个、21个、20个、15个、10个或更少个核苷酸。在某些实施方案中,所述同向重复序列在长度上为55-70个核苷酸,例如55-65个核苷酸,例如60-65个核苷酸,例如62-65个核苷酸,例如63-64个核苷酸。在某些实施方案中,所述同向重复序列在长度上为15-30个核苷酸,例如15-25个核苷酸,例如20-25个核苷酸,例如22-24个核苷酸,例如23个核苷酸。在某些实施方案中,所述同向重复序列在长度上不少于32nt,例如32nt-37nt。

[0494] 如本文中所使用的,术语“CRISPR/Cas复合物”是指,导向RNA(guide RNA)或成熟crRNA与Cas蛋白结合所形成的核糖核蛋白复合体,其包含杂交到靶序列上并且与Cas蛋白结合的导向序列。该核糖核蛋白复合体能够识别并切割能与该导向RNA或成熟crRNA杂交的多核苷酸。

[0495] 因此,在形成CRISPR/Cas复合物的情况下,“靶序列”是指被设计为具有靶向性的导向序列所靶向的多核苷酸,例如与该导向序列具有互补性的序列,其中靶序列与导向序

列之间的杂交将促进CRISPR/Cas复合物的形成。完全互补性不是必需的,只要存在足够互补性以引起杂交并且促进一种CRISPR/Cas复合物的形成即可。靶序列可以包含任何多核苷酸,如DNA或RNA。在某些情况下,所述靶序列位于细胞的细胞核或细胞质中。在某些情况下,该靶序列可位于真核细胞的一个细胞器例如线粒体或叶绿体内。可被用于重组到包含该靶序列的靶基因座中的序列或模板被称为“编辑模板”或“编辑多核苷酸”或“编辑序列”。在某些实施方案中,所述编辑模板为外源核酸。在某些实施方案中,该重组是同源重组。

[0496] 在本发明中,表述“靶序列”或“靶多核苷酸”可以是对细胞(例如,真核细胞)而言任何内源或外源的多核苷酸。例如,该靶多核苷酸可以是一种存在于真核细胞的细胞核中的多核苷酸。该靶多核苷酸可以是一个编码基因产物(例如,蛋白质)的序列或一个非编码序列(例如,调节多核苷酸或无用DNA)。在某些情况下,据信该靶序列应该与原间隔序列临近基序(PAM)相关。对PAM的精确序列和长度要求取决于使用的Cas效应酶而不同,但是PAM典型地是临近原间隔序列(也即,靶序列)的2-5个碱基对序列。本领域技术人员能够鉴定与给定的Cas效应蛋白一起使用的PAM序列。

[0497] 在某些情况下,靶序列或靶多核苷酸可以包括多个疾病相关基因和多核苷酸以及信号传导生化途径相关基因和多核苷酸。此类靶序列或靶多核苷酸的非限制性实例,包括分别提交于2012年12月12日和2013年1月2日的美国临时专利申请61/736,527和61/748,427、提交于2013年12月12日的国际申请PCT/US2013/074667中所列举的那些,其全部通过引用并入本文。

[0498] 在某些情况下,靶序列或靶多核苷酸的实例包括与信号传导生化途径相关的序列,例如信号传导生化途径相关基因或多核苷酸。靶多核苷酸的实例包括疾病相关基因或多核苷酸。“疾病相关”基因或多核苷酸是指与非疾病对照的组织或细胞相比,在来源于疾病影响的组织的细胞中以异常水平或以异常形式产生转录或翻译产物的任何基因或多核苷酸。在改变的表达与疾病的出现和/或进展相关的情况下,它可以是一个以异常高的水平被表达的基因;或者,它可以是一个以异常低的水平被表达的基因。疾病相关基因还指具有一个或多个突变或直接负责或与一个或多个负责疾病的病因学的基因连锁不平衡的遗传变异的基因。转录的或翻译的产物可以是已知的或未知的,并且可以处于正常或异常水平。

[0499] 如本文中所使用的,术语“野生型”具有本领域技术人员通常理解的含义,其表示生物、菌株、基因的典型形式或者当它在自然界存在时区别于突变体或变体形式的特征,其可从自然中的来源分离并且没有被人为有意地修饰。

[0500] 如本文中所使用的,术语“非天然存在的”或“工程化的”可互换地使用并且表示人工的参与。当这些术语用于描述核酸分子或多肽时,其表示该核酸分子或多肽至少基本上从它们在自然界中或如发现于自然界中的与其结合的至少另一种组分游离出来。

[0501] 如本文中所使用的,术语“直系同源物(orthologue, ortholog)”具有本领域技术人员通常理解的含义。作为进一步指导,如本文中所述的蛋白质的“直系同源物”是指属于不同物种的蛋白质,该蛋白质执行与作为其直系同源物的蛋白相同或相似的功能。

[0502] 如本文中所使用的,术语“同一性”用于指两个多肽之间或两个核酸之间序列的匹配情况。当两个进行比较的序列中的某个位置都被相同的碱基或氨基酸单体亚单元占据时(例如,两个DNA分子的每一个中的某个位置都被腺嘌呤占据,或两个多肽的每一个中的某个位置都被赖氨酸占据),那么各分子在该位置上是同一的。两个序列之间的“百分数同一

性”是由这两个序列共有的匹配位置数目除以进行比较的位置数目 $\times 100$ 的函数。例如,如果两个序列的10个位置中有6个匹配,那么这两个序列具有60%的同一性。例如,DNA序列CTGACT和CAGGTT共有50%的同一性(总共6个位置中有3个位置匹配)。通常,在将两个序列比对以产生最大同一性时进行比较。这样的比对可通过使用,例如,可通过计算机程序例如Align程序(DNAstar, Inc.)方便地进行的Needleman等人(1970) J. Mol. Biol. 48:443-453的方法来实现。还可使用已整合入ALIGN程序(版本2.0)的E. Meyers和W. Miller (Comput. Appl Biosci., 4:11-17 (1988))的算法,使用PAM120权重残基表(weight residue table)、12的缺口长度罚分和4的缺口罚分来测定两个氨基酸序列之间的百分数同一性。此外,可使用已整合入GCG软件包(可在www.gcg.com上获得)的GAP程序中的Needleman和Wunsch (J Mol Biol. 48:444-453 (1970))算法,使用Blossum 62矩阵或PAM250矩阵以及16、14、12、10、8、6或4的缺口权重(gap weight)和1、2、3、4、5或6的长度权重来测定两个氨基酸序列之间的百分数同一性。

[0503] 如本文中所使用的,术语“载体”是指,可将多聚核苷酸插入其中的一种核酸运载工具。当载体能使插入的多核苷酸编码的蛋白获得表达时,载体称为表达载体。载体可以通过转化,转导或者转染导入宿主细胞,使其携带的遗传物质元件在宿主细胞中获得表达。载体是本领域技术人员公知的,包括但不限于:质粒;噬菌粒;柯斯质粒;人工染色体,例如酵母人工染色体(YAC)、细菌人工染色体(BAC)或P1来源的人工染色体(PAC);噬菌体如 λ 噬菌体或M13噬菌体及动物病毒等。可用作载体的动物病毒包括但不限于,逆转录酶病毒(包括慢病毒)、腺病毒、腺相关病毒、疱疹病毒(如单纯疱疹病毒)、痘病毒、杆状病毒、乳头瘤病毒、乳头多瘤空泡病毒(如SV40)。一种载体可以含有多种控制表达的元件,包括但不限于,启动子序列、转录起始序列、增强子序列、选择元件及报告基因。另外,载体还可含有复制起始位点。

[0504] 如本文中所使用的,术语“宿主细胞”是指,可用于导入载体的细胞,其包括但不限于,如大肠杆菌或枯草菌等的原核细胞,如酵母细胞或曲霉菌等的真菌细胞,如S2果蝇细胞或Sf9等的昆虫细胞,或者如纤维原细胞,CHO细胞,COS细胞,NS0细胞,HeLa细胞,BHK细胞,HEK 293细胞或人细胞等的动物细胞。

[0505] 本领域技术人员将理解,表达载体的设计可取决于诸如待转化的宿主细胞的选择、所希望的表达水平等因素。一种载体可以被引入到宿主细胞中而由此产生转录物、蛋白质、或肽,包括由如本文所述的蛋白、融合蛋白、分离的核酸分子等(例如,CRISPR转录物,如核酸转录物、蛋白质、或酶)。

[0506] 如本文中所使用的,术语“调节元件”旨在包括启动子、增强子、内部核糖体进入位点(IRES)、和其他表达控制元件(例如转录终止信号,如多聚腺苷酸化信号和多聚U序列),其详细描述可参考戈德尔(Goeddel),《基因表达技术:酶学方法》(GENE EXPRESSION TECHNOLOGY:METHODS IN ENZYMOLOGY)185,学术出版社(Academic Press),圣地亚哥(San Diego),加利福尼亚州(1990)。在某些情况下,调节元件包括指导一个核苷酸序列在许多类型的宿主细胞中的组成型表达的那些序列以及指导该核苷酸序列只在某些宿主细胞中表达的那些序列(例如,组织特异型调节序列)。组织特异型启动子可主要指导在感兴趣的期望组织中的表达,所述组织例如肌肉、神经元、骨、皮肤、血液、特定的器官(例如肝脏、胰腺)、或特殊的细胞类型(例如淋巴细胞)。在某些情况下,调节元件还可以时序依赖性方式

(如以细胞周期依赖性或发育阶段依赖性方式)指导表达,该方式可以是或者可以不是组织或细胞类型特异性的。在某些情况下,术语“调节元件”涵盖的是增强子元件,如WPRES;CMV增强子;在HTLV-I的LTR中的R-U5'片段(Mol.Cell.Biol.,第8(1)卷,第466-472页,1988);SV40增强子;以及在兔 β -珠蛋白的外显子2与3之间的内含子序列(Proc.Natl.Acad.Sci.USA.,第78(3)卷,第1527-31页,1981)。

[0507] 如本文中所使用的,术语“启动子”具有本领域技术人员公知的含义,其是指一段位于基因的上游能启动下游基因表达的非编码核苷酸序列。组成型(constitutive)启动子是这样的核苷酸序列:当其与编码或者限定基因产物的多核苷酸可操作地相连时,在细胞的大多数或者所有生理条件下,其导致细胞中基因产物的产生。诱导型启动子是这样的核苷酸序列,当可操作地与编码或者限定基因产物的多核苷酸相连时,基本上只有当对应于所述启动子的诱导物在细胞中存在时,其导致所述基因产物在细胞内产生。组织特异性启动子是这样的核苷酸序列:当可操作地与编码或者限定基因产物的多核苷酸相连时,基本上只有当细胞是该启动子对应的组织类型的细胞时,其才导致在细胞中产生基因产物。

[0508] 如本文中所使用的,术语“可操作地连接”旨在表示感兴趣的核苷酸序列以一种允许该核苷酸序列的表达的方式被连接至该一种或多种调节元件(例如,处于一种体外转录/翻译系统中或当该载体被引入到宿主细胞中时,处于该宿主细胞中)。

[0509] 如本文中所使用的,术语“互补性”是指核酸与另一个核酸序列借助于传统的沃森-克里克或其他非传统类型形成一个或多个氢键的能力。互补百分比表示一个核酸分子中可与一个第二核酸序列形成氢键(例如,沃森-克里克碱基配对)的残基的百分比(例如,10个之中有5、6、7、8、9、10个即为50%、60%、70%、80%、90%、和100%互补)。“完全互补”表示一个核酸序列的所有连续残基与一个第二核酸序列中的相同数目的连续残基形成氢键。如本文使用的“基本上互补”是指在一个具有8、9、10、11、12、13、14、15、16、17、18、19、20、21、22、23、24、25、30、35、40、45、50个或更多个核苷酸的区域上至少为60%、65%、70%、75%、80%、85%、90%、95%、97%、98%、99%、或100%的互补程度,或者是指在严格条件下杂交的两个核酸。

[0510] 如本文中所使用的,对于杂交的“严格条件”是指与靶序列具有互补性的一个核酸主要地与该靶序列杂交并且基本上不杂交到非靶序列上的条件。严格条件通常是序列依赖性的,并且取决于许多因素而变化。一般而言,该序列越长,则该序列特异性地杂交到其靶序列上的温度就越高。严格条件的非限制性实例描述于蒂森(Tijssen)(1993)的《生物化学和分子生物学中的实验室技术-核酸探针杂交》(Laboratory Techniques In Biochemistry And Molecular Biology-Hybridization With Nucleic Acid Probes),第I部分,第二章,“杂交原理概述和核酸探针分析策略”(“Overview of principles of hybridization and the strategy of nucleic acid probe assay”),爱思唯尔(Elsevier),纽约。

[0511] 如本文中所使用的,术语“杂交”是指其中一个或多个多核苷酸反应形成一种复合物的反应,该复合物经由这些核苷酸残基之间的碱基的氢键键合而稳定化。氢键键合可以借助于沃森-克里克碱基配对、Hoogsteen结合或以任何其他序列特异性方式而发生。该复合物可包含形成一个双链体的两条链、形成多链复合物的三条或多条链、单个自我杂交链、或这些的任何组合。杂交反应可以构成一个更广泛的过程(如PCR的开始、或经由一种酶的

多核苷酸的切割)中的一个步骤。能够与一个给定序列杂交的序列被称为该给定序列的“互补物”。

[0512] 如本文中所使用的,术语“表达”是指,藉此从DNA模板转录成多核苷酸(如转录成mRNA或其他RNA转录物)的过程和/或转录的mRNA随后藉此翻译成肽、多肽或蛋白质的过程。转录物和编码的多肽可以总称为“基因产物”。如果多核苷酸来源于基因组DNA,表达可以包括真核细胞中mRNA的剪接。

[0513] 如本文中所使用的,术语“接头”是指,由多个氨基酸残基通过肽键连接形成的线性多肽。本发明的接头可以为人工合成的氨基酸序列,或天然存在的多肽序列,例如具有铰链区功能的多肽。此类接头多肽是本领域众所周知的(参见例如,Holliger,P.等人(1993) Proc.Natl.Acad.Sci.USA 90:6444-6448;Poljak,R.J.等人(1994) Structure 2:1121-1123)。

[0514] 如本文中所使用的,术语“治疗”是指,治疗或治愈病症,延缓病症的症状的发作,和/或延缓病症的发展。

[0515] 如本文中所使用的,术语“受试者”包括但不限于各种动物,例如哺乳动物,例如牛科动物、马科动物、羊科动物、猪科动物、犬科动物、猫科动物、兔科动物、啮齿类动物(例如,小鼠或大鼠)、非人灵长类动物(例如,猕猴或食蟹猴)或人。在某些实施方式中,所述受试者(例如人)患有病症(例如,疾病相关基因缺陷所导致的病症)。

[0516] 发明的有益效果

[0517] 与现有技术相比,本发明的Cas蛋白及系统具有显著的有利方面。例如,本发明的Cas效应蛋白具有严格的错配容忍度,使其可能具有更低的脱靶率。例如,本发明的Cas效应蛋白拥有更加严谨的PAM识别方式,从而显著降低脱靶效应。

附图说明

[0518] 图1为cas12j蛋白对pre-crRNA加工的凝胶电泳结果。

[0519] 图2A-2B为cas12j蛋白的PAM结构域分析的结果。

[0520] 图3为CRISPR/Cas12j系统的DNA切割方式的鉴定结果。

[0521] 图4为Cas12j.4、Cas12j.19、Cas12j.22体外切割位点分析的结果。

[0522] 图5为Cas12j.19在不同温度下体外酶切活性检测的结果。

[0523] 图6为CRISPR/Cas12j.19系统中不同spacer长度对酶切活性影响的结果。

[0524] 图7为CRISPR/Cas12j.19系统中不同repeat长度对酶切活性影响的结果。WT表示未经截短的repeat序列。

[0525] 图8为CRISPR/Cas12j.19系统对于spacer错配容忍的结果。WT表示未经突变的spacer序列。

[0526] 序列信息

[0527] 本发明涉及的部分序列的信息提供于下面的表1中。

[0528] 表1:序列的描述

[0529]

SEQ ID NO:	描述
1	Cas12j.3 的氨基酸序列
2	Cas12j.4 的氨基酸序列
3	Cas12j.5 的氨基酸序列
4	Cas12j.6 的氨基酸序列
5	Cas12j.7 的氨基酸序列
6	Cas12j.8 的氨基酸序列
7	Cas12j.9 的氨基酸序列
8	Cas12j.10 的氨基酸序列
9	Cas12j.11 的氨基酸序列
10	Cas12j.12 的氨基酸序列
11	Cas12j.13 的氨基酸序列
12	Cas12j.14 的氨基酸序列
13	Cas12j.15 的氨基酸序列
14	Cas12j.16 的氨基酸序列
15	Cas12j.17 的氨基酸序列
16	Cas12j.18 的氨基酸序列
17	Cas12j.19 的氨基酸序列
18	Cas12j.20 的氨基酸序列

[0530]

19	Cas12j.21 的氨基酸序列
20	Cas12j.22 的氨基酸序列
21	Cas12j.3 编码核酸序列
22	Cas12j.4 编码核酸序列
23	Cas12j.5 编码核酸序列
24	Cas12j.6 编码核酸序列
25	Cas12j.7 编码核酸序列
26	Cas12j.8 编码核酸序列
27	Cas12j.9 编码核酸序列
28	Cas12j.10 编码核酸序列
29	Cas12j.11 编码核酸序列
30	Cas12j.12 编码核酸序列
31	Cas12j.13 编码核酸序列
32	Cas12j.14 编码核酸序列
33	Cas12j.15 编码核酸序列
34	Cas12j.16 编码核酸序列
35	Cas12j.17 编码核酸序列
36	Cas12j.18 编码核酸序列
37	Cas12j.19 编码核酸序列
38	Cas12j.20 编码核酸序列
39	Cas12j.21 编码核酸序列
40	Cas12j.22 编码核酸序列
41	Cas12j.3 原型同向重复序列
42	Cas12j.4 原型同向重复序列
43	Cas12j.5 原型同向重复序列
44	Cas12j.6 原型同向重复序列
45	Cas12j.7 原型同向重复序列
46	Cas12j.8 原型同向重复序列
47	Cas12j.9 原型同向重复序列
48	Cas12j.10 原型同向重复序列
49	Cas12j.11 原型同向重复序列
50	Cas12j.12 原型同向重复序列
51	Cas12j.13 原型同向重复序列
52	Cas12j.14 原型同向重复序列
53	Cas12j.15 原型同向重复序列
54	Cas12j.16 原型同向重复序列
55	Cas12j.17 原型同向重复序列
56	Cas12j.18 原型同向重复序列
57	Cas12j.19 原型同向重复序列
58	Cas12j.20 原型同向重复序列
59	Cas12j.21 原型同向重复序列
60	Cas12j.22 原型同向重复序列
61	Cas12j.3 原型同向重复序列的编码核酸序列

[0531]

62	Cas12j.4 原型同向重复序列的编码核酸序列
63	Cas12j.5 原型同向重复序列的编码核酸序列
64	Cas12j.6 原型同向重复序列的编码核酸序列
65	Cas12j.7 原型同向重复序列的编码核酸序列
66	Cas12j.8 原型同向重复序列的编码核酸序列
67	Cas12j.9 原型同向重复序列的编码核酸序列
68	Cas12j.10 原型同向重复序列的编码核酸序列
69	Cas12j.11 原型同向重复序列的编码核酸序列
70	Cas12j.12 原型同向重复序列的编码核酸序列
71	Cas12j.13 原型同向重复序列的编码核酸序列
72	Cas12j.14 原型同向重复序列的编码核酸序列
73	Cas12j.15 原型同向重复序列的编码核酸序列
74	Cas12j.16 原型同向重复序列的编码核酸序列
75	Cas12j.17 原型同向重复序列的编码核酸序列
76	Cas12j.18 原型同向重复序列的编码核酸序列
77	Cas12j.19 原型同向重复序列的编码核酸序列
78	Cas12j.20 原型同向重复序列的编码核酸序列
79	Cas12j.21 原型同向重复序列的编码核酸序列
80	Cas12j.22 原型同向重复序列的编码核酸序列
81	NLS 序列
82	Cas12j.3-NLS 融合蛋白的氨基酸序列
83	Cas12j.4-NLS 融合蛋白的氨基酸序列
84	Cas12j.5-NLS 融合蛋白的氨基酸序列
85	Cas12j.6-NLS 融合蛋白的氨基酸序列
86	Cas12j.7-NLS 融合蛋白的氨基酸序列
87	Cas12j.8-NLS 融合蛋白的氨基酸序列
88	Cas12j.9-NLS 融合蛋白的氨基酸序列
89	Cas12j.10-NLS 融合蛋白的氨基酸序列
90	Cas12j.11-NLS 融合蛋白的氨基酸序列
91	Cas12j.12-NLS 融合蛋白的氨基酸序列
92	Cas12j.13-NLS 融合蛋白的氨基酸序列
93	Cas12j.14-NLS 融合蛋白的氨基酸序列
94	Cas12j.15-NLS 融合蛋白的氨基酸序列
95	Cas12j.16-NLS 融合蛋白的氨基酸序列
96	Cas12j.17-NLS 融合蛋白的氨基酸序列
97	Cas12j.18-NLS 融合蛋白的氨基酸序列
98	Cas12j.19-NLS 融合蛋白的氨基酸序列
99	Cas12j.20-NLS 融合蛋白的氨基酸序列
100	Cas12j.21-NLS 融合蛋白的氨基酸序列
101	Cas12j.22-NLS 融合蛋白的氨基酸序列
102	表达 Cas12j.3 系统的质粒
103	PAM 文库序列
104	Pre-crRNA 加工及 PAM 消耗导向 RNA

[0532]	105	Cas12j.19 导向 RNA
	106	靶向的双链 DNA 序列
	107	Cas12j.1 氨基酸序列
	108	Cas12j.2 氨基酸序列

具体实施方式

[0533] 现参照下列意在举例说明本发明(而非限定本发明)的实施例来描述本发明。

[0534] 除非特别指明,否则基本上按照本领域内熟知的以及在各种参考文献中描述的常规方法进行实施例中描述的实验和方法。例如,本发明中所使用的免疫学、生物化学、化学、分子生物学、微生物学、细胞生物学、基因组学和重组DNA等常规技术,可参见萨姆布鲁克(Sambrook)、弗里奇(Fritsch)和马尼亚蒂斯(Maniatis),《分子克隆:实验室手册》(MOLECULAR CLONING:A LABORATORY MANUAL),第2次编辑(1989);《当代分子生物学实验手册》(CURRENT PROTOCOLS IN MOLECULAR BIOLOGY)(F.M.奥苏贝尔(F.M.Ausubel)等人编辑,(1987));《酶学方法》(METHODS IN ENZYMOLOGY)系列(学术出版公司):《PCR 2:实用方法》(PCR 2:A PRACTICAL APPROACH)(M.J.麦克弗森(M.J.MacPherson)、B.D.黑姆斯(B.D.Hames)和G.R.泰勒(G.R.Taylor)编辑(1995))、哈洛(Harlow)和拉内(Lane)编辑(1988)《抗体:实验室手册》(ANTIBODIES,A LABORATORY MANUAL),以及《动物细胞培养》(ANIMAL CELL CULTURE)(R.I.弗雷谢尼(R.I.Freshney)编辑(1987))。

[0535] 另外,实施例中未注明具体条件者,按照常规条件或制造商建议的条件进行。所用试剂或仪器未注明生产厂商者,均为可以通过市购获得的常规产品。本领域技术人员知晓,实施例以举例方式描述本发明,且不意欲限制本发明所要求保护的。本文中提及的全部公开案和其他参考资料以其全文通过引用合并入本文。

[0536] 以下实施例涉及的部分试剂的来源如下:

[0537] LB液体培养基:10g胰蛋白胍(Tryptone),5g酵母提取物(Yeast Extract),10g NaCl,定容至1L,灭菌。若需加抗生素,则待培养基冷却后加,50 μ g/ml的终浓度。

[0538] 氯仿/异戊醇:240ml的氯仿加10ml的异戊醇,混匀。

[0539] RNP缓冲液:100mM氯化钠,50mM Tris-HCl,10mM MgCl₂,100 μ g/ml BSA,pH 7.9。

[0540] 原核表达载体pACYC-Duet-1和pUC19购自金斯瑞生物有限公司。

[0541] 大肠杆菌感受态EC100购自Epicentre公司。

[0542] 实施例1.Cas12j基因和Cas12j导向RNA的获得

[0543] 1、CRISPR和基因的注释:使用Prodigal对将NCBI和JGI数据库的微生物基因组和宏基因组数据进行基因注释得到所有蛋白,同时用Piler-CR进行CRISPR座的注释,参数均为默认参数。

[0544] 2、蛋白质的过滤:通过序列一致性对注释蛋白去冗余,去除序列完全一致的蛋白,同时将长度大于800个氨基酸的蛋白划分为大分子蛋白。由于目前发现的所有第二类CRISPR/Cas系统的效应蛋白长度多大于900个氨基酸,所以为了降低计算复杂度,我们在挖掘CRISPR效应蛋白的时候只对大于800个氨基酸的大分子蛋白进行考虑。

[0545] 3、CRISPR相关大分子蛋白的获得:将每一个CRISPR座上下游延伸10Kb,将对CRISPR邻近区间内的非冗余大分子蛋白进行鉴定。

[0546] 4、CRISPR相关大分子蛋白质的聚类：使用BLASTP对非冗余大分子CRISPR相关蛋白进行内部的两两比对，输出Evalue<1E-10的比对结果。使用MCL对BLASTP的输出结果进行聚类分析，CRISPR相关蛋白质家族。

[0547] 5、CRISPR富集大分子蛋白质家族的鉴定：使用BLASTP对CRISPR相关蛋白质家族的蛋白比对到去除CRISPR相关蛋白的非冗余大分子蛋白数据库，输出Evalue<1E-10的比对结果。如果一个非CRISPR相关蛋白数据库发现的同源蛋白小于100%，那么则说明这个家族的蛋白在CRISPR区域是富集的，通过这种方法我们对CRISPR富集大分子蛋白质家族进行鉴定。

[0548] 6、蛋白功能和结构域的注释：利用Pfam数据库，NR数据库以及从NCBI收集的Cas蛋白对CRISPR富集大分子蛋白质家族进行注释，得到新的CRISPR/Cas蛋白质家族。利用Mafft对每个CRISPR/Cas家族蛋白进行多重序列比对，然后用JPred和HHpred进行保守结构域分析，鉴定含有RuvC结构域的蛋白质家族。

[0549] 在此基础上，本发明人获得了一种全新的Cas效应蛋白，即Cas12j，以其22种活性同源物序列，分别命名为Cas12j.3(SEQ ID NO:1)、Cas12j.4(SEQ ID NO:2)、Cas12j.5(SEQ ID NO:3)、Cas12j.6(SEQ ID NO:4)、Cas12j.7(SEQ ID NO:5)、Cas12j.8(SEQ ID NO:6)、Cas12j.9(SEQ ID NO:7)、Cas12j.10(SEQ ID NO:8)、Cas12j.11(SEQ ID NO:9)、Cas12j.12(SEQ ID NO:10)、Cas12j.13(SEQ ID NO:11)、Cas12j.14(SEQ ID NO:12)、Cas12j.15(SEQ ID NO:13)、Cas12j.16(SEQ ID NO:14)、Cas12j.17(SEQ ID NO:15)、Cas12j.18(SEQ ID NO:16)、Cas12j.19(SEQ ID NO:17)、Cas12j.20(SEQ ID NO:18)、Cas12j.21(SEQ ID NO:19)、Cas12j.22(SEQ ID NO:20)、Cas12j.1(SEQ ID NO:107)、Cas12j.2(SEQ ID NO:108)，20种同源物的编码DNA分别如SEQ ID NOs:21-40所示。Cas12j.3、Cas12j.4、Cas12j.5、Cas12j.6、Cas12j.7、Cas12j.8、Cas12j.9、Cas12j.10、Cas12j.11、Cas12j.12、Cas12j.13、Cas12j.14、Cas12j.15、Cas12j.16、Cas12j.17、Cas12j.18、Cas12j.19、Cas12j.20所对应的原型同向重复序列(pre-crRNA中所含有的repeat序列)分别如SEQ ID NOs:41-60所示。

[0550] 实施例2.Cas12j基因对pre-crRNA的加工

[0551] 1、Cas12j蛋白的体外表达及纯化

[0552] Cas12j蛋白的体外表达及纯化的步骤具体如下：

[0553] 1、人工合成编码带有核定位信号的Cas12j蛋白(SEQ ID NO:82-101)的DNA序列。

[0554] 2、将步骤1合成的双链DNA分子与原核表达载体pET-30a(+)连接，得到重组质粒pET-30a-CRISPR/Cas12j。

[0555] 3、将重组质粒pET-30a-CRISPR/Cas12j导入大肠杆菌EC100，得到重组菌，将该重组菌命名为EC100-CRISPR/Cas12j。

[0556] 取EC100-CRISPR/Cas12j的单克隆，接种至100mL LB液体培养基(含50μg/mL氨苄霉素)，37℃、200rpm振荡培养12h，得到培养菌液。

[0557] 4、取培养菌液，按体积比为1:100接种至50mL LB液体培养基(含50μg/mL氨苄霉素)，37℃、200rpm振荡培养至OD_{600nm}值为0.6，然后加入IPTG并使其浓度为1mM，28℃、220rpm振荡培养4h，4℃、10000rpm离心10min，收集菌体沉淀。

[0558] 5、取菌体沉淀，加入100mL pH 8.0、100mM的Tris-HCl缓冲液，重悬后超声破碎(超声波功率600W，循环程序为：破碎4s，停6s，共20min)，然后4℃、10000rpm离心10min，收集上

清液甲。

[0559] 6、取上清液甲,4℃、12000rpm离心10min,收集上清液乙。

[0560] 7、采用GE公司生产的镍柱对上清液乙进行纯化(纯化的具体步骤参考镍柱的说明书),然后采用赛默飞世尔公司生产的蛋白定量试剂盒对Cas12j蛋白进行定量。

[0561] 二、Cas12j蛋白导向RNA的转录及纯化:

[0562] 1、设计导向RNA转录的模板,转录模板的结构为:T7启动子+Cas12j的原型的repeat (SEQ ID NO:41-60)+spacer (SEQ ID NO:104),引物的设计使用Primer5.0软件,保证正向引物和反向引物有至少18bp的重叠序列。

[0563] 2、配置如下反应体系,轻轻吹打混匀后短暂离心,置于PCR仪中缓慢退火:

[0564] PCR扩增反应

组分	体积 (μl)
正向引物(100nM)	7.5
反向引物(100nM)	7.5
2*KAPA Mix	25
ddH ₂ O	10
总体积	50

[0566] 引物退火PCR反应程序

温度 (℃)	时间	ramp at(℃/s)
98℃	5min	2℃/s

	85°C/95°C	0.05s	—
	85°C	1min	0.03°C/s
	75°C/85°C	0.05s	—
	75°C	1min	0.03°C/s
	72°C/75°C	0.05s	—
	72°C	1min	0.03°C/s
	55°C/65°C	0.05s	—
	55°C	1min	0.03°C/s
[0568]	45°C/55°C	0.05s	—
	45°C	1min	0.03°C/s
	35°C/45°C	0.05s	—
	35°C	1min	0.03°C/s
	30°C/35°C	0.05s	—
	30°C	1min	0.03°C/s
	25°C	1min	—
	10°C	forever	—

[0569] 3、使用MinElute PCR Purification Kit进行模板的纯化,步骤如下:

[0570] 1) 向PCR产物中加入5倍体积的PB,将一个MinElute柱子放至2ml收集管上,室温静置2min,12000g/2min;

[0571] 2) 弃废液,加入750 μ l Buffer PE(用之前记得加乙醇),12000g/2min;

[0572] 3) 弃废液,加入350 μ l Buffer PE,12000g/2min,弃废液,12000g,空离2min;

[0573] 4) 将MinElute柱子换至新的1.5ml离心管上,开盖,65°C静置2min;

[0574] 5) 加入20 μ l预热的EB溶液,静置2min后,12000g/2min,为了提高回收率,可将离心管内容物过2-3遍MinElute离心柱;

[0575] 6) 用Nanodrop测定浓度,冻存-20°C备用。

[0576] 4、导向RNA的纯化:酚:氯仿:异戊醇(25:24:1)抽提去除体系内的DNaseI

[0577] 1) 向转录后的反应体系中加入80 μ l RNA free H₂O,调整体积至100 μ l;

[0578] 2) 取出2ml的Phase Lock Gel (PLG) Heavy,15000g,离心2min,加入100 μ l酚:氯仿:异戊醇(25:24:1)、100 μ l经过DNaseI消化的RNA,用手轻轻弹Phase-Lock tube 5-10次,使其混合均匀,之后15°C/16000g离心12min;

[0579] 3) 取一个新的RNA-free的1.5ml离心管,将上步离心的上清吸出至离心管中,注意不要吸到凝胶,加入与上清等体积的异丙醇以及十分之一体积的醋酸钠溶液,用枪头吸打混匀后放入-20°C冰箱1h或过夜静置;

[0580] 4) 4℃/16000g, 离心30min, 弃上清, 加入75%预冷的乙醇, 将沉淀吸打混匀, 4℃/16000g, 离心12min, 弃上清, 在通风橱静置2-3min, 晾干RNA表面的乙醇, 加入100μl的RNA free H₂O, 吸打混匀。

[0581] 5) 用Nanodrop测定纯化后的crRNA浓度, 并统一稀释至250ng/μl, 分装至200μl的PCR离心管中, 冻存-80℃备用。

[0582] 4、Cas12f的precrRNA转录采用NEB的HiScribe T7高效RNA合成试剂盒, 反应体系如下表所示:

[0583] DNA转录体系

组分	体积 (μl)
ATP(100mM)	2
GTP(100nM)	2
CTP (100nM)	2
[0584] UTP(100nM)	2
10*Reaction buffer	2
T7 RNA Polymerase Mix	2
DNA 模板	8
总计	20

[0585] 设置PCR反应程序为: 37℃/3h或31℃/forever, 加入DNaseI, 37℃/45min

[0586] 5、precrRNA的纯化:

[0587] (1) 酚:氯仿:异戊醇(25:24:1) 抽提去除体系内的DNaseI

[0588] 1) 向转录后的反应体系中加入80μl RNA free H₂O, 调整体积至100μl;

[0589] 2) 取出2ml的Phase Lock Gel (PLG) Heavy, 15000g, 离心2min, 加入100μl酚:氯仿:异戊醇(25:24:1)、100μl经过DNaseI消化的RNA, 用手轻轻弹Phase-Lock tube 5-10次, 使其混合均匀, 之后15℃/16000g离心12min;

[0590] 3) 取一个新的RNA-free的1.5ml离心管, 将②步离心的上清吸出至离心管中, 注意不要吸到凝胶, 加入与上清等体积的异丙醇以及十分之一体积的醋酸钠溶液, 用枪头吸打混匀后放入-20℃冰箱1h或过夜静置;

[0591] 4) 4℃/16000g, 离心30min, 弃上清, 加入75%预冷的乙醇, 将沉淀吸打混匀, 4℃/16000g, 离心12min, 弃上清, 在通风橱静置2-3min, 晾干RNA表面的乙醇, 加入100μl的RNA free H₂O, 吸打混匀。

[0592] (2) 跑胶并从聚丙烯酰胺凝胶中纯化precrRNA, 使用ZYMO RESEARCH的ZR Small-RNATM PAGE Recovery Kit试剂盒纯化回收precrRNA。步骤如下:

[0593] 1) precrRNA条带大小为90bp左右, 切割相应条带的RNA片段, 转移至1.5ml RNA-free的离心管中;

[0594] 2) 使用SquisherTM-single将胶完全捣碎, 加入400μl的RNA Recovery Buffer, 65

℃水浴锅加热15min;

[0595] 3) 液氮速冻5min, 立即取出放入65℃水浴锅加热5min;

[0596] 4) 取出Zymo-Spin™ IV的柱子于收集管上, 然后将溶解后的凝胶加入其中, 12000g离心5min, 并保留收集管中液体;

[0597] 5) 取出Zymo-Spin™ IIIC的柱子于新的收集管上, 将上步收集的液体加入其中, 2000g离心2min, 保留收集管中液体;

[0598] 6) 估算收集管中液体体积, 加入2倍体积的RNA MAX Buffer, 上下颠倒混匀;

[0599] 7) 取出Zymo-Spin™ IC的柱子于新的收集管中, 将⑥步收集管中的液体加入其中, 静置2min后, 12000g离心2min;

[0600] 8) 加入800μl RNA Wash Buffer (注意用之前按照说明书加入一定体积的无水乙醇), 12000g离心2min, 弃收集管中液体;

[0601] 9) 加入400μl RNA Wash Buffer, 12000g离心2min, 弃收集管中液体, 再空离2min;

[0602] 10) 65℃烘箱静置1min, 加入20μl RNA-free H₂O, 用nanodrop测定所收集的precrRNA的浓度, 并统一调整浓度至200ng/μl, 分装在PCR离心管中, 负80℃冻存储用。

[0603] 6、建立体外pre-crRNA酶切体系

[0604] (1) 配置如下反应体系, 轻轻吹打混匀后短暂离心。置于37℃, 1hour;

[0605] 体外pre-crRNA酶切体系

	试剂	用量
	pre-crRNA	400 ng
[0606]	Cas protein	1 μg
	RNA Cleavage Buffer	1 μL
	RNA-free H ₂ O	补至 10 μL

[0607] (2) 向以上反应体系中加入10μl 2×RNA loading dye, 置于98℃, 3min。反应结束后立即置于冰上2min;

[0608] (3) 10%TBE-Urea聚丙烯酰胺凝胶上样孔中上样10μl, 150V/40min;

[0609] (4) 在1×TBE电泳缓冲液中加入SYBR Gold nucleic acid gel stain dye, 置入凝胶, 室温染色10-15min后扫胶。

[0610] 扫胶结果如图1所示, 结果显示, Cas12j.1、Cas12j.4、Cas12j.18、Cas12j.19、Cas12j.21、Cas12j.22在体外具有pre-crRNA切割活性。

[0611] 实施例3. Cas12j蛋白的PAM结构域鉴定

[0612] 1. 构建重组质粒pACYC-Duet-1+CRISPR/Cas12j并测序。根据测序结果, 对重组质粒pACYC-Duet-1+CRISPR/Cas12j进行结构描述如下: 将载体pACYC-Duet-1的限制性内切酶Pml I和Kpn I识别序列间的小片段替换为Cas12j基因 (SEQ ID NO:21-40所示的序列中自5'端起第1位至3'末端最后一位所示的双链DNA分子)。重组质粒pACYC-Duet-1+CRISPR/Cas12j表达Cas12j蛋白 (SEQ ID NO:1-20、107、108) 和SEQ ID NO:104所示的Cas12j导向RNA。

[0613] 2. 重组质粒pACYC-Duet-1+CRISPR/Cas12j中含有表达盒, 该表达盒的核苷酸序列

由Cas12j基因分别与SEQ ID NO:104连接构成。例如SEQ ID NO:102所示。SEQ ID NO:102所示的序列中,自5'末端起第1至44位为pLacZ启动子的核苷酸序列,第45至3056位为Cas12j.3基因的核苷酸序列,第3057至3143位为rrnB T1终止子的核苷酸序列(用于终止转录)。自5'末端起第3144至3178位为J23119启动子的核苷酸序列,第3179至3241位为CRISPR阵列的核苷酸序列,第3244至3268位为rrnB-T2终止子的核苷酸序列(用于终止转录)。

[0614] 3. 重组大肠杆菌的获得:将重组质粒pACYC-Duet-1+CRISPR/Cas12j导入大肠杆菌EC100中,得到重组大肠杆菌,命名为EC100/pACYC-Duet-1+CRISPR/Cas12j。将重组质粒pACYC-Duet-1导入大肠杆菌EC100中,得到重组大肠杆菌,命名为EC100/pACYC-Duet-1。

[0615] 4. PAM文库的构建:人工合成SEQ ID NO:103所示的序列,并连接到pUC19载体,其中SEQ ID NO:103所示的序列包括5'端八个随机碱基和靶序列。对PAM文库的靶标序列5'端前面设计了8个随机碱基构建质粒文库。将质粒分别转入到含有Cas12j基因座的大肠杆菌中和不含有Cas.12j基因座的大肠杆菌中。在37℃下处理1小时后,我们对质粒进行提取,并对PAM区域序列进行PCR扩增和测序。

[0616] 5. PAM文库结构域的获得:分别统计实验组和对照组中65,536种组合的PAM序列出现次数,并用各自组所有的PAM序列数目进行标准化。对于任意一条PAM序列,当 \log_2 (对照组标准化值/实验组标准化值)大于3.5时,我们认为这条PAM被显著消耗。我们用Weblogo对显著消耗的PAM序列进行预测,发现各个蛋白的PAM结构域,其中,Cas12j.1为5'-TTWV,Cas12j.4、Cas12j.12为5'-TTN,Cas12j.18为5'-AYR,Cas12j.19为5'-ATG,Cas12j.21为5'-VTTG,Cas12j.22为5'-KTR。PAM结构域分析结果见附图2A-2B。

[0617] 实施例4. CRISPR/Cas12j系统的DNA切割方式的鉴定

[0618] 一、Cas12j蛋白的体外表达及纯化

[0619] Cas12j蛋白的体外表达及纯化的步骤具体如下:

[0620] 1、人工合成编码带有核定位信号的Cas12j蛋白(SEQ ID NO:82-101)的DNA序列。

[0621] 2、将步骤1合成的双链DNA分子与原核表达载体pET-30a(+)连接,得到重组质粒pET-30a-CRISPR/Cas12j。

[0622] 3、将重组质粒pET-30a-CRISPR/Cas12j导入大肠杆菌EC100,得到重组菌,将该重组菌命名为EC100-CRISPR/Cas12j。

[0623] 取EC100-CRISPR/Cas12j的单克隆,接种至100mL LB液体培养基(含50 μ g/mL氨苄霉素),37℃、200rpm振荡培养12h,得到培养菌液。

[0624] 4、取培养菌液,按体积比为1:100接种至50mL LB液体培养基(含50 μ g/mL氨苄霉素),37℃、200rpm振荡培养至OD_{600nm}值为0.6,然后加入IPTG并使其浓度为1mM,28℃、220rpm振荡培养4h,4℃、10000rpm离心10min,收集菌体沉淀。

[0625] 5、取菌体沉淀,加入100mL pH 8.0、100mM的Tris-HCl缓冲液,重悬后超声破碎(超声波功率600W,循环程序为:破碎4s,停6s,共20min),然后4℃、10000rpm离心10min,收集上清液甲。

[0626] 6、取上清液甲,4℃、12000rpm离心10min,收集上清液乙。

[0627] 7、采用GE公司生产的镍柱对上清液乙进行纯化(纯化的具体步骤参考镍柱的说明书),然后采用赛默飞世尔公司生产的蛋白定量试剂盒对Cas12j蛋白进行定量。

[0628] 二、Cas12j蛋白导向RNA的转录及纯化:

[0629] 1、设计导向RNA转录的模板,转录模板的结构为:T7启动子+Cas12j的原型repeat (SEQ ID NO:41-60)+spacer(SEQ ID NO:105),引物的设计使用Primer5.0软件,保证正向引物和反向引物有至少18bp的重叠序列。

[0630] 2、配置如下反应体系,轻轻吹打混匀后短暂离心,置于PCR仪中缓慢退火:

[0631] PCR扩增反应

	组分	体积 (μl)
[0632]	正向引物(100nM)	7.5
	反向引物(100nM)	7.5
	2*KAPA Mix	25
[0633]	ddH ₂ O	10
	总体积	50

[0634] 3、使用MinElute PCR Purification Kit进行模板的纯化,步骤如下:

[0635] 1) 向PCR产物中加入5倍体积的PB,将一个MinElute柱子放至2ml收集管上,室温静置2min,12000g/2min;

[0636] 2) 弃废液,加入750μl Buffer PE(用之前记得加乙醇),12000g/2min;

[0637] 3) 弃废液,加入350μl Buffer PE,12000g/2min,弃废液,12000g,空离2min;

[0638] 4) 将MinElute柱子换至新的1.5ml离心管上,开盖,65℃静置2min;

[0639] 5) 加入20μl预热的EB溶液,静置2min后,12000g/2min,为了提高回收率,可将离心管内容物过2-3遍MinElute离心柱;

[0640] 6) 用Nanodrop测定浓度,冻存-20℃备用。

[0641] 4、导向RNA的纯化:酚:氯仿:异戊醇(25:24:1)抽提去除体系内的DNaseI

[0642] 1) 向转录后的反应体系中加入80μl RNA free H₂O,调整体积至100μl;

[0643] 2) 取出2ml的Phase Lock Gel (PLG) Heavy,15000g,离心2min,加入100μl酚:氯仿:异戊醇(25:24:1)、100μl经过DNaseI消化的RNA,用手轻轻弹Phase-Lock tube 5-10次,使其混合均匀,之后15℃/16000g离心12min;

[0644] 3) 取一个新的RNA-free的1.5ml离心管,将上步离心的上清吸出至离心管中,注意不要吸到凝胶,加入与上清等体积的异丙醇以及十分之一体积的醋酸钠溶液,用枪头吸打混匀后放入-20℃冰箱1h或过夜静置;

[0645] 4) 4℃/16000g,离心30min,弃上清,加入75%预冷的乙醇,将沉淀吸打混匀,4℃/16000g,离心12min,弃上清,在通风橱静置2-3min,晾干RNA表面的乙醇,加入100μl的RNA free H₂O,吸打混匀。

[0646] 5、用Nanodrop测定纯化后的crRNA浓度,并统一稀释至250ng/μl,分装至200μl的PCR离心管中,冻存-80℃备用。

[0647] 6、双链DNA酶切体系的建立:

[0648] (1) 配置如下反应体系,轻轻吹打混匀后短暂离心。置于37℃,15min;

[0649] DNA切割反应体系

	组分	加样量
	12j-crRNA (250ng/μl)	600ng
[0650]	12j protein (0.5μg/μl)	0.5μg
	10*DNA Cleavage buffer	1μl
	RNA-Free H₂O	补至 7μl
[0651]	(2) 加入300ng底物DNA (SEQ ID NO:106) (100ng/μl), 3μL, 轻轻吹打混匀后短暂离心。置于37°C, 8hour;	
[0652]	(3) 加入RNase, 置于37°C, 15min, 充分消化体系中的RNA杂质;	
[0653]	(4) 加入蛋白酶K, 置于58°C, 15min, 消化Cas12j蛋白;	
[0654]	(5) 琼脂糖跑胶检测。	
[0655]	跑胶结果如图3所示, Cas12j.4, Cas12j.19以及Cas12j.22均能够有效的切割双链DNA, 但Cas12j.22的切割活性很弱。	
[0656]	三、Cas12j.4、Cas12j.19、Cas12j.22体外切割位点分析的结果	
[0657]	接下来我们对这三个具有DNA双链切割活性的蛋白的体外切割活性位点进行了分析。我们将上一步的切割后的条带进行回收, 并送公司进行Sanger测序。测序结果用seqman软件进行比对, 比对结果如附图4所示, 由峰图我们可以看出: Cas12j.4, Cas12j.19, Cas12j.22具有不同的切割方式, Cas12j.4和Cas12j.22的切割位点位于PAM末端18nt和25nt处, 切割后形成了7nt的粘性末端, 而Cas12j.19在距PAM远端25nt处有一个切割位点, 形成约1nt的末端。	
[0658]	实施例5. Cas12j.19在不同温度下体外酶切活性检测的结果	
[0659]	将Cas12j.19 (SEQ ID NO:17) 和导向RNA (SEQ ID NO:105) 在25°C下孵育15分钟, 形成RNA和蛋白的混合物, 通常称为RNP, 之后向反应体系中加入双链DNA (SEQ ID NO:106), 并分别置于设置的不同温度中, 设置的温度有: 17°C, 22°C, 27°C, 32°C, 37°C, 42°C, 47°C, 52°C, 62°C, 67°C, 72°C, 反应8h, 反应结束后加入RNase, 37°C消化15分钟RNA, 以及蛋白酶K, 58°C反应15分钟消化蛋白, 通过琼脂糖凝胶电泳检测其对于DNA消耗的结果。结果如图5所示, 结果显示, 发现Cas12j.19在27°C~42°C之间都具有双链DNA切割活性。	
[0660]	实施例6. Cas12j.19不同spacer长度对酶切活性影响的结果	
[0661]	由于Cas12j.19的切割位点在靶序列之外, 我们进一步测试了Cas12j.19导向RNA (SEQ ID NO:105) 含有靶位点的序列, 通常也称为spacer序列的长度对切割活性的影响。将导向RNA含有靶位点的序列进行截短 (14~28nt) 获得图6中所示的截短体, 将Cas12j.19和截短的导向RNA在25°C下孵育15分钟, 形成RNP, 之后向反应体系中加入双链DNA (SEQ ID NO:106), 37°C, 反应8h。反应结束后加入RNase, 37°C消化15分钟RNA, 以及蛋白酶K, 58°C反应15分钟消化蛋白, 通过琼脂糖凝胶电泳检测酶切结果。结果如图6所示, 结果显示, Cas12j.19发挥切割活性所需的spacer长度至少为14nt。	
[0662]	实施例7. Cas12j.19不同repeat长度对酶切活性影响的结果	
[0663]	同样我们测试了导向RNA的同向重复序列repeat的长度对于Cas12j.19双链DNA切割活性的影响。我们将导向RNA (SEQ ID NO:105) 中的同向重复序列进行截短为24~34nt获	

得图7中的截短体,将Cas12j.19与相应的不同repeat长度的导向RNA在25℃下孵育15分钟,形成RNP,之后向反应体系中加入双链DNA,37℃,反应8h。反应结束后加入RNase,37℃消化15分钟RNA,以及蛋白酶K,58℃反应15分钟消化蛋白,通过琼脂糖凝胶电泳检测酶切结果。结果如图7所示,结果显示,检测所需的最短的同向重复序列repeat长度为32nt。

[0664] 实施例8.Cas12j.19对于spacer错配容忍的结果

[0665] 导向RNA中含有靶位点的序列与原始的靶向序列的互补配对对于DNA的重组和切割具有重要的意义。将导向RNA (SEQ ID NO:105) 中含有靶序列部分依次进行点突变(即spacer 5'端开始的1,3,5,7,9,11,13,15,17位点的碱基),以获得图8中的突变体,从而与靶序列形成错配。通过将Cas12j.19与相应的含有突变位点的导向RNA在25℃下孵育15分钟,形成RNP,之后向反应体系中加入双链DNA (SEQ ID NO:106),37℃,反应8h。反应结束后加入RNase,37℃消化15分钟RNA,以及蛋白酶K,58℃反应15分钟消化蛋白,通过琼脂糖凝胶电泳检测酶切结果。结果如图8所示,结果显示,spacer序列5'端前5nt内,靶序列碱基的突变对于Cas12j.19双链DNA的切割具有重要的影响。另外,第13nt靶序列的错配对Cas12j.19双链DNA的切割活性影响很大。Cas12j.19严格的错配容忍度使其可能具有更低的脱靶率。

[0666] 尽管本发明的具体实施方式已经得到详细的描述,但本领域技术人员将理解:根据已经公布的所有教导,可以对细节进行各种修改和变动,并且这些改变均在本发明的保护范围之内。本发明的全部分为由所附权利要求及其任何等同物给出。

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 [0003] <120> CRISPR-Cas12j酶和系统
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[0336]	Cys Met Ile Gln Ser Arg Ile Ala Asp Thr Ser Arg Arg Leu Asn Ser
[0337]	565 570 575
[0338]	Phe Lys Tyr Gln Met Asn Lys Glu Gly Tyr Gln Asp Leu Ala Glu Ala
[0339]	580 585 590
[0340]	Leu Arg Leu Leu Asp Ala Met Asp Ser Tyr Asn Ser Leu Leu Glu Ser
[0341]	595 600 605
[0342]	Tyr Gln Arg Met His Leu Ser Pro Gly Glu Gln Ser Pro Lys Glu Ala
[0343]	610 615 620
[0344]	Lys Phe Asp Thr Lys Arg Ala Ser Phe Arg Asp Leu Leu Arg Arg Arg
[0345]	625 630 635 640
[0346]	Val Ala His Thr Ile Val Glu Tyr Phe Asp Asp Cys Asp Ile Val Phe
[0347]	645 650 655
[0348]	Phe Glu Asp Leu Asp Gly Pro Ser Asp Ser Asp Ser Arg Asn Asn Ala
[0349]	660 665 670
[0350]	Leu Val Lys Leu Leu Ser Pro Arg Thr Leu Leu Leu Tyr Ile Arg Gln
[0351]	675 680 685
[0352]	Ala Leu Glu Lys Arg Gly Ile Gly Met Val Glu Val Ala Lys Asp Gly
[0353]	690 695 700
[0354]	Thr Ser Gln Asn Asn Pro Ile Ser Gly His Val Gly Trp Arg Asn Lys
[0355]	705 710 715 720
[0356]	Gln Asn Lys Ser Glu Ile Tyr Phe Tyr Glu Asp Lys Glu Leu Leu Val
[0357]	725 730 735
[0358]	Met Asp Ala Asp Glu Val Gly Ala Met Asn Ile Leu Cys Arg Gly Leu
[0359]	740 745 750
[0360]	Asn His Ser Val Cys Pro Tyr Ser Phe Val Thr Lys Ala Pro Glu Lys
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[0362]	Lys Asn Asp Glu Lys Lys Glu Gly Asp Tyr Gly Lys Arg Val Lys Arg
[0363]	770 775 780
[0364]	Phe Leu Lys Asp Arg Tyr Gly Ser Ser Asn Val Arg Phe Leu Val Ala
[0365]	785 790 795 800
[0366]	Ser Met Gly Phe Val Thr Val Thr Thr Lys Arg Pro Lys Asp Ala Leu
[0367]	805 810 815
[0368]	Val Gly Lys Arg Leu Tyr Tyr His Gly Gly Glu Leu Val Thr His Asp
[0369]	820 825 830
[0370]	Leu His Asn Arg Met Lys Asp Glu Ile Lys Tyr Leu Val Glu Lys Glu
[0371]	835 840 845
[0372]	Val Leu Ala Arg Arg Val Ser Leu Ser Asp Ser Thr Ile Lys Ser Tyr
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 [0386] 20 25 30
 [0387] Met Lys Glu Ile Leu Pro Val Met Asn Asn Ile Val His Tyr Gln Asn
 [0388] 35 40 45
 [0389] Asn Leu Leu Lys Leu Met Leu Ile Leu Arg Gly Glu Lys Tyr Thr Leu
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 [0391] Asp Gly Gln Phe Phe Ser Gln Lys Asp Val Asp Arg Gln Phe Gly Asp
 [0392] 65 70 75 80
 [0393] Leu Cys Lys Glu His Asn Ile Lys Gly Ser Ile Cys Ser Leu Lys Glu
 [0394] 85 90 95
 [0395] Lys Ser Arg Lys Leu Tyr Glu Val Phe Ser Cys Tyr Ile Asp Lys Lys
 [0396] 100 105 110
 [0397] Gly Asn Leu Lys Thr Asn Ser Lys Ala Arg Ser Phe Ala Gly Val Leu
 [0398] 115 120 125
 [0399] Leu Asn Pro Lys Asp Val Lys Leu Pro Pro Gln Ile Asp Ser Ile Ser
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 [0401] Ser Phe Val Val Glu Leu Arg Ala Lys Gly Val Leu Pro Ile Lys His
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 [0403] Glu Gly Asn Tyr Leu Ser Gly His Pro Ser Leu Lys Tyr Ser Val Ala
 [0404] 165 170 175
 [0405] Gln Asn Val Leu Val Lys Leu Thr Ser Met Glu Lys Leu Gln Lys Ile
 [0406] 180 185 190
 [0407] Tyr Ser Asp Glu Lys Ala Gly Trp Glu Asn Ile Val Ser Glu Val Arg
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 [0409] Ser Asp Leu Pro Lys Ile Glu Arg Tyr Glu Arg Met Leu Leu Ser Ile
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 [0415] Leu Lys Gln Gly Met Arg Thr Tyr Phe Val Asn Met Leu Glu Ser Lys
 [0416] 260 265 270
 [0417] Lys Asp Tyr Arg Phe Glu Glu Ser Asp Arg Tyr Leu Phe Gly Tyr Ala
 [0418] 275 280 285
 [0419] Pro Glu Val Met Asn Leu Val Tyr His Asp Phe Arg Asp Leu Trp Gln

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[0421]	Gly Glu Asp Ile Ile Gly Ser Gln Ser Pro Glu Lys Lys Asp Arg Asp		
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[0423]	Tyr Val Asp Val Ile Phe Asn Tyr Phe Asn Trp Arg Lys Glu Ser Ile		
[0424]		325	330
[0425]	Asn Ile Ser Ser Phe Asp Ser Tyr Gly Lys Thr Ala Gln Ile Lys Leu		
[0426]		340	345
[0427]	Gly Asp Asn Tyr Val Pro Phe Ser Asn Phe Gln Tyr Asp Lys Ile Leu		
[0428]		355	360
[0429]	Asp Ala Trp Thr Leu Glu Ile Ala Asn Val Ser Gly Glu Gly Asp Asn		
[0430]	370	375	380
[0431]	His Lys Leu Val Ile Ala Arg Ser Pro Gln Phe Asp Ser His Ser Ser		
[0432]	385	390	395
[0433]	Val Lys Asp Ile Val Met Lys Asn Leu Lys Gly Lys Glu Ala Ser Lys		
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[0435]	Thr Thr Leu Glu Phe Arg Tyr Ser Gly Asp Ser Lys Lys Ser Thr Trp		
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[0437]	Tyr Arg Gly Thr Leu Lys Glu Pro Thr Leu Arg Tyr Ser Ser Ser Lys		
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[0439]	Asn Cys Leu Tyr Val Asp Phe Ala Leu Ser Asn His Ile Val Glu Gly		
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[0441]	Leu Ile Ser Asp Asn Leu Gly Ile Ser Asp Lys Met Tyr Lys Phe Arg		
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[0443]	Gly Glu Phe Met Lys Ala Ser Pro Ser Ser Gly Lys Gln Ser Asn Ser		
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[0445]	Ile Asn Leu Pro Ile Lys Lys Leu Arg Ala Met Gly Val Asp Phe Asn		
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[0447]	Leu Arg Arg Pro Phe Gln Ala Ser Ile Tyr Asp Val Glu Asn Lys Asn		
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[0449]	Gly Asn Leu Glu Phe Ser Phe Val Lys His Val Gln Ser Phe Ser Asn		
[0450]	530	535	540
[0451]	Glu Asn Asp Glu Glu Arg Ala Lys Glu Leu Leu Asn Ile Glu Arg Asn		
[0452]	545	550	555
[0453]	Ile Leu Ala Leu Lys Ile Leu Ile Trp Gln Thr Val Gly Tyr Val Thr		
[0454]		565	570
[0455]	Gly Lys Asn Asp Thr Ile Asp Gly Val Val Thr Arg Lys Asn Asn Ala		
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[0457]	Val Asp Ile Glu Lys Thr Leu Gly Ile Asn Met Lys Glu Tyr Met Ala		
[0458]		595	600
[0459]	Tyr Leu Asn Gln Phe Arg Ser Tyr Glu Asp Lys Asn Lys Ala Phe Met		
[0460]	610	615	620
[0461]	Asp Leu Arg Lys Arg Glu Tyr Ala Trp Ile Val Pro Pro Leu Ile Phe		

[0462]	625	630	635	640
[0463]	Gln Cys Arg Ser Arg Leu Ile Ser Phe Arg Ser Glu Tyr Phe Asn Thr			
[0464]		645	650	655
[0465]	Pro Lys Asp Glu Lys Ser His Tyr Cys Gln His Arg Asn Phe Val Asp			
[0466]		660	665	670
[0467]	Tyr Ser Thr Phe Leu Lys Lys Asn Val Val Lys Lys Met Met Glu Leu			
[0468]		675	680	685
[0469]	Arg Arg Ser Tyr Ser Thr Phe Gly Met Ser Ser Glu Gln Ser Ile Trp			
[0470]		690	695	700
[0471]	Val Thr Asn Asn Asp His Ala Lys Asp Gly Ser Lys Lys Asn Gly Asn			
[0472]	705	710	715	720
[0473]	Met Phe Asp Asp Asp Leu His Gln Trp Tyr Asn Gly Leu Val Arg Lys			
[0474]		725	730	735
[0475]	Cys Ser Ser Leu Ala Ser Ser Ile Ile Asn Val Ala Arg Asp Asn Gly			
[0476]		740	745	750
[0477]	Ala Ile Leu Val Phe Ile Glu Asp Leu Asp Cys His Pro Ser Ala Phe			
[0478]		755	760	765
[0479]	Asp Ser Glu Glu Asp Asn Ser Leu Lys Ser Ile Trp Gly Trp Gly Ser			
[0480]		770	775	780
[0481]	Ile Lys Ala Ser Leu Ala His Gln Ala Arg Lys His Asn Ile Ala Val			
[0482]	785	790	795	800
[0483]	Val Ala Asn Asp Pro His Leu Thr Ser Leu Val Ser Ser Thr Thr Gly			
[0484]		805	810	815
[0485]	Glu Leu Gly Ile Ala Lys Gly Arg Asp Val Leu Phe Phe Asp Ser Lys			
[0486]		820	825	830
[0487]	Gly Lys Leu Thr Ser Lys Val Asn Arg Asp Glu Asn Ala Ala Gln Asn			
[0488]		835	840	845
[0489]	Ile Ala Ile Arg Gly Phe Val Arg His Ser Asp Leu Arg Glu Phe Val			
[0490]		850	855	860
[0491]	Ala Glu Lys Ile Glu Glu Asn Arg Tyr Arg Val Val Val Asn Lys Thr			
[0492]	865	870	875	880
[0493]	His Lys Arg Lys Ala Gly Ala Ile Tyr Arg His Ile Gly Ser Thr Glu			
[0494]		885	890	895
[0495]	Cys Ile Met Ser Lys Gln Ala Asp Gly Ser Leu Lys Ile Asp Lys Thr			
[0496]		900	905	910
[0497]	Glu Leu Thr Pro Leu Glu Ile Lys Met Glu Lys Lys Asn Asp Lys Lys			
[0498]		915	920	925
[0499]	Met Tyr Val Ile Leu His Gly Lys Thr Trp Arg Leu Arg His Glu Leu			
[0500]		930	935	940
[0501]	Asn Glu Lys Leu Glu Lys Asp Leu Asp Asn His Leu Lys Ser Lys Ser			
[0502]	945	950	955	960
[0503]	Ser Val Ile Ser			

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 [0513] Thr Leu Arg Gly Lys Val Tyr Asn His Asp Thr Ala Met Glu Ala Phe
 [0514] 20 25 30
 [0515] Ala Pro Val Met Lys Gly Met Val Pro Tyr Ala Asn Asn Leu Met Arg
 [0516] 35 40 45
 [0517] Ile Leu Leu Thr Leu Arg Leu Glu Lys Tyr Thr Leu Asp Gly Ile His
 [0518] 50 55 60
 [0519] His Thr Lys Glu Glu Val Glu Lys Asp Leu Arg Gly Leu Met Lys Glu
 [0520] 65 70 75 80
 [0521] Tyr Gly Ile Asn Leu Ser Phe Ala Lys Phe Ser Glu Met Ala Gly Glu
 [0522] 85 90 95
 [0523] Val Tyr Arg Val Phe Val Cys Tyr Val Asp Ala Lys Gly Lys Leu Lys
 [0524] 100 105 110
 [0525] Val Asn Gly Lys Ala Arg Gly Phe Ala Asn Val Phe Phe Ser Glu Asp
 [0526] 115 120 125
 [0527] Asp Ala Thr Ile Pro Glu Asn Cys Pro Ser Met Glu Leu Leu Arg Lys
 [0528] 130 135 140
 [0529] Lys Gly Met Phe Pro Ile Leu Val Asp Gly Lys Pro Ile Ser Ser Ile
 [0530] 145 150 155 160
 [0531] Ser Arg Glu Lys Thr Pro Leu Lys Tyr Ser Val Ala Gln Asp Val Leu
 [0532] 165 170 175
 [0533] Thr Lys Leu Thr Ser Met Glu Glu Ile Ser Lys Glu Tyr Glu Lys Ala
 [0534] 180 185 190
 [0535] Lys Thr Asp Trp Glu Asn Glu Cys Gln Lys Val Ile Ser Gln Leu Pro
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 [0537] Leu Ile Gly Arg Tyr Glu Ala Leu Leu Thr Thr Ile Pro Leu Ile Pro
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 [0541] Trp Arg Asp Tyr Val Asn Glu Asp Gly Glu Leu Val Arg Gly Gly Met
 [0542] 245 250 255
 [0543] Lys Thr Tyr Phe Leu Asp Leu Leu Ser Lys Asp Thr Ser His Lys Phe
 [0544] 260 265 270
 [0545] Asn Glu Glu Glu Arg Tyr Leu Phe Gly Tyr Cys Pro Glu Phe Met Asn

[0546]	275	280	285
[0547]	Leu Ile Tyr His Asp Phe Arg Asp Leu Trp Ser Lys Glu Asp Ile Ile		
[0548]	290	295	300
[0549]	Gly Ser Gln Arg Lys Gly Lys Gly Leu Lys Gly Lys Asp Tyr Val Asp		
[0550]	305	310	315
[0551]	Val Ile Phe Asn Cys Phe His Trp Arg Arg Glu Ser Ile Asn Ile Ser		
[0552]		325	330
[0553]	Ser Phe Gly Asn Asn Asp Lys Val Met Asn Ile His Leu Gly Asp Asn		335
[0554]		340	345
[0555]	Phe Val Pro Phe Glu Leu Lys Ser Gln Asn Gly Ile Trp Glu Val His		350
[0556]		355	360
[0557]	Val Gln Asn Leu His Gly Gln Asn Asp Pro His Arg Val Ile Val Cys		365
[0558]		370	375
[0559]	Arg Cys Pro Gln Phe Asn Glu Asp Ser Ser Met Lys Met Val His Pro		380
[0560]		385	390
[0561]	Leu Ala Lys Asn Gly Glu Glu Ser Asp Lys Glu Asn Ile Glu Phe Arg		395
[0562]		405	410
[0563]	Tyr Ser Gly Asp Ser Lys Arg Glu Thr Trp Tyr Thr Gly Leu Leu Lys		415
[0564]		420	425
[0565]	Glu Pro Thr Leu Arg Tyr Asp Val Glu Arg Lys Ser Leu Tyr Val Asp		430
[0566]		435	440
[0567]	Phe Ile Leu Ser Asn His Arg Val Glu Gly Val Val Thr Asn Glu Tyr		445
[0568]		450	455
[0569]	Leu Lys Asp Pro Arg Asp Leu Phe Gly Val Arg Gly Tyr Phe Leu Ser		460
[0570]		465	470
[0571]	Ser Ser Val Ser Asn Pro Arg Gln Lys Asp Lys Thr Ser Leu Pro Asp		475
[0572]		485	490
[0573]	Gly Lys Phe Asn Val Met Gly Val Asp Leu Gly Leu Lys Cys Pro Tyr		495
[0574]		500	505
[0575]	Glu Cys Ala Ile Tyr Gly Ile Thr Val Lys Asn Gly Lys Met Gln His		510
[0576]		515	520
[0577]	Lys Trp Ser His Asn Val Ser Ala Glu Asp Asn Asn Asn Val Ser Glu		525
[0578]		530	535
[0579]	Arg Leu Ala Asn Leu Lys Lys Ile Asp Glu Lys Ile Leu Ala Thr Gln		540
[0580]		545	550
[0581]	Val Leu Ile Ser Leu Thr Lys Met Cys Val Val Lys Asp Glu Glu Ile		555
[0582]		565	570
[0583]	Pro Asp Ser Tyr Thr Leu Arg Glu His Arg Val Asp Ile Ala Lys Ser		575
[0584]		580	585
[0585]	Leu Asp Leu Asp Met Asp Lys Tyr Arg Arg Tyr Val Glu Lys Cys Lys		590
[0586]		595	600
[0587]	Lys Asn Pro Asp Lys Ile Gln Ala Leu Lys Asp Ile Arg Lys Ser Glu		605

[0588]	610	615	620
[0589]	Asn Asn Trp Ile Val Ala Glu Lys Ile Asn Glu Ile Arg Ser Leu Ile		
[0590]	625	630	635 640
[0591]	Ser Glu Ile Arg Ser Glu Tyr Tyr Ala Ser Lys Asp Lys Arg Asn Tyr		
[0592]		645	650 655
[0593]	Cys Arg Asn Leu Asn Gly Val Asp Leu Ser Val Phe Leu Lys Lys Lys		
[0594]		660	665 670
[0595]	Val Val Lys Asn Trp Ile Ser Leu Leu Arg Ser Phe Ser Thr Phe Gly		
[0596]		675	680 685
[0597]	Met Thr Pro Gln Glu Ser Ala Tyr Ile Arg Lys Asp Phe Ala Lys Asn		
[0598]	690	695	700
[0599]	Leu Ser Lys Trp Tyr Lys Gly Leu Val Arg Lys Cys Gly Ser Ile Ala		
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[0601]	Ala His Ile Val Asn Ile Ala Arg Asp Asn Lys Val Met Val Ile Phe		
[0602]		725	730 735
[0603]	Ile Glu Asp Leu Asp Ala Arg Thr Ser Ala Phe Asp Ser Lys Glu Asp		
[0604]		740	745 750
[0605]	Asn Glu Leu Lys Ile Leu Trp Gly Trp Gly Glu Ile Lys Lys Trp Ile		
[0606]		755	760 765
[0607]	Gly His Gln Ala Arg Lys His Asn Ile Ala Val Val Ala Val Asp Pro		
[0608]	770	775	780
[0609]	His Leu Thr Ser Leu Val Asn His Glu Ser Gly Leu Leu Gly Ile Ala		
[0610]	785	790	795 800
[0611]	Gly Ser Gly Asn Asp Arg Asn Ile Tyr Thr Phe Gln Lys Asn Lys Lys		
[0612]		805	810 815
[0613]	Tyr Val Val Ile Asn Arg Asp Asn Asn Ala Ala His Asn Ile Ala Leu		
[0614]		820	825 830
[0615]	Arg Gly Leu Ser Lys His Thr Asp Ile Arg Glu Phe Tyr Val Glu Gln		
[0616]		835	840 845
[0617]	Ile Asp Val Asp His Tyr Arg Leu Met Tyr Gly Pro Glu Ala Glu Asn		
[0618]	850	855	860
[0619]	Gly Lys Arg Arg Ser Gly Ala Ile Tyr Lys His Ile Gly Ser Thr Glu		
[0620]	865	870	875 880
[0621]	Cys Val Phe Ser Lys Gln Lys Asn Gly Thr Leu Lys Val Glu Lys Thr		
[0622]		885	890 895
[0623]	Ser Leu Thr Lys Asp Glu Lys Glu Met Pro Lys Ile Asn Gly Lys Gly		
[0624]		900	905 910
[0625]	Val Tyr Ala Ile Leu His Gly Asn Glu Trp Arg Leu Arg His Glu Leu		
[0626]		915	920 925
[0627]	Asn Glu Glu Leu Gly Ala Lys Leu Asp Gly Ile Ser Val Lys Arg Val		
[0628]	930	935	940
[0629]	Val Ser Glu Pro Asn Lys Val Lys Thr Ser Leu Val Lys Gly Ser Val		

[0630]	945	950	955	960
[0631]	Arg Ala			
[0632]	<210> 6			
[0633]	<211> 907			
[0634]	<212> PRT			
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[0641]	Glu Asn Lys Met Tyr Pro Asp Lys Asp Thr Asp Phe Pro Val Asn Ser			
[0642]		20	25	30
[0643]	Gln Phe Ser Arg Ser Ile Ser Ile Arg Ala Asn Val Asp Pro Lys Asp			
[0644]		35	40	45
[0645]	Leu Leu Val Leu Lys Arg Thr Phe Glu Glu Thr Thr Lys Ile Ser Asp			
[0646]		50	55	60
[0647]	Glu Leu Leu Ser Thr Leu Leu Met Leu Arg Gly Lys Asp Tyr Cys Leu			
[0648]	65	70	75	80
[0649]	Asp Asn Val Val Cys Lys Gly Glu Glu Val Leu Glu Asn Leu Tyr Lys			
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[0651]	Lys Leu Ser Lys Asn Ala Thr Val Asn Arg Asp Lys Phe Ile Ser Thr			
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[0653]	Ala Lys Ala Phe Tyr Glu Tyr Phe His Gly Cys Ser Tyr His Lys Gly			
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[0655]	Phe Lys Ser Phe Phe Phe Ser Ser Lys Glu Ile Asp Ser Ile Gln Ser			
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[0657]	Glu Lys Phe Gly Tyr Leu Arg Glu Ile Gly Leu Phe Pro Ile Lys Ile			
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[0659]	Asp Ala Gln Ile Ser Asn Asp Leu Gln Tyr Ser Ile Val Ala Ser Asn			
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[0661]	His Ala Lys Ile Lys Gly Phe Glu Lys Ile Asp Lys Glu Tyr Gln Ala			
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[0663]	Asn Lys Glu Lys Trp Asn Lys Thr Ile Gly Glu Ser Thr Leu Lys His			
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[0665]	Leu Asn Arg Tyr Gly Glu Met Leu Lys Gly Leu Ser Asp Leu Gly Thr			
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[0667]	Met Gly Asn Phe Asn Gly Lys Lys Tyr Asp Arg Phe Met Gly His Trp			
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[0669]	Arg Asn Glu Gln Lys Ile Pro Asp His Ile Ser Met Leu Asp Phe Phe			
[0670]		245	250	255
[0671]	Arg Lys Ile Tyr Gln Glu Lys Gly Lys Ser His Arg Phe Thr Ala Ile			

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[0673]	Asp Asn Phe Thr Tyr Gly Tyr Glu Ser Glu Phe Met Asn His Ile Tyr		
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[0675]	Leu Asn Phe Ser Asp Leu Trp Leu Lys Glu Asp Val Ile Gly Asp Glu		
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[0677]	Glu Tyr Val Ser Leu Ile Arg Gly Ala Tyr His Trp Gln Lys Asp Val		
[0678]	305	310	315
[0679]	Val Gly Ile Ala Ser Phe Ser Gly Tyr Asn Lys Tyr Glu Lys Leu Phe		
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[0681]	Met Gly Asp Asn Lys Ile Asn Tyr Ala Leu Asp Phe Ser Asn Lys Asp		
[0682]	340	345	350
[0683]	Gln Trp Leu Met Lys Phe Asn Asn Val Ile Ser Lys Glu Pro Glu Thr		
[0684]	355	360	365
[0685]	Ile Thr Leu Arg Leu Cys Lys Asn Gly Tyr Phe Asn Asn Leu Ser Val		
[0686]	370	375	380
[0687]	Leu Glu Lys Asn Asp Glu Asn Gly Arg Tyr Lys Ile Arg Phe Ser Thr		
[0688]	385	390	395
[0689]	Glu Lys Gln Gly Lys Tyr Phe Tyr Glu Ala Phe Ile Arg Glu Pro Phe		
[0690]	405	410	415
[0691]	Leu Arg Tyr Asn Lys Asp Asn Asp Lys Ile Tyr Val His Phe Cys Leu		
[0692]	420	425	430
[0693]	Ser Glu Glu Ile Lys Glu Asn Cys Pro Asn His Leu Asp Thr Arg Ser		
[0694]	435	440	445
[0695]	Asp Lys Tyr Leu Phe Lys Ser Ala Leu Leu Thr Asn Ser Arg Gln Lys		
[0696]	450	455	460
[0697]	Leu Gly Lys Leu His Tyr Arg Asp Phe His Ile Val Gly Val Asp Leu		
[0698]	465	470	475
[0699]	Gly Ile Asn Pro Val Ala Lys Ile Thr Val Cys Lys Val His Val Asp		
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[0701]	Lys Asn Glu Asn Leu Lys Ile Thr Lys Ile Ile Thr Glu Glu Thr Arg		
[0702]	500	505	510
[0703]	Lys Asn Ile Asp Thr Asn Tyr Leu Asp Gln Leu Asn Leu Leu Tyr Lys		
[0704]	515	520	525
[0705]	Lys Ile Val Ser Leu Lys Arg Leu Ile Arg Ala Thr Val Ala Phe Lys		
[0706]	530	535	540
[0707]	Lys Asp Gly Glu Glu Ile Pro Lys Met Phe Lys Met Gly Lys Lys Ser		
[0708]	545	550	555
[0709]	Pro Tyr Phe Leu Asn Trp Thr Glu Val Leu Asn Val Asn Tyr Asp Asp		
[0710]	565	570	575
[0711]	Tyr Ile Lys Glu Ile Ser Thr Phe Ser Val Asp Arg Leu Ser Gly Leu		
[0712]	580	585	590
[0713]	Thr Leu Pro Met Gln Trp Ala Arg Ser Gln Asn Lys Trp Val Val Lys		

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[0715]	Asp Leu Thr Lys Met Val Arg Lys Gly Ile Ser Asp Leu Ile Tyr Ala		
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[0717]	Arg Tyr Phe Asn Cys Ser Asp Lys Thr Gln Tyr Val Thr Glu Asn Asn		
[0718]	625	630	635
[0719]	Ala Val Asp Ile Thr Thr Phe Lys Lys His Asp Ile Ile Ser Glu Ile		
[0720]	645	650	655
[0721]	Ile Gly Leu Gln Lys Met Phe Ser Gly Gly Gly Lys Asp Val Ala Lys		
[0722]	660	665	670
[0723]	Lys Asp Tyr Leu Tyr Leu Arg Gly Leu Arg Lys His Ile Gly Asn Tyr		
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[0725]	Thr Ala Ser Ala Ile Val Ser Ile Ala Gln Lys Tyr Asn Ala Val Phe		
[0726]	690	695	700
[0727]	Ile Phe Ile Glu Asp Leu Asp Leu Lys Ile Ser Gly Met Asn Gly Lys		
[0728]	705	710	715
[0729]	Lys Glu Asn Lys Val Lys Ile Leu Trp Gly Val Gly Gln Leu Lys Lys		
[0730]	725	730	735
[0731]	Arg Leu Ser Glu Lys Ala Glu Lys Phe Gly Ile Gly Ile Val Pro Val		
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[0734]	755	760	765
[0735]	Tyr Arg Asn Pro Thr Asn Lys Lys Glu Leu Tyr Val Lys Arg Asp Asp		
[0736]	770	775	780
[0737]	Lys Ile Glu Ile Leu Asp Ala Asp Glu Thr Ala Ser Tyr Asn Val Ala		
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[0739]	Leu Arg Gly Leu Gly His His Ala Asn Leu Ile Gln Phe Arg Ala Asp		
[0740]	805	810	815
[0741]	Lys Met Pro Asn Gly Cys Phe Arg Val Met Pro Asp Arg Lys Tyr Lys		
[0742]	820	825	830
[0743]	Gln Gly Ala Leu Tyr Gly Tyr Leu Asn Ser Thr Ala Val Leu Phe Lys		
[0744]	835	840	845
[0745]	Asp Lys Gly Asp Gly Val Leu Thr Ile His Lys Ser Lys Leu Thr Lys		
[0746]	850	855	860
[0747]	Lys Glu Arg Asp Ser Arg Pro Ile Lys Gly Lys Lys Thr Phe Val Val		
[0748]	865	870	875
[0749]	Lys Asn Gly Lys Arg Trp Ile Leu Arg His Val Leu Asp Glu Glu Val		
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[0764]	Phe Lys Val Ile Thr Lys Asp Ala Asp Phe Leu Leu Lys Asn Leu Leu	
[0765]		35 40 45
[0766]	Ile Met Glu Gly Gly Glu Tyr Met Leu Asn Arg Glu Ile His Asn Gly	
[0767]		50 55 60
[0768]	Asp Lys Glu Phe Asp Lys Ile Ile Ser Lys Leu Gly Leu Ser Lys Lys	
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[0770]	Glu Lys Glu Asn Leu Lys Met Lys Cys Lys Asp Phe Phe Phe Asp Phe	
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[0772]	Val Lys Leu Gln Asn Gly Arg Ser Leu Ala Asn Ile Leu Phe Glu Thr	
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[0774]	Lys Gly Thr Thr Leu Ile Gly Cys Gly Lys Asp Lys Lys Gly Glu Lys	
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[0776]	Val Asp Gly Glu Tyr Pro Thr Ile Tyr His Asp His Glu Thr Leu Arg	
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[0778]	Ser Thr Gly Leu Leu Pro Leu Lys Phe Ser Lys Asn Ile Asp Asp Val	
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[0780]	Asp Tyr Lys Tyr Leu Ile Cys Tyr Leu Val His Asn Val Leu Ser Ser	
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[0782]	Phe Ile Glu Lys Arg Asp Ala Tyr Asn Asp Asn Lys Lys Glu Trp Glu	
[0783]		180 185 190
[0784]	Ser Lys Leu Ser Asn Ser Asn Leu Pro Gln Leu Glu Arg Met Ser Glu	
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[0786]	Phe Leu Asn Gly Ile Asn His Leu Gly Asn Ile Ile Gly Trp Asn Gly	
[0787]		210 215 220
[0788]	Lys Lys Tyr Ile Gly Phe Ile Lys Lys Trp Thr Asp Glu Glu Ser Ser	
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[0790]	Met Tyr Asp Phe Phe Val Gln Lys Leu Gln Asp Asn Pro Lys Tyr Lys	
[0791]		245 250 255
[0792]	Phe Gly Lys Lys Asp Gln Phe Leu Tyr Gly Tyr Glu Pro Glu Phe Leu	
[0793]		260 265 270
[0794]	Asn Tyr Leu Phe His Asp Phe Arg Asp Leu Trp His Pro Asp Asn Leu	
[0795]		275 280 285
[0796]	Ile Gly Lys Asp Glu Tyr Val Asp Leu Ile Ser Gly Lys Asn Asn Thr	
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[0798]	Asp Ala Glu Thr Ala Asn Lys Gly Ala Tyr His Trp Leu Lys Asp Phe
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[0800]	Ile Asn Ile Ser Ser Phe Asp Ala Tyr Gly Lys Met Ala Thr Ile Gly
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[0802]	Met Gly Asn Asn Leu Ile Asn Tyr Ser Met Asn Ile Asp Lys Asp Gly
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[0804]	Lys Ile Ile Val Asn Met Asp Asn Ile Phe Asp Arg Ser Lys Pro Ile
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[0806]	Val Phe Asn Val Tyr Arg Asn Ser Tyr Phe Arg Asn Phe Lys Ile Ile
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[0808]	Glu Ser Asp Asp Lys Lys Gly Ile Tyr Lys Val Glu Phe Ser Thr Ser
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[0812]	Phe Ala Thr Lys Gly Gly Thr Ile Lys Ile Asp Phe Pro Ile Ser Asp
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[0814]	Lys Arg Ile Lys Gly Gly Arg Glu Met Asn Thr Asp Leu Met Trp Phe
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[0816]	Leu Asn Arg Ala Ser Pro Cys Ser Thr Lys Asn Lys Glu Val Asn Ser
[0817]	450 455 460
[0818]	Phe Ile Gly Lys Asn Phe Val Gly Leu Ala Ile Asp Arg Gly Ile Asn
[0819]	465 470 475 480
[0820]	Pro Leu Met Ala Trp Tyr Val Ala Glu Trp Thr Tyr Asp Lys Asp Gly
[0821]	485 490 495
[0822]	Lys Ala Lys Ile Val Arg Ser Ile Ala Asn Gly Arg Val Asp Ser Gly
[0823]	500 505 510
[0824]	His Asn Glu Ser Glu Val Lys Phe Val Arg Glu Thr Thr Asn Arg Ile
[0825]	515 520 525
[0826]	Val Gly Ile Lys Ser Leu Val Trp Asn Thr Val Lys Tyr Arg Thr Gly
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[0828]	Gly Ser Glu Gly Ile Asp Arg Cys Arg Lys Ser Gln Asn Gly Gln Val
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[0830]	Asp Leu Phe Glu Met Phe Asp Ile Asp Tyr Asn Asn Tyr Leu Lys Glu
[0831]	565 570 575
[0832]	Val Asn Asn Leu Pro Tyr Asp Pro Asn Ser Glu Arg Ser Ile Ile Gln
[0833]	580 585 590
[0834]	Thr Trp Val Ser Ser Pro Trp Lys Val Lys Asp Leu Val Lys Asp Ala
[0835]	595 600 605
[0836]	Lys Asn Arg Met Val Gln Ile Lys Thr Gln Tyr His Asn Ala Lys Asp
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[0838]	Lys Glu Lys Tyr Ile Thr Thr Gln Asn Arg Ala Gly Phe Tyr Asp Phe
[0839]	625 630 635 640

[0840]	Leu Lys Ile Glu Met Glu Lys Gln Phe Thr Ser Leu Gln Arg Met Phe
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[0842]	Ser Gly Gly Gln Lys Asp Ile Cys Lys Asn Asn Glu Glu Tyr Arg Arg
[0843]	660 665 670
[0844]	Gly Leu Arg Arg Arg Ile Asn Leu Tyr Thr Ser Ser Val Ile Met Ser
[0845]	675 680 685
[0846]	Leu Ala Arg Lys Phe Asn Val Asp Cys Ile Phe Leu Glu Asp Leu Asp
[0847]	690 695 700
[0848]	Ser Ser Lys Ser Ser Trp Asp Asp Ala Lys Lys Asn Ser Leu Lys Asp
[0849]	705 710 715 720
[0850]	Leu Trp Ser Thr Gly Gly Ala Asp Asp Ile Leu Gly Lys Met Ala Asn
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[0852]	Lys Tyr Lys Tyr Pro Ile Val Lys Val Asn Ser His Leu Thr Ser Leu
[0853]	740 745 750
[0854]	Val Asp Asn Lys Thr Gly Lys Ile Gly Tyr Arg Asp Pro Lys Lys Lys
[0855]	755 760 765
[0856]	Ser Asn Leu Tyr Val Glu Arg Gly Lys Lys Ile Glu Ile Ile Asp Ser
[0857]	770 775 780
[0858]	Asp Glu Asn Ala Ala Ile Asn Ile Leu Lys Arg Gly Ile Ser Lys His
[0859]	785 790 795 800
[0860]	Ile Asp Ile Arg Glu Phe Phe Ala Glu Lys Ile Glu Val Ser Gly Lys
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[0862]	Thr Leu Tyr Arg Ile Ser Asn Lys Leu Gly Lys Gln Arg Met Gly Ser
[0863]	820 825 830
[0864]	Leu Tyr Tyr Leu Glu Gly Asn Lys Glu Ile Leu Phe Gly Leu Gly Lys
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[0867]	850 855 860
[0868]	Leu Ala Pro Arg Ile Ala Glu Lys Lys Ser Thr Tyr Leu Ile Met Asn
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[0870]	Gly Ser Lys Trp Met Phe Arg His Glu Ala Lys Lys Ile Val Glu Thr
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[0885]	Arg Asn Gly Phe Ser Asp Ile Gly Val Asp Pro Ser Leu Val Ser Tyr			
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[0887]	Ala Ser Lys Phe Leu Asp Ser Met Phe Ile Cys Phe Ser Arg Val Lys			
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[0889]	Gly Glu Lys Asn Phe Lys Ala Lys Asn Val Arg Lys Asn Met Ser Ser			
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[0891]	Ala Glu Lys Lys Ala Gln Lys Lys Lys Glu Tyr Gln Glu Tyr Tyr Gln			
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[0893]	Gly Val Met Ala Gln Gln Asp Ala Tyr Ala Gln Leu Leu Ser Asp Pro			
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[0895]	Thr Gln Glu Asn Leu Asp Lys Leu Asn Glu Leu Ile Ser Met Ser Val			
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[0897]	Asn Gly Ser Leu Val Glu Asp Phe Phe Pro Ala Leu Lys Asn Met Ile			
[0898]		130	135	140
[0899]	Gln Lys Ala Asp Tyr Ser Ile Asp Lys Lys Gly Leu Leu Asp Phe Ser			
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[0901]	Cys Cys Met Met Asp Arg Tyr Glu Asp Arg Ser Leu Thr Arg Ala Ile			
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[0903]	Ser Ile Ser Ala Phe Asn Ile His Ser Gly Gly Leu Arg Lys Ala Leu			
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[0905]	Ser Asp Ile Ser Glu Lys Val Gln Asp Leu Ser Asn Thr Leu Leu Ile			
[0906]		195	200	205
[0907]	Arg Ile Leu Tyr Met Lys Gly Glu Glu Leu Ser Ile Asp Gly Glu Lys			
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[0909]	Ile Ser Lys Glu Glu Val Gln Arg Gln Leu Lys Ala Asp Tyr Glu Glu			
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[0911]	His Lys Glu Tyr Phe Glu Asp Phe Glu Asp Phe Ala Lys Lys Cys Arg			
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[0913]	Phe Phe Tyr Asn Lys Phe Ser Lys Lys Lys Lys Thr Arg Gly Phe Gly			
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[0915]	Thr Tyr Phe Phe Gly Asp Lys Lys Lys Glu Ile Ser Ser Ala Glu Tyr			
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[0917]	Lys Ala His Lys Glu Leu Arg Asp Ser Gly Tyr Leu Trp Phe Asp Ile			
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[0919]	Gly Trp Ser Glu Ser Ser Asp Phe Lys Tyr Val Ile Val Gly Asn Val			
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[0921]	Ser Gly Lys Leu Lys Ser Phe Glu Glu Thr Ser Glu Glu Tyr Gln Lys			
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[0923]	Ser Lys Asn Cys Trp Glu Ala Glu Arg Val Lys Leu Tyr Glu Gln Asp			

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[0925]	Ser Asp Phe Val Leu Phe Val Glu Asp Met Ile Glu Ser Lys Tyr Gly					
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[0927]	Pro Ile Glu Lys Met Lys Leu Arg Thr Phe Lys Thr Ile Val Lys Lys					
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[0929]	Leu Asp Lys Glu Phe Gly Lys Arg Gly Asp Lys Thr Pro Ser Ile His					
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[0931]	Asp Tyr Phe Glu Ser Leu Asp Pro Asn His Thr Phe Ser Gln Ser Glu					
[0932]		405		410		415
[0933]	Gln Phe Met Tyr Gly Leu Asp Val Thr Leu Met Gln Phe Leu Phe Asn					
[0934]		420		425		430
[0935]	Asn Lys Lys Gln Phe Tyr Lys Leu Cys Lys Asp His Asp Gly Lys Arg					
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[0937]	Thr Phe Ala Lys Val Val Glu Glu Ser Tyr His Trp Gly Lys Asn Ser					
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[0939]	Ile Asn Val Ser Thr Phe Gln Asn Ser Thr Ser Ile Leu Leu Gly Gly					
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[0941]	Asn Tyr Leu Asn Tyr Ser Met Ser Ile Glu Gly Glu Gly Leu Val Ile					
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[0943]	Lys Phe Asp Asn Pro Leu Ser Gly Lys Glu Val His Phe Val Val Cys					
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[0945]	Asn Asn Lys Tyr Leu Ser Asp Leu Glu Ile Leu Ser Gly Asn Pro Asn					
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[0947]	Arg Lys Asp Asn Asn Tyr Thr Ile Ser Tyr Ser Thr Gly Gly Lys Ala					
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[0949]	Arg Phe Ile Ala Lys Ser Lys Glu Pro Arg Ile Phe Phe Asn Arg Lys					
[0950]		545		550		555
[0951]	Thr Lys Lys Trp Glu Ile Ala Phe Gln Leu Ser Asp Val Ser Pro Leu					
[0952]		565		570		575
[0953]	Asn Gly Lys Phe Gly Lys Gln Gly Glu Phe Leu Ser Asn Leu Arg Lys					
[0954]		580		585		590
[0955]	Phe Val Tyr Asn His Val Ala Lys Ser Pro Ser Lys Leu Asn Ile Ser					
[0956]		595		600		605
[0957]	Asp Asn Asn Cys Arg Ala Val Ala Tyr Asp Leu Gly Ile Arg Asn Val					
[0958]		610		615		620
[0959]	Gly Ala Trp Ser Ser Phe Asp Phe Ser Tyr Lys Asp Gly Val Leu Gly					
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[0961]	Gly Tyr Lys Tyr Leu Thr Ser Gly Ser Leu Arg Ser Lys Ser Glu Ser					
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[0963]	Ser Glu Met Asp Gln Gly Tyr Tyr Phe Val Leu Asn Leu Lys Lys Ile					
[0964]		660		665		670
[0965]	Val Lys Leu Ile Pro Val Val Lys Lys Ser Ile Ile Asp Asp Pro Glu					

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[0969]	Gly Leu Gly Asn Ile Gly Lys Leu Asp Ile Ala Ser Arg Lys Ala Val		
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[0971]	Gln Ser Phe His Asn Cys Ile Gln Gln Ile Asn Tyr Tyr Val Asp Thr		
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[0973]	Tyr Ala Asp His Ile Asp Lys Ile Ser Ala Lys Asp Phe Val Asp Asp		735
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[0975]	Ile Asp Gly Ile Lys Val Leu Asp Glu Asp Asp Pro Tyr Val Val Lys		750
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[0977]	Ile Leu Ser His Leu Pro Glu Asp Val Glu Gly Asn Gln Asp Asp Ile		765
[0978]		770	775
[0979]	Leu Asn Ile Ser Leu Leu Lys Trp Lys Thr Ser Asn Ala Gln Phe Val		780
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[0981]	Pro Pro Leu Ile Gln Glu Ala Lys Ala Ile Met Ser Arg Ile Lys Arg		795
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[0983]	Glu Asn Leu Asp Asn Ile Arg Gly Lys Lys Thr Gln Val Val Thr Gln		815
[0984]		820	825
[0985]	Lys Thr Phe His Lys Ile Lys Phe Ala Lys Ala Leu Leu Ser Leu Met		830
[0986]		835	840
[0987]	Lys Ser Trp Ser Ser Ile Gly Thr Val Arg Val Val Lys Thr Asp Gln		845
[0988]		850	855
[0989]	Ile Tyr Gly Lys Lys Ile Trp Asp Tyr Ile Asn Gly Leu Arg Arg Asn		860
[0990]		865	870
[0991]	Val Leu Thr Tyr Leu Ser Ser Ala Ile Val Asn Asn Ala Leu Asp Leu		875
[0992]		885	890
[0993]	Gly Ala His Met Ile Ile Leu Glu Asp Leu Asp Ser Ser Val Ser Lys		895
[0994]		900	905
[0995]	Tyr Arg Glu Lys Asp Lys Asn Ala Ile Gln Ser Leu Trp Gly Ser Gly		910
[0996]		915	920
[0997]	Glu Leu Lys Lys Arg Ile Glu Glu Lys Ala Glu Lys His Arg Val Val		925
[0998]		930	935
[0999]	Val Gln Tyr Val Ser Pro Tyr Leu Thr Ser Gln Leu Asp Asn Glu Thr		940
[1000]		945	950
[1001]	Lys Asp Ile Gly Tyr Arg Lys Gly Gly Arg Leu Tyr Val Val Arg Asn		955
[1002]		965	970
[1003]	Gly Lys Ile Lys Ser Ile Asp Ala Asp Ile Asn Ala Ser Lys Asn Ile		975
[1004]		980	985
[1005]	Gly Glu Arg Phe Phe Asp Arg Asp Leu Ile Gln Thr Leu Ser Gly Val		990
[1006]		995	1000
[1007]	Val Val Glu Asp Gln Ser Thr Val Tyr Ile Leu Gln Lys Arg Asn		1005

[1008]	1010	1015	1020
[1009]	Val Ser Ser Asp Asn Arg Lys Arg Phe Tyr Lys Lys Phe Leu Glu		
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[1011]	Asp Val Gly Gly Lys Ser Lys Lys Asp Ala Val Leu Lys Met Gly		
[1012]	1040	1045	1050
[1013]	Asp His Gly Glu Leu Glu Val Glu Arg Leu Ile Asp Gly Lys Lys		
[1014]	1055	1060	1065
[1015]	Leu Asp Ile Asp Gly Lys Lys Ile Leu Val Asp Gly Glu Lys Val		
[1016]	1070	1075	1080
[1017]	Pro Phe Arg Asn Thr Ser Val Tyr Tyr Ser Pro Lys Lys Lys Lys		
[1018]	1085	1090	1095
[1019]	Trp Val Ser Lys Glu Leu Arg Cys Asn His Ile Lys Leu Thr Val		
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[1021]	Glu Glu Gln Asp Ile Lys		
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[1034]	Lys Leu Asn Gln Lys Glu Val Lys Lys Gln Ile Glu Arg Ser Glu Tyr		
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[1036]	Ile Arg Ser Asn Cys Gly Tyr Ile Asn Ile Glu Arg Pro Gln Lys Ser		
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[1038]	Leu Ser Phe Leu Ser Tyr Ser Thr Ile Lys Ser Ala Cys Met Ser Val		
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[1040]	Asn Phe Arg Ala Phe Gln Asn Pro Ile Asn Asp Tyr Glu Thr Ala Ile		
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[1042]	Cys Asn Gly Ile Asn Glu Cys Glu Arg Phe Phe Tyr Gln Gln Ile Asp		
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[1044]	Ser Ile Tyr Met Ser Gln Ile Ile Glu Gln Leu Phe Asp Phe Tyr Ile		
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[1046]	Ala Ser Arg Gln His Asp Met Phe Ile Asn Asn Thr Val Val Pro Tyr		
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[1048]	Asp Val Asn Lys Leu Lys Ser Tyr Tyr Thr Ala Asn Glu Lys Tyr Ser		
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			155
			160

[1050]	Phe Glu Gln Phe Cys Asp Asp Ile Lys Glu Phe Thr Asn Lys Gly Phe
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[1052]	Thr Ser Gly Gly Val Ser Cys Ile Leu Asn Leu Phe Tyr Lys Gly Ser
[1053]	180 185 190
[1054]	Val Lys Asp Ser Lys Asn Lys Lys Asp Tyr Ile Lys Ser Val Lys Arg
[1055]	195 200 205
[1056]	Leu Glu Thr Asn Gly Leu Phe Lys Lys Leu Asn Ile Phe Glu Lys Asn
[1057]	210 215 220
[1058]	Gly Ile Ser Lys Tyr Phe Ala Ala Ser Thr Leu Ser Thr Phe Phe Ala
[1059]	225 230 235 240
[1060]	Thr Ile Ser Ser Trp Lys Lys Gln Asn Asp Asp Trp Thr Gly Val Ala
[1061]	245 250 255
[1062]	Lys Asp Gly Thr Ser Leu Leu Ser Lys Leu Glu Asn Lys Thr Ile Thr
[1063]	260 265 270
[1064]	Leu Gln Ser Ile Ile Lys His His Arg Val Ile Asn Glu Leu Ala Val
[1065]	275 280 285
[1066]	Leu Ile Val Lys Ala Tyr Lys Asp Pro Val Lys Thr Leu Asn Asn Leu
[1067]	290 295 300
[1068]	Phe Glu Glu Arg Ser Asp Asn Asn Asn Asp Phe Lys Tyr Thr Cys Ser
[1069]	305 310 315 320
[1070]	Asp Asp Glu Asp Lys Tyr Pro Met Tyr Ile Lys Arg Glu Ile Ala Glu
[1071]	325 330 335
[1072]	Phe Val Lys Lys His Lys Thr Val Trp Glu Glu Ile Arg Tyr Phe Asp
[1073]	340 345 350
[1074]	Glu Ser Asp Thr Lys Lys Lys Lys Arg Asp Lys Lys Glu Ser Ser Ser
[1075]	355 360 365
[1076]	Asp Asp Lys Ser Tyr Leu Cys Cys Gly Asp Ser Trp Asp Tyr Leu Lys
[1077]	370 375 380
[1078]	Thr Trp Val Arg Leu Tyr Gly Glu Tyr Tyr Phe Phe Asp Asn Ala Leu
[1079]	385 390 395 400
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[1084]	Tyr Lys Ile Gly Lys Val Glu Val Val Glu Arg Asn Asn Gln Arg Phe
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[1086]	Leu Leu Val Tyr Val Ser Val Pro Glu Met Glu Asn Tyr Ile Ile Ile
[1087]	450 455 460
[1088]	Pro Leu Gln Leu Asn Lys Tyr Phe Gly Asn Phe Gln Cys Lys Ile Phe
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[1091]	485 490 495

[1092]	Leu Lys Asn Asn Lys Pro Gln Pro Ser Pro Asn Ile Ser Val Arg Ile
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[1094]	Asn Glu Phe His Phe Ala Leu Arg Ser Tyr Arg Lys Gln Gln Ile Ser
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[1106]	Tyr Asn Ile Lys Pro Leu Gln Thr Gly Lys Pro Ala Thr Asp Trp Val
[1107]	610 615 620
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[1109]	625 630 635 640
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[1111]	645 650 655
[1112]	Ser Ser Asn Glu Ile Tyr Ser Ile Ala Thr Leu Thr Phe Arg Asn Ala
[1113]	660 665 670
[1114]	Asp Gly Lys Leu Glu Asn Arg Ser Phe Ser Arg Phe Tyr His Glu Leu
[1115]	675 680 685
[1116]	Asn Asn Thr Leu Asn Ile Ile Glu Gln Ile Lys Gly Thr Phe Asn Phe
[1117]	690 695 700
[1118]	Ile His Ser Ile Asn Thr Gln Phe Lys Glu Ile Lys Ala Leu Lys Thr
[1119]	705 710 715 720
[1120]	Thr Glu Glu Phe Ser Ser Tyr Val Ser Thr Leu Thr Trp Asp Gln Phe
[1121]	725 730 735
[1122]	Ile Glu Asp Ser Arg Lys Thr Ala Arg Tyr Ser Lys Tyr Trp Ile His
[1123]	740 745 750
[1124]	Ile Ile Asn Glu Asn Pro Lys Arg Arg Thr Ile Ala Thr Leu Asn Glu
[1125]	755 760 765
[1126]	Thr Leu Lys Leu Val Asp Glu Lys His Arg Phe Thr Val Thr Ile Gln
[1127]	770 775 780
[1128]	Glu Ile Phe Asp Leu Val Lys Tyr Cys Gln Gln His Gly Tyr Tyr Pro
[1129]	785 790 795 800
[1130]	Lys Ser Asn Val Met Ser Lys Leu Arg Asn Leu Ala Ile Lys Leu Ile
[1131]	805 810 815
[1132]	Asn Asp Leu Ile Arg Tyr Gln Lys Ile Gly Ile His Ser Cys Tyr Leu
[1133]	820 825 830

[1134]	Asp Phe Cys Val Leu Ile Lys Asn His Ile Ala Leu Leu Asn Ser Ser
[1135]	835 840 845
[1136]	Thr Ala Phe Ile Ile Asn Phe Ser Arg Asn Lys Glu Asn Ile Ile Arg
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[1139]	865 870 875 880
[1140]	Arg Arg Gln Met Ile Lys Asn Leu Cys Ser Gln Ile Leu Lys Ile Ala
[1141]	885 890 895
[1142]	Ala Lys Asn Lys Val His Ile Val Val Val Glu Lys Leu Asn Asn Met
[1143]	900 905 910
[1144]	Arg Thr Asn Asn Arg Asn Asn Glu Asp Lys Asn Asn Met Ile Asp Leu
[1145]	915 920 925
[1146]	Leu Ala Thr Gly Gln Phe Arg Lys Gln Leu Ser Asp Gln Ala Lys Trp
[1147]	930 935 940
[1148]	Tyr Gly Ile Ala Val Val Asp Thr Ala Glu Tyr Asn Thr Ser Lys Val
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[1153]	980 985 990
[1154]	Lys Lys Ala Ser Glu Asn Ile Leu Leu Ala Phe Val Thr Gln Ser Leu
[1155]	995 1000 1005
[1156]	Leu Leu Asn His Leu Lys Val Leu Ile Thr Glu Asp Gly Lys Thr
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[1158]	Ala Val Ile Asp Leu Ser Glu Arg Thr Thr Glu Pro Gln Lys Ile
[1159]	1025 1030 1035
[1160]	Arg Ser Lys Ile Trp Thr Asn Ser Asp Val Gln Lys Ile Ile Phe
[1161]	1040 1045 1050
[1162]	Cys Lys Gln Glu Asn Gly Ser Tyr Val Leu Lys Lys Gly Ser Thr
[1163]	1055 1060 1065
[1164]	Asp Ile Lys Glu Lys Met His Lys Ala Val Leu His Arg His Gly
[1165]	1070 1075 1080
[1166]	Ser Leu Trp Tyr Asp Tyr Leu Asn His Lys Asn Met Ile Glu Asp
[1167]	1085 1090 1095
[1168]	Ile Lys Asn Leu His Leu Ser Asn Cys Ser Leu Thr Thr Ser Thr
[1169]	1100 1105 1110
[1170]	Asn Ser Asp Val Ile Asn Ser His Ser Gly Ser Ser Arg Ser Leu
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[1172]	Asp Lys Thr Lys Thr Tyr Ala
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 [1185] His Ser Asn Leu His Asp Ala Ala Glu Ile Gly Ile Asn Glu Cys Arg
 [1186] 35 40 45
 [1187] Trp Trp Ile Gly Asp Gly Glu Ile Tyr Glu Arg Asp Pro Ala Cys Arg
 [1188] 50 55 60
 [1189] Ser Ile Lys Lys Gly Asn Asp Ile Arg Thr Val Thr Ser Glu Lys Ile
 [1190] 65 70 75 80
 [1191] Lys Glu Leu Trp Thr Lys His Thr Asp His Ser Val Pro Leu Val Asp
 [1192] 85 90 95
 [1193] Phe Ile Asp Met Leu Lys Phe Val Ala Gln Cys Ala Ile Tyr Gly Asp
 [1194] 100 105 110
 [1195] Ser Arg Ala Leu Ala Ser Thr Leu Phe Gly Lys Ser Lys Ala Glu Thr
 [1196] 115 120 125
 [1197] Arg Gly Val Ser Thr Glu Asp Met Thr Val Ile Arg Ala Trp Ile Ala
 [1198] 130 135 140
 [1199] Glu Thr Asp Ala Val Leu Ala Ser Gly Leu Ser Pro Lys Lys Lys Lys
 [1200] 145 150 155 160
 [1201] Lys Lys Glu Lys Glu Ala Gly Lys Lys Glu Arg Lys Pro Asp Val Lys
 [1202] 165 170 175
 [1203] Met Glu Met Cys Arg Arg Ile Arg Cys Thr Met Val Gln Cys Gly Tyr
 [1204] 180 185 190
 [1205] Phe Arg Arg Phe Pro Phe Glu Ala Lys Ile Asp Asn Gly Gly Glu Arg
 [1206] 195 200 205
 [1207] Gly Lys Met Asp Ser Glu Leu Ser Tyr Val Ser Ala Arg Asn Leu Leu
 [1208] 210 215 220
 [1209] Arg Cys Leu Ser Thr Trp Arg Ala Ser Ser Val Met Arg Arg Asp Ser
 [1210] 225 230 235 240
 [1211] Tyr Leu Ile Glu Glu Glu Arg Ile Lys Glu Ala Glu Ser Lys Met Thr
 [1212] 245 250 255
 [1213] Pro Glu Ile Ile Asp Gly Leu Arg Arg Leu Tyr Arg Tyr Cys Ala Val
 [1214] 260 265 270
 [1215] Asp His Asp Phe Leu Lys Trp Phe Gly Gly Arg Ile Ile Arg His Ile
 [1216] 275 280 285
 [1217] Asp Ser Cys Leu Ala Pro Ala Ile Ala Gly Asn Thr Gly Arg Pro Thr

[1218]	290	295	300
[1219]	Gly Gly Glu Ser Phe Thr Val Ile Tyr Asp Arg Arg Lys Lys Arg Asp		
[1220]	305	310	315 320
[1221]	Val Lys Ile Thr Tyr Ser Val Pro Glu Glu Ile Tyr Gly Tyr Leu Ser		
[1222]		325	330 335
[1223]	Ser His Pro Glu Leu Val Ala Ile Gly Lys Asp Gly Met Thr Pro Ile		
[1224]		340	345 350
[1225]	Ser Arg His Ala Asp Tyr Leu Glu Met Ile Ala Ser His Glu Lys His		
[1226]		355	360 365
[1227]	Arg Trp Tyr Ala Thr Phe Pro Thr Val Gly Lys Glu Asp Gly Tyr Arg		
[1228]	370	375	380
[1229]	Thr Ser Val Leu Leu Gly Lys Asn Tyr Leu Thr Tyr Asp Leu Ser Tyr		
[1230]	385	390	395 400
[1231]	Asp Gly Glu Ser Val Pro Asp Lys Lys Ile Asn Val Ile Ser Lys Gly		
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[1233]	Gln Pro Val Cys Leu Asp Leu His Asp Gly Arg Arg Val Ser Ser Leu		
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[1235]	Tyr Leu Thr Val Gly Glu Ser Ala Ala Tyr Asp Ile Ala Val Arg Lys		
[1236]		435	440 445
[1237]	Asn Lys Arg His His Gly Lys Pro Ala Asp Tyr Cys Arg Met Arg Val		
[1238]	450	455	460
[1239]	His Leu Thr Gln Glu Arg Glu Asp Lys Thr Tyr Asn Asp Pro Tyr Phe		
[1240]	465	470	475 480
[1241]	Ser Asn Met Glu Ile Trp Arg Ala Gly Asp Gln Val Tyr Ala Ile Glu		
[1242]		485	490 495
[1243]	Phe Asp Arg His Gly Ala Arg Tyr Thr Ala Ile Val Lys Glu Pro Ser		
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[1245]	Val Glu Tyr Arg Asn Lys Lys Leu Tyr Leu Arg Val Asn Met Val Leu		
[1246]		515	520 525
[1247]	Asp Ser Pro Ser Arg Gln Asp Asp Lys Asp Met Tyr Tyr Ala Tyr Met		
[1248]	530	535	540
[1249]	Thr Ala Tyr Pro Ser Ser Asn Pro Pro Val Glu Thr Ser Asp Asn Lys		
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[1251]	Lys Arg Phe Glu Arg Leu Gly Pro Gly Arg Arg Ala Ile Gly Gly Ile		
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[1253]	Asp Ile Gly Ile Gly Arg Pro Tyr Val Ala Val Val Ala Ser Tyr Glu		
[1254]		580	585 590
[1255]	Val Gly Pro Ala Gly Thr Glu Gln Lys Phe Gln Ile Glu Asp Arg Leu		
[1256]		595	600 605
[1257]	Ile Glu Asp Asp Gly Ser Ser Pro Tyr Asp Ser Leu Tyr Asn Asp Phe		
[1258]	610	615	620
[1259]	Leu Thr Asp Ile Arg Thr Val Ser Arg Ile Ile Glu Ala Ala Lys Lys		

[1260]	625	630	635	640
[1261]	Ile Ser Glu Gly Asp Leu Glu Asp Ile Pro Ser Asp Met Ser Val Asp			
[1262]		645	650	655
[1263]	Glu Asp Gly Ser Ile Ala Ala Thr Met Lys Arg Met Ser Ala Arg Ile			
[1264]		660	665	670
[1265]	Ala Glu Arg His His Leu Tyr Gly Glu Arg Lys Ser Glu Ala Tyr Ala			
[1266]		675	680	685
[1267]	Thr Phe Leu Lys Met Asn His Lys Gln Arg Leu Asp Ile Leu Leu Thr			
[1268]		690	695	700
[1269]	Gln Lys Ala Ser Asn Ala Thr Leu Lys Gln Leu Val Glu Glu Asp Pro			
[1270]	705	710	715	720
[1271]	Ser Phe Leu Pro Arg Ile Cys Val Tyr Tyr Val Ile Ser Val Glu Arg			
[1272]		725	730	735
[1273]	Glu Leu Lys Asn Lys His Arg Asn Ala Tyr Leu Asp Gly Leu Thr Val			
[1274]		740	745	750
[1275]	Asp Glu Lys Tyr Ser Gly Glu Thr Lys Arg Gly Tyr Ala Gln Lys Arg			
[1276]		755	760	765
[1277]	Leu Asn Ser Met Leu Arg Ala Tyr Ser Ala Leu Gly Glu Glu Glu Thr			
[1278]		770	775	780
[1279]	Asp Glu Val Arg Thr Phe Ser Thr Arg Ser Glu Lys Val Arg Asn Met			
[1280]	785	790	795	800
[1281]	Ala Lys Asn Ala Ile Lys Arg Asn Ala Arg Lys Leu Val Asn Phe Tyr			
[1282]		805	810	815
[1283]	Val Gly Lys Gly Ile Arg Thr Ile Val Ala Glu Asp Thr Asp Pro Thr			
[1284]		820	825	830
[1285]	Lys Ser Arg Asn Asp Gly Lys Lys Ser Asn Arg Ile Lys Ala Ala Trp			
[1286]		835	840	845
[1287]	Ser Pro Lys Gln Phe Leu Ala Ala Val Lys Asn Ala Ala Gln Trp His			
[1288]		850	855	860
[1289]	Gly Leu Glu Ile Ala Glu Val Asp Pro Arg Met Thr Ser Gln Val His			
[1290]	865	870	875	880
[1291]	Pro Glu Thr Gly Leu Ile Gly Tyr Arg Asp Gly Asp Thr Leu His Cys			
[1292]		885	890	895
[1293]	Pro Asp Gly Ser Lys Ile Asp Ala Asp Val Ala Gly Ala Ala Asn Val			
[1294]		900	905	910
[1295]	Cys Arg Val Phe Ala Gly Arg Gly Leu Trp Arg Phe Ser Ile Asn Thr			
[1296]		915	920	925
[1297]	Asn Ile Asp Ile Ser Asn Lys Asp Glu Lys Lys Arg Leu Arg Ala Tyr			
[1298]		930	935	940
[1299]	Ile Val His His Phe Gly Ser Glu Ser Asn Trp Glu Lys Phe Arg Lys			
[1300]	945	950	955	960
[1301]	Gln Tyr Pro Ser Gly Thr Thr Leu Tyr Leu His Gly Arg Glu Trp Leu			

[1302]		965		970		975
[1303]	Thr Ala Glu Glu His Lys Ser Ala Ile Asp Arg Ile Arg Asp Asp Val					
[1304]		980		985		990
[1305]	Gly Arg Asp Ala Glu Asn Asp His Val Ala Ile Val Thr Ala Ala Glu					
[1306]		995		1000		1005
[1307]	Lys Val Glu Ile Phe					
[1308]		1010				
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[1318]	Lys Thr His Ser Asp Gln Asn Ala Lys Gln Val Ala Glu Glu Val Lys					
[1319]		20		25		30
[1320]	Lys Glu His Leu Asn Tyr Leu Leu Ile Lys Asn Glu Met Leu Ile Ser					
[1321]		35		40		45
[1322]	Ile Val Pro Glu Ala Lys Asp Asp Asp Gly Asn Asp Ile Asp Phe Lys					
[1323]		50		55		60
[1324]	Lys Gln Leu Lys Ser Leu Tyr Lys Glu Thr Asp Gln Ser Val Ser Phe					
[1325]	65	70		75		80
[1326]	Ser Val Phe Cys Gln Met Met Lys Phe Arg Asn Ile Ala Leu Leu Tyr					
[1327]		85		90		95
[1328]	Ala Lys Gly Gln Ser Arg Trp Ala Val Ser Ser Tyr Phe Thr Gly Asn					
[1329]		100		105		110
[1330]	Arg Arg Lys Asp Asp Tyr Ala Lys Asp Leu Ser Leu Leu Asp Glu Ala					
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[1332]	Ile Glu Leu Leu Glu Cys Lys Arg Arg Lys Lys Ala Glu Glu Glu Asn					
[1333]		130		135		140
[1334]	Glu Glu Glu Asn Glu Thr Pro Lys Lys Lys Glu Asp Asn Pro Ser Asn					
[1335]	145	150		155		160
[1336]	Ile Ser Glu Glu Gln Ile Met Lys Leu Phe Tyr Ala Val Asn Lys Lys					
[1337]		165		170		175
[1338]	Leu Lys Glu Ile Gly Tyr Leu Asp Arg Tyr Ser His Ile Glu Lys Gln					
[1339]		180		185		190
[1340]	Glu Gln Tyr Ala Ile Ile Gly Val Thr Ser Arg Thr Val Lys Ala Trp					
[1341]		195		200		205
[1342]	Asp Tyr Ala Asn Phe Ala Thr Arg Asn His Tyr Gln Ser Val Gln Asn					
[1343]		210		215		220

[1344]	Glu Tyr Gln Lys Lys Leu Lys Ala Leu Pro Gly Thr Lys Lys Asp Lys			
[1345]	225	230	235	240
[1346]	Val Cys Leu Glu Lys Phe Phe Asp His Leu Asn Glu Asn Asn Ile Ala			
[1347]		245	250	255
[1348]	Ala Asp Trp Asp Lys Trp Arg Leu Lys Lys His Ile Leu Gln Cys Ile			
[1349]		260	265	270
[1350]	Ile Pro Ala Ala Lys Ile Gly Leu Lys Glu Leu Lys Gln Ser Phe Tyr			
[1351]		275	280	285
[1352]	Val Asp Asn Lys Gly Asn Lys His Asn Tyr Phe Val Asn Gly Leu Tyr			
[1353]		290	295	300
[1354]	Glu Glu Ile Leu Lys Arg Pro Phe Leu Tyr Ser Ala Glu Asp Pro Glu			
[1355]		305	310	315
[1356]	Glu Ser Ile Leu Tyr Leu Gly Val Glu Val Ala Ser Leu His Ser Lys			
[1357]		325	330	335
[1358]	Leu Asn His Leu Arg Ser Glu Ala Arg Phe Ser Phe Glu Thr Pro Asp			
[1359]		340	345	350
[1360]	Asp Ile Cys Lys Tyr Met Thr Ile Cys Gly Asp Asn Tyr His Asn Phe			
[1361]		355	360	365
[1362]	Thr Met Ser Ala Ile Gly Glu Asp Val Glu Asp Ile Glu Val Glu Val			
[1363]		370	375	380
[1364]	Tyr Asp Tyr Asn His Ser Lys Lys Tyr Glu Thr Met Arg Phe Ile Asn			
[1365]		385	390	395
[1366]	Gly Lys Arg Thr Thr Asp Leu Ser Leu Asn Phe Lys Gly Ile Pro Val			
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[1368]	Arg Leu Cys Leu Glu Gly Lys Arg Asn Asn Ser Tyr Phe Ala Asp Ala			
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[1370]	Ile Val Trp Glu Leu Asp Asn Lys Asp Lys Thr Gly Tyr Leu Ile Glu			
[1371]		435	440	445
[1372]	Tyr Gly Lys Ser Asn Asn Arg Leu Tyr Met Leu Val Lys Glu Pro Leu			
[1373]		450	455	460
[1374]	Ile Gly Cys Arg Arg Lys Phe Gly Lys Asp Val Leu Phe Val Ser Leu			
[1375]		465	470	475
[1376]	Ser Gly Thr Leu Val Asn Lys Tyr Ile Glu Asp Asp Ile Val Ser Ala			
[1377]		485	490	495
[1378]	Arg Tyr Leu Met Gln Thr Ala Ala Pro Ile Phe Lys Thr Ser Arg Ala			
[1379]		500	505	510
[1380]	Lys Lys Gln Asp Lys Ile Gly Asp Lys Trp Phe Glu His Cys Gln Gly			
[1381]		515	520	525
[1382]	Ser Thr Ile Lys Ile Ala Gly Ile Asp Ile Gly Ile Asn Pro Ile Ala			
[1383]		530	535	540
[1384]	Ala Ile Thr Val Ala Asn Val Thr Phe Asp Arg Ala Leu Gly Asn Lys			
[1385]		545	550	555
				560

[1386]	Ile Lys Asn Gln Lys Gln Ile Val Ile Asp Cys Tyr Ala Glu Asp Tyr
[1387]	565 570 575
[1388]	Lys Ile Asp Pro Val Val Val Lys Arg Met Glu Asp Ile Arg His Ile
[1389]	580 585 590
[1390]	Lys Tyr Thr Ile Asn Ser Trp Tyr His Leu Ala Asp Cys Cys Arg Leu
[1391]	595 600 605
[1392]	Lys Ala Ala Asn Lys Glu Tyr Val Val Asn Glu Arg Lys Gln Gly Phe
[1393]	610 615 620
[1394]	Phe Arg Glu Asn Ile Glu Tyr Leu Lys Glu Val Ala Lys Lys Ala Ile
[1395]	625 630 635 640
[1396]	Thr Glu Ser Asp Gln Gln Ile Lys Glu Gln Lys Ala Ala Leu Lys Arg
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[1398]	Phe Asp Gly Glu Lys Lys Lys Glu Ile Gln Ala Thr Ile Asn Gly Phe
[1399]	660 665 670
[1400]	Asn Leu Lys Ile Lys Ile Leu Lys Lys Phe Val Arg Gln Ser Ala Lys
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[1402]	Lys Ile Phe Asp Ser Thr Leu Glu Thr Leu Glu Lys Tyr Asp Asn Asn
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[1404]	Ile Glu Gln Ala Lys Arg Asp Arg Glu Phe Gly Leu Lys Ile Ile Tyr
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[1406]	Asp Leu Ile Ile Lys Tyr Tyr Lys Arg Ser Lys Lys Glu Arg Glu Met
[1407]	725 730 735
[1408]	Asn Gln Arg Ile Tyr Val Asp Asp Tyr Asn Gln Glu Glu Ile Asp Thr
[1409]	740 745 750
[1410]	Glu Arg Thr Lys Lys Ile Arg Lys Glu Thr Ile Thr Phe Cys Asp Asn
[1411]	755 760 765
[1412]	Asp Trp Asn Ser Leu Thr Lys Arg Ile His Asp Leu Glu Lys Lys Met
[1413]	770 775 780
[1414]	Lys Lys Ile Gly Ile Ser Glu Pro Gly Arg Val Glu Gln Glu Ile Asn
[1415]	785 790 795 800
[1416]	Asp Arg Asp Tyr Tyr Asn Asn Ile Gln Asp Asn Thr Lys Lys Arg Gln
[1417]	805 810 815
[1418]	Ala Lys Ile Ile Val Asp Ala Leu Lys Glu Glu Gly Val Ser Ile Ile
[1419]	820 825 830
[1420]	Val Val Glu Asp Leu Thr Gly Gly Gly Ser Glu Asn Thr Lys Glu Ile
[1421]	835 840 845
[1422]	Asn Lys Ser Phe Asp Ala Phe Ala Pro Ile Arg Phe Leu Asn Ala Leu
[1423]	850 855 860
[1424]	Lys Asn Cys Ala Glu Thr Asn Gly Ile Gln Val Thr Glu Val Leu Ser
[1425]	865 870 875 880
[1426]	Pro Met Ser Ser Lys Met Val Pro Ser Thr Gly Glu Ile Gly His Arg
[1427]	885 890 895

[1428]	Asp Lys Arg Asp Lys Gln Leu Tyr Tyr Lys Asp Gly Glu Glu Leu Lys
[1429]	900 905 910
[1430]	Ser Ile Asp Gly Asp Ile Ser Ala Ser Glu Ile Leu Leu Arg Arg Gly
[1431]	915 920 925
[1432]	Val Ser Arg His Thr Glu Leu Ile Gly Thr Met Asn Val Glu Asp Val
[1433]	930 935 940
[1434]	Leu Asp Lys Asn Asn Asn Lys Asn Lys Cys Ile Lys Gly Tyr Val Cys
[1435]	945 950 955 960
[1436]	Asn Arg Trp Gly Asn Ile Gln Asn Phe Glu Lys Ile Leu Lys Glu Lys
[1437]	965 970 975
[1438]	Gly Ile Gly Glu Arg Glu Ile Ile Tyr Leu His Gly Asp Lys Ile Leu
[1439]	980 985 990
[1440]	Thr Met Asp Glu Lys Arg Thr Leu Gln Ala Ser Ile Arg Lys Glu Leu
[1441]	995 1000 1005
[1442]	Lys Glu Met Arg Glu Arg Glu Ser Gly Glu Glu Asn Ala Gly Thr
[1443]	1010 1015 1020
[1444]	Ala Arg Lys Lys Ser Lys Pro Lys Lys Lys Lys Lys Ile Lys Arg
[1445]	1025 1030 1035
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[1447]	1040 1045 1050
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[1458]	20 25 30
[1459]	Phe Tyr Lys Glu Ser Phe Glu Leu Phe Lys Gln Phe Thr Asn Glu Phe
[1460]	35 40 45
[1461]	Val Ala Trp Gly Asn Asp Glu Ile Tyr Gln Tyr Gly Ser Ser Lys Arg
[1462]	50 55 60
[1463]	Lys Lys Asp Asp Gln Lys Ile Ser Leu Ile Pro Val Ile Glu Asp Ile
[1464]	65 70 75 80
[1465]	Tyr Lys Ser Val Glu Lys Lys Ala Thr Ala Glu Gly Ile Ser Lys Thr
[1466]	85 90 95
[1467]	Asp Phe Arg Ala Val Leu Lys Tyr Leu Tyr His Gln Ile Ile Asn Val
[1468]	100 105 110
[1469]	Gly Asn Ser Gly Arg Ser Tyr Gly Thr Ser Leu Phe Gly Gly Cys Glu

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[1471]	Val Lys Glu Lys Leu Ser Lys Gln Asp Ile Ser Asn Ile Val Glu Cys		
[1472]	130	135	140
[1473]	Val Lys Glu Leu Glu Leu Cys Lys Ser Lys Gln Glu Glu Ser Asp Ala		
[1474]	145	150	155
[1475]	Tyr Asp Lys Ile Leu Leu Lys Glu Lys Ile Thr His Ile Val Lys Ser		
[1476]	165	170	175
[1477]	Gly Glu Thr Ala Gly Asp Ile Thr Lys Lys Tyr Asn Gln Ala Thr Thr		
[1478]	180	185	190
[1479]	Gly Arg Lys Thr Ser Ser Lys Gly Phe Phe Asp Lys Ser Thr Lys Thr		
[1480]	195	200	205
[1481]	Glu Val Lys Tyr Lys Asp Ile Lys Asp Asp Thr Leu Leu Gln Asp Gly		
[1482]	210	215	220
[1483]	Ser Thr Ile Phe Ile Lys Ser Ser Val Asp Leu Phe Val Lys Lys Val		
[1484]	225	230	235
[1485]	Cys Asn Thr Leu Arg Glu Ile Asn Phe Phe Asp Arg Leu Pro Phe Lys		
[1486]	245	250	255
[1487]	Asn Asn His Ser Asn Asn Tyr Gly Leu Leu Phe Ser Met Leu Ser Gln		
[1488]	260	265	270
[1489]	Ile Glu Ser Trp Lys Thr Ile Ser Glu Thr Thr Lys Lys Ser His Glu		
[1490]	275	280	285
[1491]	Glu His Gly Glu Lys Ile Ala Ser Met Val Lys Lys Leu Asp Leu Thr		
[1492]	290	295	300
[1493]	Gln Thr Glu Leu Met Lys Asp Phe Ala Ala Phe Cys Ile Glu Asn Asn		
[1494]	305	310	315
[1495]	Ile Thr Lys Lys Phe Asp His Lys Phe Lys Arg His Met Glu Asp Cys		
[1496]	325	330	335
[1497]	Val Ile Pro Ser Phe Lys Asn Gly Lys Ile Pro Asp Lys Leu Phe Tyr		
[1498]	340	345	350
[1499]	Phe Asn Ile Ile Leu Ala Lys Lys Thr Asp Glu Gln Ile Asp Tyr Ser		
[1500]	355	360	365
[1501]	Leu Ser Ser Glu Phe Tyr Thr Lys Leu Phe Ser Met Pro Asn Leu Trp		
[1502]	370	375	380
[1503]	Gln Glu Glu Glu Ala Phe Ile Val Lys Asn Ile Asn Leu Ile Glu Glu		
[1504]	385	390	395
[1505]	Ile Thr Ile Phe Asn Lys Arg Arg Asn Tyr Ala Cys Cys Pro Leu Ile		
[1506]	405	410	415
[1507]	Lys Glu Lys Glu Tyr Asp Arg Phe Gln Ile Gln Leu Asn Glu Thr Asn		
[1508]	420	425	430
[1509]	Phe Leu Lys Phe Gln Phe Asp Pro Lys Asn Val Val Asn Ile Asp Glu		
[1510]	435	440	445
[1511]	Asn Thr Thr Glu Ala Thr Val Gly Phe Asp Glu Lys Leu Lys Leu Val		

[1512]	450	455	460
[1513]	Val Cys Ala Asp Lys Lys Tyr Ala Phe Ser Ile Phe Thr Gln Cys Lys		
[1514]	465	470	475 480
[1515]	Tyr His Gly Asn Lys His Lys Pro Asn Thr Tyr Phe Asn Asn Leu Lys		
[1516]		485	490 495
[1517]	Ile Ile Lys Val Ile Glu Ser Lys Ser Asn Ser Val Lys Ser Met Lys		
[1518]		500	505 510
[1519]	Tyr Thr Phe Glu Phe Thr Lys Arg Asn Glu Leu Lys Arg Ala Glu Ile		
[1520]		515	520 525
[1521]	Lys Gln Pro Ser Ile Val Tyr Lys Asn Asn Asn Tyr Tyr Ile Arg Ile		
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[1523]	Asn Met Asn Val Ile Leu Asp Ala Asp Gln Thr Ser Tyr Lys Ile Ile		
[1524]		545	550 555 560
[1525]	Asn Asn Asn Gln Thr Ala Ser Leu Pro Ser Tyr Phe Gln Ser Ser Leu		
[1526]		565	570 575
[1527]	Pro Phe Glu Asn Asn Arg Gly Lys Ile His Asp Lys Gly Ile Val His		
[1528]		580	585 590
[1529]	Trp Glu Lys Ile Lys Asn Arg Lys Ile Ile Ala Met Gly Val Asp Leu		
[1530]		595	600 605
[1531]	Gly Val Arg Arg Pro Phe Ser Tyr Ala Ile Gly Asn Phe Thr Leu Asn		
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[1533]	Lys Asp Ile Leu Asp Lys Asn Asp Val Asn Ile Val Ala Ser Gly Phe		
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[1537]	Lys Thr Leu Ala Lys Phe Ile Gly Lys Leu Lys Ser His Asn Lys Gly		
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[1539]	Leu Lys Val Asp Phe Glu Lys Asp Lys Lys Tyr Ile Phe Asp Leu Val		
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[1541]	Asn Asp Ala Lys Ala Tyr Phe Lys Asp Met Ser Ala Lys Arg Ile Asn		
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[1543]	Asp Thr Lys Asp Asn Ile Ser Asn Thr Val Thr Asn Lys Glu Arg Ile		
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[1545]	Tyr Gly Ser Phe Val Ser Glu Ser Ala Glu Ser Ala Ile Gln Cys Ala		
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[1547]	Ile Asp Arg Ser Glu Lys Glu Ser Gly Leu Thr Leu Lys Lys Asp Ile		
[1548]		740	745 750
[1549]	Ser Trp Leu Val Asn Val Leu Ser Lys Tyr Leu Glu Arg Lys Phe Lys		
[1550]		755	760 765
[1551]	Glu Val Lys Asn Asn Arg Lys Tyr Thr Asn Val Asn Lys Cys Asp Asn		
[1552]		770	775 780
[1553]	Cys Phe Asn Trp Leu Arg Val Ile Glu Asn Ile Lys Arg Leu Lys Arg		

[1554]	785	790	795	800
[1555]	Ser Ile Ser Tyr	Leu Gly Glu Asp	Leu Gln Lys Asn Pro	Glu Leu Lys
[1556]		805	810	815
[1557]	Ile Glu Leu Lys	Asn Leu Asn Glu Tyr	Gly Asn Asn Val	Lys Ser Asp
[1558]		820	825	830
[1559]	Phe Leu Lys Gln	Ile Ala Ser Asn	Ile Ile Lys Val	Ala Ile Glu His
[1560]		835	840	845
[1561]	Lys Cys Asp Ile	Val Phe Ile Glu	Lys Leu Gly Lys	Ala Asp Ser Arg
[1562]		850	855	860
[1563]	Ser Arg Lys Leu	Asn Glu Met Phe	Ser Phe Trp Ser	Pro Lys Ala Ile
[1564]	865	870	875	880
[1565]	Lys Lys Ala Ile	Glu Asn Ala Ala	Ser Trp His Gly	Ile Pro Val Val
[1566]		885	890	895
[1567]	Glu Val Asp Pro	Ser Cys Thr Ser	Lys Val His Tyr	Glu Thr Asn Leu
[1568]		900	905	910
[1569]	Phe Gly His Arg	Ile Gly Asn Asp	Leu Tyr Tyr Val	Glu Asp Gln Cys
[1570]		915	920	925
[1571]	Leu Lys Lys Val	Asp Ala Asp Ile	Asn Ala Ala Lys	Gln Ile Leu Val
[1572]		930	935	940
[1573]	Arg Gly Ala Thr	Arg His Gly Asn	Ile Ser Ser Ile	Asn Ile Lys Tyr
[1574]	945	950	955	960
[1575]	Leu Gln Ala Lys	Ile Ala Glu Leu	Asn Ser Glu Ala	Asn Ser Glu Glu
[1576]		965	970	975
[1577]	Asp Lys Glu Glu	Ile Lys Gln Gly	Gly Lys Arg Ile	Gln Gly Phe Leu
[1578]		980	985	990
[1579]	Trp Lys Lys Tyr	Gly Asn Ile Thr	Asn Ile Thr Asn	Gln Leu Thr Ala
[1580]		995	1000	1005
[1581]	Ala His Lys Glu	Arg Glu Ser Lys	Phe Asp Tyr Ile	Tyr Leu His
[1582]		1010	1015	1020
[1583]	Asn Asp Lys Trp	Ile Ala Tyr Glu	Asp Arg Asn Glu	Ile Lys Lys
[1584]		1025	1030	1035
[1585]	Asp Ile Glu Lys	Arg Leu Glu		
[1586]		1040	1045	
[1587]	<210>	13		
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[1589]	<212>	PRT		
[1590]	<213>	人工序列		
[1591]	<220>			
[1592]	<223>	Cas12j.15的氨基酸序列		
[1593]	<400>	13		
[1594]	Met Thr Ala Lys	Lys Thr Ala Lys	Lys Tyr Phe Pro	Pro Lys Cys Leu
[1595]	1	5	10	15

[1596]	Arg Ser Ser His Phe Lys Ile Tyr Gly Ile Pro Thr Ala Ile Arg Ala
[1597]	20 25 30
[1598]	Leu Glu Glu Thr Asn Thr Phe Val Asn Lys Ala Ala Ala Asp Leu Met
[1599]	35 40 45
[1600]	Glu Met Phe Phe Leu Met Arg Gly Gln Pro Tyr Arg Arg Arg Ile Gly
[1601]	50 55 60
[1602]	Ser Glu Glu Lys Gln Val Thr Gln Glu His Ile Asp Ala Arg Leu Arg
[1603]	65 70 75 80
[1604]	Val Leu Val Gly Asp Tyr Ser Leu Asn Glu Val Lys Pro Leu Leu Arg
[1605]	85 90 95
[1606]	Gln Leu Tyr Asp Gly Ile Lys Ala Lys Gln Asn Tyr Ala Pro Thr His
[1607]	100 105 110
[1608]	Phe Val Arg Phe Phe Ile Gln Pro Thr Lys Gly Ala Ile Asp Lys Lys
[1609]	115 120 125
[1610]	Ser Pro Val Ser Gln Arg Ala Lys Lys Ala Gly Gln Lys Leu Gln Lys
[1611]	130 135 140
[1612]	Met Gly Val Leu Pro Ile Leu Pro Leu Ser Pro Gly Phe Lys Phe Trp
[1613]	145 150 155 160
[1614]	Thr Ala Ala Met Met Met Ala Cys Ser Arg Met Asn Ser Trp Glu Ala
[1615]	165 170 175
[1616]	Cys Asn Glu Lys Thr Ile Glu Asn His Lys Ala Phe Leu Glu Gly Ile
[1617]	180 185 190
[1618]	Glu Asn Tyr Lys Lys Glu Ile Arg Phe Glu Asp Leu Cys Glu Glu Trp
[1619]	195 200 205
[1620]	Ser Leu Phe Ser Asp Trp Leu Thr Glu Ala Glu Ser Asp Asn Glu Gly
[1621]	210 215 220
[1622]	Gly Cys Lys Phe Lys Leu Thr Pro Arg Phe Leu Gln Arg Trp Glu Arg
[1623]	225 230 235 240
[1624]	Ile Tyr Leu Lys Gln Met Arg Lys Gly Lys Ile Pro Ala Arg His Asn
[1625]	245 250 255
[1626]	Leu Gly Pro Val Met Glu Ala Leu Ala Gly Asp Lys Tyr Arg Gln Leu
[1627]	260 265 270
[1628]	Trp Asp Asn Gly Glu Glu Arg Asp Tyr Ile Thr Glu Leu Gly Asp Leu
[1629]	275 280 285
[1630]	Val Thr Ser Gln Arg Lys Ala Val Arg Leu Ser Arg Asp Ser Ala Val
[1631]	290 295 300
[1632]	Thr Phe Pro Asp Glu Glu Leu Ser Pro Val Gly Thr Glu Phe Gly His
[1633]	305 310 315 320
[1634]	Asn Tyr Met Ser Phe Ser Ile Asp Gln Glu Asn Ser His Leu Val Thr
[1635]	325 330 335
[1636]	Leu Glu Val Ile Gly Gly Lys Tyr Gln Phe Glu Ile Ser Lys Ser Asp
[1637]	340 345 350

[1638]	Tyr Phe Arg Asp Leu Ile Val Glu Glu Ala Gly Lys Gln Ser Lys Phe
[1639]	355 360 365
[1640]	Tyr Asn Val Ser Tyr Arg Lys Gly Asn Val Arg Glu Glu Asn Leu Ala
[1641]	370 375 380
[1642]	Gly Asp Phe Lys Glu Ala Thr Val Arg Asn Arg Arg Ser Leu Lys Thr
[1643]	385 390 395 400
[1644]	Gly Lys Arg Arg Leu Tyr Phe Tyr Met Ser His Ser Ile Pro Thr Arg
[1645]	405 410 415
[1646]	Phe Asp Asp Asp Leu Tyr Ala Gln Phe Thr Glu Lys Gly Gln Pro Asp
[1647]	420 425 430
[1648]	Phe Ser Lys Leu Tyr Lys Ala Val Thr Tyr Phe Gln Cys Ser Leu Gly
[1649]	435 440 445
[1650]	Asn Lys Lys Ala Asp Thr Tyr Arg Val Tyr Val Lys Met Gly Thr Arg
[1651]	450 455 460
[1652]	Phe Leu Gly Val Asp Ile Gly Val Ser Arg Leu Phe Gly Phe Ser Leu
[1653]	465 470 475 480
[1654]	Phe Glu Leu Arg Glu Glu Lys Pro Glu Lys Asn Pro Phe Phe Glu Leu
[1655]	485 490 495
[1656]	Pro Asp Asp Leu Gly Tyr Ala Val Cys Leu Glu Ser Trp Val Asp Gly
[1657]	500 505 510
[1658]	Val Glu Lys Asn His Lys Val Ala Gln Glu Met Lys Asp Trp Arg Arg
[1659]	515 520 525
[1660]	Glu Cys Leu Ala Ala Gln Arg Leu Ile His Tyr Ala Lys Phe Leu Lys
[1661]	530 535 540
[1662]	Lys Arg Asp Lys Asn Glu Glu Ile Asp Tyr Lys His Glu Glu Ser Leu
[1663]	545 550 555 560
[1664]	Glu Thr Ile Ala Gly Leu Leu Gly Ile Glu Ile Asp Pro Glu Gln Ile
[1665]	565 570 575
[1666]	Ile Asp Val Pro Leu Lys Leu Leu Asp Leu Val Gly Gln Ala Ile Gly
[1667]	580 585 590
[1668]	Ala Leu Arg Lys Lys Tyr Leu Val Leu Lys Lys Asn Glu Val Arg Gln
[1669]	595 600 605
[1670]	Gly Arg Ile Thr Ser Glu Leu Phe Leu Trp Pro Glu Cys Val Asp Thr
[1671]	610 615 620
[1672]	Tyr Ile Arg Leu Leu Lys Ser Trp Thr Tyr Lys Asp Lys Lys Pro Tyr
[1673]	625 630 635 640
[1674]	Gln Lys Gly Glu Thr Asn Lys Asp Ala Phe Lys Lys Leu Lys Gly Tyr
[1675]	645 650 655
[1676]	Leu Ala Arg Leu Arg Lys Asp Leu Ala Pro Lys Tyr Ala Ala Val Ile
[1677]	660 665 670
[1678]	Ala Asp Ala Ala Ile Arg His Lys Val His Val Val Val Ala Glu Asn
[1679]	675 680 685

[1680]	Leu Glu Gln Phe Gly Leu Ser Met Lys Asn Glu Lys Asp Leu Asn Arg
[1681]	690 695 700
[1682]	Val Leu Ala His Trp Ser His Gln Lys Ile Trp Ser Met Val Glu Glu
[1683]	705 710 715 720
[1684]	Gln Leu Arg Pro Tyr Gly Ile Met Val Val Tyr Val Asp Pro Arg His
[1685]	725 730 735
[1686]	Thr Ser Lys Leu Asp Phe Ala Thr Asp Glu Phe Gly Gly Arg Cys Phe
[1687]	740 745 750
[1688]	Thr Ser Leu Tyr Val Met Arg Asp Gly Lys Lys Thr Thr Thr Asp Thr
[1689]	755 760 765
[1690]	Glu Lys Asn Ala Ser Gln Asn Ile Pro Lys Lys Phe Leu Thr Arg His
[1691]	770 775 780
[1692]	Arg Asn Val Ser Trp Leu Leu Ala Tyr Ala Val Asp Leu Ser Asp Ser
[1693]	785 790 795 800
[1694]	Gln Lys Lys Lys Leu Gly Ile Gly Asp Glu Lys Val Trp Leu Pro Asn
[1695]	805 810 815
[1696]	Met Gly Leu Met Ile Ser Gly Ala Leu Lys Ala Lys His Gly Lys Asn
[1697]	820 825 830
[1698]	Ser Ala Leu Leu Val Glu Asp Gly Glu Asn Tyr Arg Leu Leu Pro Ile
[1699]	835 840 845
[1700]	Thr Ala Ala Gln Ala Lys Lys Phe Val Val Lys Arg Lys Lys Glu Glu
[1701]	850 855 860
[1702]	Phe Tyr Arg His Gly Glu Ile Trp Leu Thr Lys Glu Ala His Lys Ala
[1703]	865 870 875 880
[1704]	Arg Ile Glu Tyr Leu Phe Pro Glu Ser Lys Lys Gly Arg Lys Ser
[1705]	885 890 895
[1706]	<210> 14
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[1713]	Met Lys Lys Thr Asn Tyr Lys Thr Ser His Leu Leu Ile Asp Asn Pro
[1714]	1 5 10 15
[1715]	Pro Gln Ser Ile Ile Asp Leu His Arg Asp Val Ile Glu Ile Gly Ser
[1716]	20 25 30
[1717]	Tyr Leu Thr Lys Phe Phe Leu Ala Cys Leu Gly Arg Pro Val Asp Ser
[1718]	35 40 45
[1719]	Thr Ile Leu Ser Glu Pro Ala Leu His Phe Gln Phe Val Asn Gly Ile
[1720]	50 55 60
[1721]	Leu Pro Val Lys Asn Gly Pro Gly Ala Asp Asp Ser Ser Trp Arg His

[1722]	65	70	75	80
[1723]	Ser Glu Asn Cys Tyr Ser Met Leu Phe Glu Lys Asn Ser Lys Ser Gly			
[1724]		85	90	95
[1725]	Lys Ser Asp Gly Lys Val Arg Gln Val Arg Glu Leu Lys Val Ala Leu			
[1726]		100	105	110
[1727]	Phe Gly Lys Lys Glu Lys Gly Lys Gly Ile Val Gly Lys Lys Thr Trp			
[1728]		115	120	125
[1729]	Asp Glu Leu Lys Val Val Leu Glu Ala Leu Pro Glu Glu His Gln Ile			
[1730]		130	135	140
[1731]	Leu Ser Leu Glu Ile Cys Gln Arg His Tyr Glu Ser Arg Asp Val Lys			
[1732]		145	150	155
[1733]	Ala Phe Gly Lys Leu Ala Leu Ser Ser Lys Ser Arg Pro Ser Val Glu			
[1734]		165	170	175
[1735]	Ala Gly Leu Lys Leu Arg Glu Leu Gly Leu Leu Pro Leu Asp Ser Arg			
[1736]		180	185	190
[1737]	Gly Leu Asp Lys Asn Lys Leu Leu Gly Ile Leu Ala Ala Val Thr Gly			
[1738]		195	200	205
[1739]	Arg Leu Lys Ser Trp Arg Asp Arg Asp Cys Ala Cys Lys Ala Asp Lys			
[1740]		210	215	220
[1741]	Gln Ala Leu Arg Val Lys Phe Glu Glu Arg Leu Ser Lys Val Asp Gln			
[1742]		225	230	235
[1743]	Ser Ala Tyr Gln Gln Phe Lys Gln Phe Ala Asp Glu Leu Leu Thr Gln			
[1744]		245	250	255
[1745]	Glu Gly Tyr Arg Ile Ser Gly Arg Val Leu Arg Ala Val Glu Lys Lys			
[1746]		260	265	270
[1747]	Asp Ser Asp Tyr Ser Pro Val Leu Thr Val Leu Ala Lys Tyr Pro Asp			
[1748]		275	280	285
[1749]	Leu Gln Asp Asn Phe Glu Glu Leu Cys Arg Ala Cys Leu Ala Glu Gln			
[1750]		290	295	300
[1751]	Ala Phe Asn Lys Lys Lys Ala Asp Ala Arg Val Thr Val Cys Ser Glu			
[1752]		305	310	315
[1753]	Thr Ser Pro Leu Gln Phe Pro Phe Gly Met Thr Gly Asn Gly Tyr Pro			
[1754]		325	330	335
[1755]	Phe Thr Leu Ser Ala Cys Glu Gly Arg Ile Asn Ala Thr Ile His Phe			
[1756]		340	345	350
[1757]	Pro Gly Gly Asp Leu Pro Leu Arg Leu Arg Lys Ser Lys Tyr Phe Gln			
[1758]		355	360	365
[1759]	Asn Pro Glu Ile Leu Pro Val Lys Asp Gly Phe Gln Ile Thr Phe Thr			
[1760]		370	375	380
[1761]	Arg Gly Lys Thr Pro Leu Val Gly Thr Ile Lys Glu Pro Ser Leu Leu			
[1762]		385	390	395
[1763]	Lys Lys Asn Asn His Tyr Tyr Leu Ser Leu Arg Val Asn Val Pro Ser			

[1764]		405		410		415
[1765]	Val Lys Ile Pro Lys Glu Val Arg Asp Thr Arg Ala Tyr Tyr Ser Ser					
[1766]		420		425		430
[1767]	Ala Val Gly Gly Asp Glu Thr Thr Pro Val Pro Val Lys Ala Val Ala					
[1768]		435		440		445
[1769]	Ile Asp Leu Gly Val Thr Thr Leu Ala Asp Tyr Ser Ile Ile Asp Thr					
[1770]		450		455		460
[1771]	Cys Leu Pro Gly Asp Cys Lys Val Phe Gly Gly Glu Thr Ala Ala Phe					
[1772]		465		470		475
[1773]	Thr Ala His Gly Lys Ile Gly Gln Cys Ala Asn Lys Ser Leu Arg Asp					
[1774]		485		490		495
[1775]	Arg Leu Tyr Lys Asn Thr Glu Glu Ala Leu Phe Leu Gly Lys Phe Ile					
[1776]		500		505		510
[1777]	Arg Leu Ser Lys Lys Leu Arg Asp Gly Glu Gly Leu Asn Arg Trp Glu					
[1778]		515		520		525
[1779]	Val Glu Lys Leu Pro Gly Tyr Ala Glu Arg Leu Gly Ile Thr Gln His					
[1780]		530		535		540
[1781]	Leu Asp Asn Ala Tyr Thr Arg Lys Asp Glu Ile Ala Arg Lys Phe Lys					
[1782]		545		550		555
[1783]	Gln Ile Lys Gly Asn Phe Asp Lys Leu Val Ser Glu Phe Ala Leu Arg					
[1784]		565		570		575
[1785]	Asp His Pro Ser Lys Lys Gly Glu Ser Trp Glu Thr Ile Ser Ala Glu					
[1786]		580		585		590
[1787]	Thr Ile Gln Val Leu Ala Ala Leu Lys Arg Ile Gln Ser Leu Leu Lys					
[1788]		595		600		605
[1789]	Ser Trp Thr Tyr Tyr Ser Trp Thr Ala Glu Asp Tyr Val Leu Ala Leu					
[1790]		610		615		620
[1791]	Thr Ala Asp Gly Pro Val Cys Ile Asp Gly Glu His Val Lys Ala Val					
[1792]		625		630		635
[1793]	Thr Ala Thr Ser Arg Arg Ser Phe Ala Pro Cys Gly Lys Ala Ala Leu					
[1794]		645		650		655
[1795]	Leu Arg Leu Ile Glu Ser Gly Glu Ile Val Glu Thr Gly Gly Gln Tyr					
[1796]		660		665		670
[1797]	Gln Leu Ala Thr Gly Val Lys His Arg Asn His Pro Val Asn Phe Leu					
[1798]		675		680		685
[1799]	Ser Ser Tyr Ile Lys His Phe Asn Gly Leu Arg Arg Asp Leu Thr Asn					
[1800]		690		695		700
[1801]	Lys Leu Val Arg Ala Ile Val Asn Lys Ala Gln Glu Tyr Arg Val Gln					
[1802]		705		710		715
[1803]	Ile Val Ile Val Glu Asp Phe Gly Ile Ala Asp Leu Glu Asp Arg Ile					
[1804]		725		730		735
[1805]	Lys Asp Ala Tyr Glu Asn Tyr Arg Trp Asn Leu Phe Ala Pro Ala Thr					

[1806]	740	745	750
[1807]	Ile Val Lys Lys Leu Glu Ala Ala Leu Leu Glu Val Gly Ile Ala Met		
[1808]	755	760	765
[1809]	Ala Gln Val Asp Pro Arg His Thr Ser Gln Ile Ala Pro Thr Gly Ala		
[1810]	770	775	780
[1811]	Phe Gly Phe Arg Asp His Ala Phe Leu Tyr Tyr Gln Asp Asp Gly Leu		
[1812]	785	790	795
[1813]	Cys Arg Ile Asp Ala Asn Thr Asn Ala Ser Met Arg Ile Ala Glu Arg		
[1814]	805	810	815
[1815]	Phe Phe Met Arg His Ser Val Leu Thr Gln Leu Arg Ala Ala Lys Ile		
[1816]	820	825	830
[1817]	Gly Glu Thr Glu Tyr Leu Ile Pro Glu Ser Ala Ser Lys Arg Leu Asn		
[1818]	835	840	845
[1819]	Ala Phe Val Lys Leu Gln Thr Gly Lys Pro Phe Ala Lys Leu Ile Met		
[1820]	850	855	860
[1821]	Asn Cys Ser Gly Phe Val Leu Glu Gly Leu Thr Lys Lys Gln Tyr Ala		
[1822]	865	870	875
[1823]	Lys Leu Ala Glu Thr Ala Gly Lys Lys Glu Ser Phe Tyr Gln Tyr Asp		
[1824]	885	890	895
[1825]	Asp Arg Trp Phe Asp Lys Gly His His Phe Ala Cys Arg Ala Thr Leu		
[1826]	900	905	910
[1827]	Glu Asn Lys Val Gln Val Cys Leu Asn Gly Gly Gly Arg Ile Lys Asp		
[1828]	915	920	925
[1829]	Thr Thr Pro Asp Phe Asn Pro Lys Ser Leu Leu Arg Ser Asp Leu Gln		
[1830]	930	935	940
[1831]	Thr Pro Leu Asp Gln Leu Phe Gly Asn Ser Gly Ala		
[1832]	945	950	955
[1833]	<210> 15		
[1834]	<211> 946		
[1835]	<212> PRT		
[1836]	<213> 人工序列		
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[1838]	<223> Cas12j.17的氨基酸序列		
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[1841]	1	5	10
[1842]	Gln Gln Glu Leu Ile Asp Leu His Arg Asp Ser Asn Glu Met Gly Ser		
[1843]	20	25	30
[1844]	Tyr Leu Thr Lys Phe Phe Leu Ala Ala Leu Gly Arg Pro Val Asp Asn		
[1845]	35	40	45
[1846]	Ser Ile Val Leu Pro Pro Glu Leu Ala Asp Leu Tyr Phe Gln Phe Ala		
[1847]	50	55	60

[1848]	Asn Gly Ile Leu Pro Val Asp Lys Gly Pro Gly Ser Asp Asp Pro Ser
[1849]	65 70 75 80
[1850]	Trp Leu His Ser Glu Asn Cys Tyr Ser Met Phe Phe Glu Lys Asp Ser
[1851]	85 90 95
[1852]	Met Ser Gly Asn Cys Thr Asn Lys Ile Lys Gln Tyr Gln Glu Leu Lys
[1853]	100 105 110
[1854]	Thr Ala Leu Cys Gly Gln Lys Val Lys Gly Gln Lys Gly Leu Val Gly
[1855]	115 120 125
[1856]	Lys Lys Thr Trp Ala Gln Leu Lys Lys Val Leu Thr Ala Leu Pro Gln
[1857]	130 135 140
[1858]	Lys Tyr Gln Ile Leu Ser Pro Lys Ile Cys Gln Lys Tyr Phe Lys Ser
[1859]	145 150 155 160
[1860]	Gly Asn Leu Glu Gly Phe Gly Lys Leu Ala Leu Ala Gly Lys Asn Arg
[1861]	165 170 175
[1862]	Pro Ser Met Ser Ala Gly Leu Gln Leu Arg Glu Leu Gly Leu Leu Pro
[1863]	180 185 190
[1864]	Leu Asp Ser Arg Gly Ile Asp Lys Asn Lys Leu Leu Gly Ile Leu Val
[1865]	195 200 205
[1866]	Gly Ile Thr Gly Arg Leu Lys Ser Trp Arg Asp Arg Asp Trp Ala Cys
[1867]	210 215 220
[1868]	Lys Thr Val Lys Glu Glu Leu Arg Val Thr Phe Glu Lys Gly Leu Gly
[1869]	225 230 235 240
[1870]	Glu Val Asp Pro Thr Ala Tyr Pro Gln Phe Lys Gln Phe Ala Asp Gln
[1871]	245 250 255
[1872]	Leu Phe Lys Gln Glu Gly Tyr Lys Ile Ser Gly Arg Val Leu Arg Ala
[1873]	260 265 270
[1874]	Val Glu Gly Lys Asp Ala Asp Tyr Gln Pro Val Leu Ser Leu Leu Thr
[1875]	275 280 285
[1876]	Gln Tyr Pro Asp Leu Gln Gly Asp Phe Glu Glu Leu Gly Arg Val Tyr
[1877]	290 295 300
[1878]	Leu Ala Glu Ala Glu Tyr Leu Arg Lys Lys Val Asp Ala Arg Val Thr
[1879]	305 310 315 320
[1880]	Val Cys Asp Ala Glu Thr Ser Pro Leu Gln Phe Pro Phe Gly Leu Thr
[1881]	325 330 335
[1882]	Gly Asn Gly Tyr Ser Ile Thr Leu Thr Val Val Lys Gly Gln Ile Ala
[1883]	340 345 350
[1884]	Ala Thr Leu His Leu Pro Gly Gly Asp Ile Thr Pro Arg Leu Arg Arg
[1885]	355 360 365
[1886]	Ser Lys Tyr Phe Gln Asn Pro Glu Ile Ala Pro Val Lys Asp Gly Lys
[1887]	370 375 380
[1888]	Gly Lys Val Asn Gly Phe Gln Ile Ser Phe Lys Arg Gly Lys Thr Pro
[1889]	385 390 395 400

[1890]	Leu Val Gly Ile Ile Lys Glu Pro Lys Leu Leu Lys Lys Asn Gly Asn
[1891]	405 410 415
[1892]	Tyr Tyr Leu Ser Leu Ala Val Gly Ile Asn Lys Thr Glu Ile Pro Lys
[1893]	420 425 430
[1894]	Glu Ile Cys Asp Ala Arg Ala Tyr Tyr Ser Ser Thr Ser Arg Thr Asp
[1895]	435 440 445
[1896]	Thr Pro Pro Ala Val Lys Ala Met Ser Ile Asp Leu Gly Val Thr Thr
[1897]	450 455 460
[1898]	Leu Ala Asp Tyr Ser Ile Ile Asp Thr Gly Leu Pro Gly Asp Cys Gly
[1899]	465 470 475 480
[1900]	Val Phe Gly Gly Ser Thr Ala Ala Phe Thr Glu His Gly Lys Ile Gly
[1901]	485 490 495
[1902]	Arg Cys Gly Ser Lys Ser Leu Arg Asp Gly Leu Tyr Lys Asn Thr Glu
[1903]	500 505 510
[1904]	Ala Gly Tyr Phe Leu Ala Lys Tyr Ile Arg Leu Ser Lys Asn Leu Arg
[1905]	515 520 525
[1906]	Gly Gly Val Gly Leu Asn Lys Leu Glu Lys Glu Lys Leu Leu Glu His
[1907]	530 535 540
[1908]	Val Glu Arg Leu Gly Ile Glu His Cys Ala Asp Asp Phe Ala Arg Lys
[1909]	545 550 555 560
[1910]	Asp Glu Ile His Arg Lys Phe Ser Glu Ile Lys Ser Lys Leu Glu Lys
[1911]	565 570 575
[1912]	Ser Ile Ser Glu Phe Ala Leu Arg Asp Arg Pro Asp Lys Lys Gly Ala
[1913]	580 585 590
[1914]	Ser Trp Glu Gly Ile Cys Ala Glu Thr Val Gln Val Leu Gly Ala Val
[1915]	595 600 605
[1916]	Lys Arg Trp Gln Ser Leu Ala Lys Ser Trp Thr Tyr Tyr Ser Trp Thr
[1917]	610 615 620
[1918]	Ala Glu Asp Tyr Val Leu Ala Leu Thr Gly Glu Gly Arg Thr Arg Val
[1919]	625 630 635 640
[1920]	Ser Asp Glu His Val Glu Ser Val Val Lys Thr Gly Arg Arg Gln Phe
[1921]	645 650 655
[1922]	Ala Pro Cys Gly Lys Ala Ala Leu Leu Arg Leu Leu Glu Lys Gly Lys
[1923]	660 665 670
[1924]	Ile Val Glu Val Cys Pro Gly Gln Phe Gln Leu Ala Glu Gly Val Asp
[1925]	675 680 685
[1926]	Tyr Lys Arg His Pro Thr Glu Phe Leu Ala Ala His Ile Arg His Phe
[1927]	690 695 700
[1928]	Asn Gly Leu Arg Arg Asp Leu Thr Asn Lys Leu Val Arg Ala Ile Val
[1929]	705 710 715 720
[1930]	Glu Lys Ala Gln Gln His Arg Val Gln Ile Val Ile Val Glu Asp Phe
[1931]	725 730 735

[1932]	Gly Ile Pro Asp Ile Glu Gly Arg Ile Met Asp His Tyr Asp Asn Tyr
[1933]	740 745 750
[1934]	Arg Trp Asn Leu Phe Ala Pro Ala Lys Val Ile Glu Lys Leu Glu Glu
[1935]	755 760 765
[1936]	Ala Leu Ser Glu Val Gly Ile Ala Met Ala Glu Val Asp Pro Arg His
[1937]	770 775 780
[1938]	Thr Ser Gln Leu Ala Pro Thr Gly Asp Phe Gly Phe Arg Asp His Glu
[1939]	785 790 795 800
[1940]	Asn Leu Tyr Phe Trp Glu Lys Gly Leu Cys Arg Thr Asp Ala Asn Thr
[1941]	805 810 815
[1942]	Asn Ala Ser Met Arg Ile Ala Glu Arg Phe Phe Thr Arg His Ser Val
[1943]	820 825 830
[1944]	Leu Ser Gln Leu Arg Ala Val Lys Ile Ser Glu Thr Glu Phe Leu Ile
[1945]	835 840 845
[1946]	Pro Val Ser Thr Gly Lys Arg Glu Asn Ala Phe Ile Lys Ser Gln Thr
[1947]	850 855 860
[1948]	Gly Lys Leu Phe Ala Lys Leu Val Ala Asp Ser Asn Gly Phe Val Met
[1949]	865 870 875 880
[1950]	Val Gly Leu Thr Glu Lys Gln His Gly Ala Thr Val Thr Val Gly Lys
[1951]	885 890 895
[1952]	Lys Val Ser Phe Tyr Asn His Ala Gly Arg Trp Leu Gly Lys Ala His
[1953]	900 905 910
[1954]	His Ile Ala His Arg Asp Arg Ile Lys Asn Glu Val Asn Gln Val Leu
[1955]	915 920 925
[1956]	Thr Ser Gly Arg Gly Arg Ile Arg Asn Ile Ala Pro Glu Leu Ser Pro
[1957]	930 935 940
[1958]	Lys Thr
[1959]	945
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[1963]	<213> 人工序列
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[1965]	<223> Cas12j.18的氨基酸序列
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[1967]	Met Thr Asn Gln Lys Pro Lys Phe Lys Ser Ser Asp Ile Gln Ile Lys
[1968]	1 5 10 15
[1969]	His Ile Ser Pro Thr Asp Lys Lys Arg Leu Lys Thr Phe Tyr His Gln
[1970]	20 25 30
[1971]	Leu Tyr Glu Gln Val Asn Phe Ile Leu Glu Arg Met Ile Val Met Arg
[1972]	35 40 45
[1973]	Gly Arg Pro Arg Thr Ile Arg Asn Ile Asp Gly Thr Glu Ile Phe Val

[1974]	50	55	60
[1975]	Ser Gln Glu Glu Ala Asp Gln Gln Leu Leu Ser Leu Ala Gly Gly Ser		
[1976]	65	70	75 80
[1977]	His Glu Gly Val Lys Tyr Leu Lys Gln Tyr Tyr Glu Ser Cys Val Asp		
[1978]		85	90 95
[1979]	Ala Gly Lys Pro Ala Lys Tyr Ala Ala Asn Met Phe Leu Thr Lys Thr		
[1980]		100	105 110
[1981]	Ile Ser Gly Thr Asn Pro Leu Gln Cys His Thr Ala Val Tyr Lys Leu		
[1982]		115	120 125
[1983]	Tyr Lys Lys Val Gln Ala Lys Gln Ile Thr Lys Lys Glu Phe Ile Asp		
[1984]		130	135 140
[1985]	Lys Leu Tyr Ser Lys Thr Lys Lys Lys Lys Ser Leu Lys Pro Ala Tyr		
[1986]		145	150 155 160
[1987]	Lys Val Phe Thr Glu Asn Glu His Ile Glu Phe Tyr His Lys Val Arg		
[1988]		165	170 175
[1989]	Ser Gly Lys Leu Pro Ala Ser Glu Val Arg Leu Glu Glu Ser Arg Arg		
[1990]		180	185 190
[1991]	Ala Pro Asp Val Gly Leu Glu Val Gly Leu Leu Leu Arg Glu Leu Gly		
[1992]		195	200 205
[1993]	Ile Phe Pro Phe Asn Phe Pro His Phe Thr Glu Lys Lys Tyr Leu Asp		
[1994]		210	215 220
[1995]	Leu Ala Trp Thr Ile Ala Ile Arg Trp Leu Lys Asn Trp Asn Glu Asn		
[1996]		225	230 235 240
[1997]	Asn Lys Asn Thr Ala Lys Glu Lys Ala Lys Gln Lys Ala Ile Val Asp		
[1998]		245	250 255
[1999]	Lys Leu Arg Thr Ser Leu Asp Gln Lys Glu Val Asp Leu Phe Glu Glu		
[2000]		260	265 270
[2001]	Phe Ala Glu Glu Cys Ser Gln Glu Gln Phe Gly Ile Arg Glu Gly Phe		
[2002]		275	280 285
[2003]	Val Lys Ala Lys Lys Arg Leu Lys Ser Phe Pro Lys Gly Ile Glu Lys		
[2004]		290	295 300
[2005]	Ser Ser Tyr Lys Glu Gly Met Arg Ile Leu Val Gln Asn Lys His Gly		
[2006]		305	310 315 320
[2007]	Ser Ile Trp Asp Asn Phe Glu Asn Leu Ala Tyr His His Ile Ala Leu		
[2008]		325	330 335
[2009]	Asn Glu Tyr Asn Arg Leu Arg Asp Glu Ala Ser Phe Ser Phe Pro Asp		
[2010]		340	345 350
[2011]	Pro Ile Tyr His Pro Ile Arg Ala Glu Phe Gly Leu Thr Ser Leu Pro		
[2012]		355	360 365
[2013]	Lys Phe Asn Val Gly Leu Asn Asp Arg Gly Asn Tyr Glu Phe Thr Ile		
[2014]		370	375 380
[2015]	Asn Leu Pro Asp Gly Pro Leu Met Met Leu Gly Lys Lys Ser Arg Tyr		

[2016]	385	390	395	400
[2017]	Tyr Leu Lys Pro Ile Ile Gln Gly Pro Leu Asn Asn Ala Phe Ser Phe			
[2018]		405	410	415
[2019]	Glu Phe Ile Lys Gly Asn Lys Lys Arg Pro Lys Ile Ser Ala Lys Leu			
[2020]		420	425	430
[2021]	Lys Ser Ile Thr Val Val Phe Ala Lys Ser Ser Ile Tyr Val Gly Leu			
[2022]		435	440	445
[2023]	Pro Tyr Arg Pro Ile Ser Ile Pro Ile Pro Gln Ala Val Thr Asn Ser			
[2024]		450	455	460
[2025]	Thr Tyr Tyr Phe Lys Lys Asn Leu Ser Ser Thr Ser Lys Phe Asp Lys			
[2026]		465	470	480
[2027]	Asp Val Phe Met Gly Leu Thr Ala Val Ser Val Asp Leu Gly Leu Asn			
[2028]		485	490	495
[2029]	Pro Val Phe Ser Met Ser Ala Cys Arg Leu Asp Glu Met Lys Ala Asp			
[2030]		500	505	510
[2031]	Glu His Tyr Ser Cys Glu Val Pro Gly Phe Gly Trp Ala Asn Gln Ile			
[2032]		515	520	525
[2033]	Trp Ser Lys Arg Ala Gly Gly Val Trp Asn Arg Ser Phe Arg Asp Lys			
[2034]		530	535	540
[2035]	Ile Arg Gly Phe Val Pro Gly Asn Leu Ser Asp Arg Ile Phe Cys Cys			
[2036]		545	550	560
[2037]	Lys Lys Ser Ile Ile Val Ser Lys Lys Leu Arg Asp Glu Lys Pro Leu			
[2038]		565	570	575
[2039]	Thr Gln Tyr Glu Glu Glu Asn Phe Glu Arg Trp Met Gln Val Val Gly			
[2040]		580	585	590
[2041]	Val Asp Pro Asn Glu Asp His Tyr Lys Gln Leu Arg Ile Ala Ile Arg			
[2042]		595	600	605
[2043]	Asp Ile Lys Thr Glu Tyr Glu Thr Val Arg Ser Glu Phe Ala Leu Arg			
[2044]		610	615	620
[2045]	Asp His Pro Asn Asn Ser Asn Lys Thr Thr Glu Asn Ile Cys Thr Glu			
[2046]		625	630	640
[2047]	Cys Phe Asp Met Leu Phe Val Ile Lys Asn Leu Ile Ser Leu Leu Lys			
[2048]		645	650	655
[2049]	Ser Trp Asn Arg Trp His Arg Thr Thr Gly Asp Ile Glu Glu Arg Gly			
[2050]		660	665	670
[2051]	Lys Asp Pro Asn Glu Cys Ser Thr Tyr Trp Arg His Tyr Asn Gly Leu			
[2052]		675	680	685
[2053]	Lys Thr Asp Leu Leu Lys Lys Leu Thr Asn Ile Leu Ile Glu Ser Ala			
[2054]		690	695	700
[2055]	Lys Ser Ile Gly Ala His Ile Ile Ile Leu Glu Asp Leu Thr Leu Ser			
[2056]		705	710	720
[2057]	Gln Arg Ser Ser Arg Ser Arg Arg Glu Asn Ser Leu Val Ala Ile Phe			

[2058]		725		730		735
[2059]	Gly Ala Gln Thr Ile Ile Lys Thr Ile Ser Glu Glu Ala Glu Ile Asn					
[2060]		740		745		750
[2061]	Gly Ile Leu Val Tyr Leu Glu Asp Pro Arg His Ser Ser Gln Ile Ser					
[2062]		755		760		765
[2063]	Ile Val Thr Asn Glu Phe Gly Tyr Arg Pro Lys Glu Asp Lys Ala Lys					
[2064]		770		775		780
[2065]	Leu Tyr Phe Met Asp Glu Glu Thr Val Cys Val Thr Asn Cys Asp Asp					
[2066]		785		790		795
[2067]	Ser Ala Ala Leu Met Leu Gln Gln Ser Phe Trp Ser Arg His Lys Asp					
[2068]		805		810		815
[2069]	Val Val Lys Val Lys Gly Thr Lys Val Ser Asp Thr Glu Tyr Leu Val					
[2070]		820		825		830
[2071]	Ser Ser Glu Asp Lys Asp Gly Thr Lys Met Arg Leu Arg Ser Tyr Leu					
[2072]		835		840		845
[2073]	Lys Arg Asn Val Gly Thr Ala Asn Ala Ile Leu Gln Lys Asn Cys Asp					
[2074]		850		855		860
[2075]	Gly Tyr Asp Leu Lys Lys Ile Ser Pro Gln Lys Lys Lys Lys Ile Glu					
[2076]		865		870		875
[2077]	Glu Phe Gly Lys Asp Glu Tyr Phe Tyr Arg His Gly Glu Gln Trp Phe					
[2078]		885		890		895
[2079]	Thr Ala Asp Ala His Phe Asp Lys Leu Arg Glu Phe Gly Asn Gln Val					
[2080]		900		905		910
[2081]	Phe Leu Thr Pro Gln Ser Gln Ile Lys Arg Ile Asn Leu Gln Val Glu					
[2082]		915		920		925
[2083]	Gly Thr					
[2084]		930				
[2085]	<210> 17					
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[2089]	<220>					
[2090]	<223> Cas12j.19的氨基酸序列					
[2091]	<400> 17					
[2092]	Met Pro Ser Tyr Lys Ser Ser Arg Val Leu Val Arg Asp Val Pro Glu					
[2093]	1	5		10		15
[2094]	Glu Leu Val Asp His Tyr Glu Arg Ser His Arg Val Ala Ala Phe Phe					
[2095]		20		25		30
[2096]	Met Arg Leu Leu Leu Ala Met Arg Arg Glu Pro Tyr Ser Leu Arg Met					
[2097]		35		40		45
[2098]	Arg Asp Gly Thr Glu Arg Glu Val Asp Leu Asp Glu Thr Asp Asp Phe					
[2099]		50		55		60

[2100]	Leu Arg Ser Ala Gly Cys Glu Glu Pro Asp Ala Val Ser Asp Asp Leu
[2101]	65 70 75 80
[2102]	Arg Ser Phe Ala Leu Ala Val Leu His Gln Asp Asn Pro Lys Lys Arg
[2103]	85 90 95
[2104]	Ala Phe Leu Glu Ser Glu Asn Cys Val Ser Ile Leu Cys Leu Glu Lys
[2105]	100 105 110
[2106]	Ser Ala Ser Gly Thr Arg Tyr Tyr Lys Arg Pro Gly Tyr Gln Leu Leu
[2107]	115 120 125
[2108]	Lys Lys Ala Ile Glu Glu Glu Trp Gly Trp Asp Lys Phe Glu Ala Ser
[2109]	130 135 140
[2110]	Leu Leu Asp Glu Arg Thr Gly Glu Val Ala Glu Lys Phe Ala Ala Leu
[2111]	145 150 155 160
[2112]	Ser Met Glu Asp Trp Arg Arg Phe Phe Ala Ala Arg Asp Pro Asp Asp
[2113]	165 170 175
[2114]	Leu Gly Arg Glu Leu Leu Lys Thr Asp Thr Arg Glu Gly Met Ala Ala
[2115]	180 185 190
[2116]	Ala Leu Arg Leu Arg Glu Arg Gly Val Phe Pro Val Ser Val Pro Glu
[2117]	195 200 205
[2118]	His Leu Asp Leu Asp Ser Leu Lys Ala Ala Met Ala Ser Ala Ala Glu
[2119]	210 215 220
[2120]	Arg Leu Lys Ser Trp Leu Ala Cys Asn Gln Arg Ala Val Asp Glu Lys
[2121]	225 230 235 240
[2122]	Ser Glu Leu Arg Lys Arg Phe Glu Glu Ala Leu Asp Gly Val Asp Pro
[2123]	245 250 255
[2124]	Glu Lys Tyr Ala Leu Phe Glu Lys Phe Ala Ala Glu Leu Gln Gln Ala
[2125]	260 265 270
[2126]	Asp Tyr Asn Val Thr Lys Lys Leu Val Leu Ala Val Ser Ala Lys Phe
[2127]	275 280 285
[2128]	Pro Ala Thr Glu Pro Ser Glu Phe Lys Arg Gly Val Glu Ile Leu Lys
[2129]	290 295 300
[2130]	Glu Asp Gly Tyr Lys Pro Leu Trp Glu Asp Phe Arg Glu Leu Gly Phe
[2131]	305 310 315 320
[2132]	Val Tyr Leu Ala Glu Arg Lys Trp Glu Arg Arg Arg Gly Gly Ala Ala
[2133]	325 330 335
[2134]	Val Thr Leu Cys Asp Ala Asp Asp Ser Pro Ile Lys Val Arg Phe Gly
[2135]	340 345 350
[2136]	Leu Thr Gly Arg Gly Arg Lys Phe Val Leu Ser Ala Ala Gly Ser Arg
[2137]	355 360 365
[2138]	Phe Leu Ile Thr Val Lys Leu Pro Cys Gly Asp Val Gly Leu Thr Ala
[2139]	370 375 380
[2140]	Val Pro Ser Arg Tyr Phe Trp Asn Pro Ser Val Gly Arg Thr Thr Ser
[2141]	385 390 395 400

[2142]	Asn Ser Phe Arg Ile Glu Phe Thr Lys Arg Thr Thr Glu Asn Arg Arg		
[2143]		405	410 415
[2144]	Tyr Val Gly Glu Val Lys Glu Ile Gly Leu Val Arg Gln Arg Gly Arg		
[2145]		420	425 430
[2146]	Tyr Tyr Phe Phe Ile Asp Tyr Asn Phe Asp Pro Glu Glu Val Ser Asp		
[2147]		435	440 445
[2148]	Glu Thr Lys Val Gly Arg Ala Phe Phe Arg Ala Pro Leu Asn Glu Ser		
[2149]		450	455 460
[2150]	Arg Pro Lys Pro Lys Asp Lys Leu Thr Val Met Gly Ile Asp Leu Gly		
[2151]		465	470 475 480
[2152]	Ile Asn Pro Ala Phe Ala Phe Ala Val Cys Thr Leu Gly Glu Cys Gln		
[2153]		485	490 495
[2154]	Asp Gly Ile Arg Ser Pro Val Ala Lys Met Glu Asp Val Ser Phe Asp		
[2155]		500	505 510
[2156]	Ser Thr Gly Leu Arg Gly Gly Ile Gly Ser Gln Lys Leu His Arg Glu		
[2157]		515	520 525
[2158]	Met His Asn Leu Ser Asp Arg Cys Phe Tyr Gly Ala Arg Tyr Ile Arg		
[2159]		530	535 540
[2160]	Leu Ser Lys Lys Leu Arg Asp Arg Gly Ala Leu Asn Asp Ile Glu Ala		
[2161]		545	550 555 560
[2162]	Arg Leu Leu Glu Glu Lys Tyr Ile Pro Gly Phe Arg Ile Val His Ile		
[2163]		565	570 575
[2164]	Glu Asp Ala Asp Glu Arg Arg Arg Thr Val Gly Arg Thr Val Lys Glu		
[2165]		580	585 590
[2166]	Ile Lys Gln Glu Tyr Lys Arg Ile Arg His Gln Phe Tyr Leu Arg Tyr		
[2167]		595	600 605
[2168]	His Thr Ser Lys Arg Asp Arg Thr Glu Leu Ile Ser Ala Glu Tyr Phe		
[2169]		610	615 620
[2170]	Arg Met Leu Phe Leu Val Lys Asn Leu Arg Asn Leu Leu Lys Ser Trp		
[2171]		625	630 635 640
[2172]	Asn Arg Tyr His Trp Thr Thr Gly Asp Arg Glu Arg Arg Gly Gly Asn		
[2173]		645	650 655
[2174]	Pro Asp Glu Leu Lys Ser Tyr Val Arg Tyr Tyr Asn Asn Leu Arg Met		
[2175]		660	665 670
[2176]	Asp Thr Leu Lys Lys Leu Thr Cys Ala Ile Val Arg Thr Ala Lys Glu		
[2177]		675	680 685
[2178]	His Gly Ala Thr Leu Val Ala Met Glu Asn Ile Gln Arg Val Asp Arg		
[2179]		690	695 700
[2180]	Asp Asp Glu Val Lys Arg Arg Lys Glu Asn Ser Leu Leu Ser Leu Trp		
[2181]		705	710 715 720
[2182]	Ala Pro Gly Met Val Leu Glu Arg Val Glu Gln Glu Leu Lys Asn Glu		
[2183]		725	730 735

[2184]	Gly Ile Leu Ala Trp Glu Val Asp Pro Arg His Thr Ser Gln Thr Ser
[2185]	740 745 750
[2186]	Cys Ile Thr Asp Glu Phe Gly Tyr Arg Ser Leu Val Ala Lys Asp Thr
[2187]	755 760 765
[2188]	Phe Tyr Phe Glu Gln Asp Arg Lys Ile His Arg Ile Asp Ala Asp Val
[2189]	770 775 780
[2190]	Asn Ala Ala Ile Asn Ile Ala Arg Arg Phe Leu Thr Arg Tyr Arg Ser
[2191]	785 790 795 800
[2192]	Leu Thr Gln Leu Trp Ala Ser Leu Leu Asp Asp Gly Arg Tyr Leu Val
[2193]	805 810 815
[2194]	Asn Val Thr Arg Gln His Glu Arg Ala Tyr Leu Glu Leu Gln Thr Gly
[2195]	820 825 830
[2196]	Ala Pro Ala Ala Thr Leu Asn Pro Thr Ala Glu Ala Ser Tyr Glu Leu
[2197]	835 840 845
[2198]	Val Gly Leu Ser Pro Glu Glu Glu Glu Leu Ala Gln Thr Arg Ile Lys
[2199]	850 855 860
[2200]	Arg Lys Lys Arg Glu Pro Phe Tyr Arg His Glu Gly Val Trp Leu Thr
[2201]	865 870 875 880
[2202]	Arg Glu Lys His Arg Glu Gln Val His Glu Leu Arg Asn Gln Val Leu
[2203]	885 890 895
[2204]	Ala Leu Gly Asn Ala Lys Ile Pro Glu Ile Arg Thr
[2205]	900 905
[2206]	<210> 18
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[2212]	<400> 18
[2213]	Met Ala Phe Gln Ser Lys Arg Arg Ile Val Gly Asn Phe Val Lys Glu
[2214]	1 5 10 15
[2215]	Gln Cys Leu Lys Ala Val Asp Gly Lys Val Ile Leu Thr Asp Gln Glu
[2216]	20 25 30
[2217]	Lys Arg Glu Leu Ile Lys Arg Tyr Glu Leu His Leu Glu Pro His Lys
[2218]	35 40 45
[2219]	Trp Leu Leu Arg Leu Phe Leu Ser Gly Tyr Glu Gly Arg Asp Asp Gly
[2220]	50 55 60
[2221]	Phe Tyr Glu Glu Leu Gly Asn Thr Asn Leu Asp Lys Glu Lys Phe Phe
[2222]	65 70 75 80
[2223]	Glu Val Thr Ala Gly Leu Arg Asp Ala Leu Leu Arg Gln Ser Gly Ser
[2224]	85 90 95
[2225]	Ser Arg Ala Leu Lys Ser Ser Met Leu Gly Lys Cys Pro Pro Ser Ala

[2226]	100	105	110
[2227]	Ala Val Gly Lys Ala Ala Lys His Ile Gln Thr Leu Arg Asp Ala Gly		
[2228]	115	120	125
[2229]	Ile Leu Pro Phe Lys Thr Gly Leu Thr Ser Gly Glu Asp Tyr Asn Val		
[2230]	130	135	140
[2231]	Leu Gln Gln Ala Val Gln Gln Leu Arg Ser Trp Val Ala Cys Asp His		
[2232]	145	150	155
[2233]	Arg Thr Arg Glu Ala Tyr Ala Glu Gln Gln Glu Lys Thr Ser Gln Ala		
[2234]	165	170	175
[2235]	Glu Glu Ala Ala Lys Lys Ala Ala Asn Glu Val Lys Pro Glu Asp Ala		
[2236]	180	185	190
[2237]	Lys Ser Leu Glu Arg His Glu Arg Val Leu Thr Lys Leu Arg Lys Gln		
[2238]	195	200	205
[2239]	Glu Arg Arg Leu Glu Arg Met Lys Ser His Ala Gln Phe Ser Leu Asp		
[2240]	210	215	220
[2241]	Glu Met Asp Cys Thr Gly Tyr Ser Leu Cys Met Gly Ala Asn Tyr Leu		
[2242]	225	230	235
[2243]	Lys Asp Tyr Cys Leu Glu Lys Glu Gly Arg Gly Leu Arg Leu Thr Leu		
[2244]	245	250	255
[2245]	Lys Asn Ser Thr Met Ala Gly Ser Tyr Tyr Val Ser Val Gly Asp Gly		
[2246]	260	265	270
[2247]	Gln His Ala Gly Met Lys Asn Pro Gly Thr Pro Ala Gly Gly Ser Pro		
[2248]	275	280	285
[2249]	Glu Lys Gly Arg Arg Arg Asn Ile Leu Phe Asp Phe Thr Val Glu Lys		
[2250]	290	295	300
[2251]	Cys Gly Asp Asn Tyr Leu Phe Arg Tyr Asp Glu Asn Gly Lys Arg Pro		
[2252]	305	310	315
[2253]	Arg Ala Gly Val Val Lys Glu Pro Arg Phe Cys Trp Arg Arg Lys Gly		
[2254]	325	330	335
[2255]	Asn Ser Val Glu Leu Tyr Leu Ala Met Pro Ile Asn Ile Glu Asn Ser		
[2256]	340	345	350
[2257]	Met Arg Asn Ile Phe Val Gly Lys Gln Lys Ser Gly Lys His Ser Ala		
[2258]	355	360	365
[2259]	Phe Thr Arg Gln Trp Pro Lys Glu Val Glu Gly Leu Asp Glu Leu Arg		
[2260]	370	375	380
[2261]	Asp Ala Val Val Leu Gly Val Asp Ile Gly Ile Asn Arg Ala Ala Phe		
[2262]	385	390	395
[2263]	Cys Ala Ala Leu Lys Thr Ser Arg Phe Glu Asn Gly Leu Pro Ala Asp		
[2264]	405	410	415
[2265]	Val Gln Val Met Asp Thr Thr Cys Asp Ala Leu Thr Glu Lys Gly Gln		
[2266]	420	425	430
[2267]	Glu Tyr Arg Gln Leu Arg Lys Asp Ala Thr Cys Leu Ala Trp Leu Ile		

[2268]	435	440	445
[2269]	Arg Thr Thr Arg Arg Phe Lys Ala Asp Pro Gly Asn Lys His Asn Gln		
[2270]	450	455	460
[2271]	Ile Lys Glu Lys Asp Val Glu Arg Phe Asp Ser Ala Asp Gly Ala Tyr		
[2272]	465	470	475
[2273]	Arg Arg Tyr Met Asp Ala Ile Ala Glu Met Pro Ser Asp Pro Leu Gln		
[2274]		485	490
[2275]	Val Trp Glu Ala Ala Arg Ile Thr Gly Tyr Gly Glu Trp Ala Lys Glu		495
[2276]		500	505
[2277]	Ile Phe Ala Arg Phe Asn His Tyr Lys His Glu His Ala Cys Cys Ala		510
[2278]		515	520
[2279]	Val Ser Leu Ser Leu Ser Asp Arg Leu Val Trp Cys Arg Leu Ile Asp		525
[2280]		530	535
[2281]	Arg Ile Leu Ser Leu Lys Lys Cys Leu His Phe Gly Gly Tyr Glu Ser		540
[2282]	545	550	555
[2283]	Lys His Arg Lys Gly Phe Cys Lys Ser Leu Tyr Arg Leu Arg His Asn		560
[2284]		565	570
[2285]	Ala Arg Asn Asp Val Arg Lys Lys Leu Ala Arg Phe Ile Val Asp Ala		575
[2286]		580	585
[2287]	Ala Val Asp Ala Gly Ala Ser Val Ile Ala Met Glu Lys Leu Pro Ser		590
[2288]		595	600
[2289]	Ser Gly Gly Lys Gln Ser Lys Asp Asp Asn Arg Ile Trp Asp Leu Met		605
[2290]		610	615
[2291]	Ala Pro Asn Thr Leu Ala Thr Thr Val Cys Leu Met Ala Lys Val Glu		620
[2292]	625	630	635
[2293]	Gly Ile Gly Phe Val Gln Val Asp Pro Glu Phe Thr Ser Gln Trp Val		640
[2294]		645	650
[2295]	Phe Glu Gln Arg Val Ile Gly Asp Arg Glu Gly Arg Ile Val Ser Cys		655
[2296]		660	665
[2297]	Leu Asp Ala Glu Gly Val Arg Arg Asp Tyr Asp Ala Asp Glu Asn Ala		670
[2298]		675	680
[2299]	Ala Lys Asn Ile Ala Trp Leu Ala Leu Thr Arg Glu Ala Glu Pro Phe		685
[2300]		690	695
[2301]	Cys Met Ala Phe Glu Lys Arg Asn Gly Val Val Glu Pro Lys Gly Leu		700
[2302]	705	710	715
[2303]	Arg Phe Asp Ile Pro Glu Glu Pro Thr Arg Glu Gln Asp Glu Ser Asp		720
[2304]		725	730
[2305]	Gln Asp Phe Lys Lys Arg Leu Glu Glu Arg Asp Lys Leu Ile Glu Arg		735
[2306]		740	745
[2307]	Leu Gln Ala Lys Ala Asp Arg Met Gln Ala Ile Val Gln Arg Leu Phe		750
[2308]		755	760
[2309]	Gly Asp Arg Arg Pro Trp Asp Ala Phe Ala Asp Arg Ile Pro Glu Gly		765

[2310]	770	775	780
[2311]	Lys Ser Lys Arg Leu Phe Arg His Arg Asp Gly Leu Val Leu Asn Lys		
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[2313]	Pro Phe Lys Gly Leu Cys Gly Ser Glu Asn Ser Glu Gln Lys Ala Ser		
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[2315]	Ala Arg Asn Ser Arg		
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[2326]	Gln Asp Cys Ile Lys Thr Ile Ser Ala Lys Cys Leu Leu Thr Arg Ala		
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[2328]	Gln Ile Asp Glu Leu Arg Ala Lys Tyr Asp Ala Val Leu Asp Thr Met		
[2329]		35	40 45
[2330]	Arg Pro Leu Ile Arg Leu Ile Leu Ala Gly Tyr Glu Gly Arg Asp Asp		
[2331]		50	55 60
[2332]	Gly Ile Tyr Glu Glu Ile Ala Pro Glu Met Ser Lys Lys Lys Phe Phe		
[2333]	65	70	75 80
[2334]	Glu Ala Ala Thr Glu Trp Arg Glu Ser Ile Val Lys Asn Ala Ser Pro		
[2335]		85	90 95
[2336]	Arg Ala Met Lys Ala Ser Val Phe Gly Asp Lys Glu Pro Cys Lys Ser		
[2337]		100	105 110
[2338]	Thr Gly Gly Ala Arg Ala Val Ile Gly Lys Leu Arg Lys Ser Gly Val		
[2339]		115	120 125
[2340]	Phe Pro Ile Glu Thr Gly Leu Ser Gly Gly Asp Glu Tyr Asn Leu Ile		
[2341]		130	135 140
[2342]	Glu Gln Ala Ile Glu Tyr Ala Lys Ser Trp Leu Lys Ser Asp Glu Ala		
[2343]	145	150	155 160
[2344]	Thr Arg Glu Ala Tyr Ala Asp Gln Gln Lys Asp Ile Lys Arg Leu Ile		
[2345]		165	170 175
[2346]	Gly Glu Ala Lys Lys Leu Ala Leu Lys Ile Glu Lys Ala Glu Lys Lys		
[2347]		180	185 190
[2348]	Leu Glu Ala Thr Asn Pro Gln Thr Lys Ser Trp Lys Lys Thr Thr Glu		
[2349]		195	200 205
[2350]	Ile Ile Lys Lys Ser Lys Arg Glu Phe Gly Ser Val Thr Thr Lys Thr		
[2351]		210	215 220

[2352]	Glu Lys Ala Glu Lys Arg Phe Glu Arg Met Lys Pro Phe Ser Lys Leu
[2353]	225 230 235 240
[2354]	Glu Leu Gln Asn Met Asp Cys Thr Lys Tyr Ser Thr Tyr Leu Gly Thr
[2355]	245 250 255
[2356]	Asn Tyr Ser Pro Phe Lys Leu Lys Lys Glu Gly Asp Leu Leu Gln Ile
[2357]	260 265 270
[2358]	Thr Val Thr Ser Ser Val Met Lys Gly Thr Tyr Leu Ala Ser Tyr Gly
[2359]	275 280 285
[2360]	Asp Gly Gln Tyr Gly Ser Arg Arg Asn Asn Gly Gln Ser Arg Arg Asp
[2361]	290 295 300
[2362]	Asp Phe Val Pro Asn Met Asn Gln Lys Arg Arg Arg Asn Leu Met Phe
[2363]	305 310 315 320
[2364]	Asp Cys Thr Val Glu Pro Phe Gly Asp Gly Ser Leu Leu Arg Tyr Glu
[2365]	325 330 335
[2366]	Glu Asn Gly Leu Arg Pro Arg Val Ala Glu Leu Lys Glu Pro Arg Leu
[2367]	340 345 350
[2368]	Cys Trp Arg Arg Arg Asn Gly Asn Tyr Glu Leu Tyr Leu Met Met Pro
[2369]	355 360 365
[2370]	Val Lys Met His Val Lys Ser Pro Glu Met Phe Ala Gly Asp His Leu
[2371]	370 375 380
[2372]	Ala Phe Ser Arg Tyr Trp Pro Lys Glu Val Glu Gly Leu Asp Ser Asp
[2373]	385 390 395 400
[2374]	Thr Lys Ile Thr Ala Leu Gly Val Asp Val Gly Ile Ile Arg Ser Ala
[2375]	405 410 415
[2376]	Tyr Cys Val Ala Val Thr Ala Glu Arg Phe Val Asp Gly Leu Pro Thr
[2377]	420 425 430
[2378]	Glu Met Thr Val Gly Lys Ala Ser Phe Asp Ala Gln Thr Glu Lys Gly
[2379]	435 440 445
[2380]	Arg Glu Tyr Phe Glu Leu Gly Arg Arg Ala Thr Met Leu Gly Trp Leu
[2381]	450 455 460
[2382]	Ile Lys Thr Thr Arg Arg Tyr Lys Lys Asp Pro Lys Asn Glu His Asn
[2383]	465 470 475 480
[2384]	Gln Ile Lys Glu Ser Asp Val Ala Ala Phe Asp Gly Ser Pro Gly Ala
[2385]	485 490 495
[2386]	Phe Glu His Tyr Ile Leu Ala Val Asp Glu Met Ser Asp Asp Pro Leu
[2387]	500 505 510
[2388]	Asp Val Trp Gly His Ala Asn Ile Thr Gly Tyr Gly Lys Trp Thr Lys
[2389]	515 520 525
[2390]	Gln Ile Phe Lys Glu Phe Asn Gln Leu Lys Arg Glu Arg Ala Glu Gly
[2391]	530 535 540
[2392]	Gln Val Glu Pro Asn Met Thr Asp Asp Leu Thr Trp Cys Ser Leu Ile
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- [3670] <223> Cas12j.3原型同向重复序列
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- [3673] <210> 42
- [3674] <211> 36
- [3675] <212> RNA
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- [3678] <223> Cas12j.4原型同向重复序列
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- [3680] gugucaaugc gaugcugaac aucgcaugag uaacac 36
- [3681] <210> 43
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- [3683] <212> RNA
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- [3686] <223> Cas12j.5原型同向重复序列
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- [3906] <211> 36
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[3926] <223> Cas12j.15原型同向重复序列的编码核酸序列
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[3928] gtgcagccta ttgggatcgc ccatagggat gagacac 37
[3929] <210> 74
[3930] <211> 36
[3931] <212> DNA
[3932] <213> 人工序列
[3933] <220>
[3934] <223> Cas12j.16原型同向重复序列的编码核酸序列
[3935] <400> 74
[3936] gtgccgtcac cgccttagtt gagcggggtc aagcac 36
[3937] <210> 75
[3938] <211> 37
[3939] <212> DNA
[3940] <213> 人工序列
[3941] <220>
[3942] <223> Cas12j.17原型同向重复序列的编码核酸序列
[3943] <400> 75
[3944] gtgccaacct caccggagac gagtggggca ccagcac 37
[3945] <210> 76
[3946] <211> 36
[3947] <212> DNA

- [3948] <213> 人工序列
[3949] <220>
[3950] <223> Cas12j.18原型同向重复序列的编码核酸序列
[3951] <400> 76
[3952] gtgccgctgg ctttcgaag aggggccttt aagcac 36
[3953] <210> 77
[3954] <211> 36
[3955] <212> DNA
[3956] <213> 人工序列
[3957] <220>
[3958] <223> Cas12j.19原型同向重复序列的编码核酸序列
[3959] <400> 77
[3960] gtgctgctgt ctcccagacg ggaggcagaa ctgcac 36
[3961] <210> 78
[3962] <211> 36
[3963] <212> DNA
[3964] <213> 人工序列
[3965] <220>
[3966] <223> Cas12j.20原型同向重复序列的编码核酸序列
[3967] <400> 78
[3968] gtgtaggcct cctctgaatg gggtaggctaa tgacac 36
[3969] <210> 79
[3970] <211> 36
[3971] <212> DNA
[3972] <213> 人工序列
[3973] <220>
[3974] <223> Cas12j.21原型同向重复序列的编码核酸序列
[3975] <400> 79
[3976] gtgttgatcc gttctgaatg gatggattgc tgacac 36
[3977] <210> 80
[3978] <211> 36
[3979] <212> DNA
[3980] <213> 人工序列
[3981] <220>
[3982] <223> Cas12j.22原型同向重复序列的编码核酸序列
[3983] <400> 80
[3984] atttcagtgc tggcctgtgg aagcaggctc tgtcac 36
[3985] <210> 81
[3986] <211> 11
[3987] <212> PRT
[3988] <213> 人工序列
[3989] <220>

[3990] <223> NLS序列
 [3991] <400> 81
 [3992] Ser Arg Ala Asp Pro Lys Lys Lys Arg Lys Val
 [3993] 1 5 10
 [3994] <210> 82
 [3995] <211> 1014
 [3996] <212> PRT
 [3997] <213> 人工序列
 [3998] <220>
 [3999] <223> Cas12j.3-NLS融合蛋白的氨基酸序列
 [4000] <400> 82
 [4001] Met Thr Lys Glu Lys Ile Lys Lys Thr Lys Lys Ala Lys Val Glu Lys
 [4002] 1 5 10 15
 [4003] Asp Ser Val Thr Arg Ala Gly Ile Leu Arg Ile Leu Leu Asn Pro Asp
 [4004] 20 25 30
 [4005] Gln His Gln Glu Leu Asp Thr Leu Ile Ser Asp His Gln Glu Ala Ala
 [4006] 35 40 45
 [4007] Arg Glu Ile Gln Thr Ala Thr Tyr Lys Leu Ser Gly Leu Lys Leu Tyr
 [4008] 50 55 60
 [4009] Asp Lys Thr Asn Asn Met Val Val Asp Gly Ser Lys Ala Thr Pro Glu
 [4010] 65 70 75 80
 [4011] Glu Gln Glu Ala Tyr Tyr Lys Ile Ile Asn Trp Glu Gly Gln Pro Ile
 [4012] 85 90 95
 [4013] Ser Ile Ser Asn Pro Met Val Arg Ala Thr Phe Lys Ser Ile Ala Lys
 [4014] 100 105 110
 [4015] Val Lys Glu Asp Ile Arg Arg Lys Gln Glu Glu Tyr Ala Lys Leu Glu
 [4016] 115 120 125
 [4017] Glu Ala Asp Leu Thr Lys Met Ser Thr Gly Asp Val Lys Lys His Lys
 [4018] 130 135 140
 [4019] Asn Glu Leu Arg Lys Ala Ala Asn Arg Ile Lys His Ser Glu Glu Ile
 [4020] 145 150 155 160
 [4021] Leu Gln Phe Ala Lys Trp Arg Leu Ala Asp Ile Phe Pro Leu Pro Leu
 [4022] 165 170 175
 [4023] Ser His Asn Ser Gln Leu His Leu Lys Asn Asn Tyr His Gln Asn Val
 [4024] 180 185 190
 [4025] Phe Ser Gly Phe His Ala Arg Val Lys Gly Trp Asn Ala Cys Asp Ile
 [4026] 195 200 205
 [4027] Ala Ala Gln Ala Asn Tyr Ala Glu Ile Asp Asn Arg Leu Thr Glu Leu
 [4028] 210 215 220
 [4029] Ser Ser Glu Leu Ser Gly Asp Tyr Gly Ser Glu Val Ile Thr Asp Leu
 [4030] 225 230 235 240
 [4031] Met Gly Leu Leu Gln Tyr Thr Lys Glu Leu Gly Glu Gly Tyr Thr Asp

[4032]		245		250		255
[4033]	Thr Ser Tyr Leu Asn Tyr Lys Phe Leu Ser Phe Phe Lys Glu Cys Trp					
[4034]		260		265		270
[4035]	Arg Pro Asn Ala Ile Ala Asn Asn Thr Gly Leu Leu Glu Gly Phe Trp					
[4036]		275		280		285
[4037]	Leu Ala Asn Asn Lys His Thr Asn Lys Lys Asn Gln Val Ala Tyr Ser					
[4038]		290		295		300
[4039]	Phe Asn Pro Lys Ile Ser Glu Glu Leu Phe Arg Arg Arg Ser Leu Trp					
[4040]		305		310		315
[4041]	Glu Ser Asp Lys Cys Leu Leu Ser Asp Pro Arg Phe Glu Lys Tyr Val					
[4042]		325		330		335
[4043]	Glu Leu Phe Asp Lys His Gly Arg Tyr Arg Lys Gly Ala Ser Leu Thr					
[4044]		340		345		350
[4045]	Leu Ile Ser Lys Glu Ser Pro Ile Pro Ile Gly Phe Ser Met Asp Arg					
[4046]		355		360		365
[4047]	Asn Ala Ala Lys Leu Val Arg Ile Asp Asn Asp Thr Ala Asn Arg Gln					
[4048]		370		375		380
[4049]	Leu Thr Ile Thr Ile Glu Leu Pro Asn Lys Glu Glu Arg Ser Tyr Val					
[4050]		385		390		395
[4051]	Ala Ala Tyr Gly Arg Lys His Glu Thr Lys Cys Tyr Tyr Asn Gly Leu					
[4052]		405		410		415
[4053]	Thr Thr Arg Leu Pro Arg Ser Glu Lys Glu Leu Leu Ala Leu Ala Lys					
[4054]		420		425		430
[4055]	Ala Glu Asn Arg Glu Leu Thr Asp Lys Glu Ile His Glu Ala Ser Leu					
[4056]		435		440		445
[4057]	Glu Lys Cys Tyr Ile Phe Glu Tyr Ala Arg Ala Gly Lys Ile Pro Val					
[4058]		450		455		460
[4059]	Phe Ala Val Val Lys Thr Leu Tyr Phe Arg Arg Asn Pro Ser Asn Gly					
[4060]		465		470		475
[4061]	Glu Tyr Tyr Val Ile Leu Pro Thr Asn Ile Phe Val Glu Tyr His Ala					
[4062]		485		490		495
[4063]	Asn Asn Glu Phe Asn Ser Lys Glu Leu Phe Lys Ile Arg Ser Glu Leu					
[4064]		500		505		510
[4065]	Gln Lys Ala Trp Asp Glu Val Arg Thr Pro Lys Arg Asn Val Gln Ser					
[4066]		515		520		525
[4067]	Cys Val Leu Asp Lys Asp Leu Ser Lys Arg Phe Ala Gly Arg Thr Leu					
[4068]		530		535		540
[4069]	Lys Tyr Ala Gly Ile Asp Leu Gly Tyr Ser Asn Pro Tyr Thr Val Ser					
[4070]		545		550		555
[4071]	Tyr Tyr Asn Val Val Gly Thr Glu Glu Gly Ile Gln Ile Lys Glu Thr					
[4072]		565		570		575
[4073]	Gly Asn Glu Ile Val Ser Thr Val Phe Asn Glu Gln Tyr Ile Gln Leu					

[4074]		580		585		590	
[4075]	Lys Gly Asn Ile Tyr Gln Leu Ile Asn Ile Ile Arg Ala Ser Arg Arg						
[4076]		595		600		605	
[4077]	Tyr Leu Gln Glu Ser Gly Glu Leu Lys Leu Ser Lys Asp Asp Ile Lys						
[4078]		610		615		620	
[4079]	Ser Phe Asp Gln Leu Met Glu Leu Leu Pro Ser Glu Gln Arg Ile Thr						
[4080]		625		630		635	640
[4081]	Ile Asp Gln Phe Ile Lys Asp Ile Lys Lys Ala Lys Gln Glu Gly Lys						
[4082]		645		650		655	
[4083]	Leu Ile Arg Asp Ile Lys Gly Lys Leu Pro Val Glu Gly Lys Lys Lys						
[4084]		660		665		670	
[4085]	Glu Tyr Trp Val Ile Ser Asn Leu Met Tyr Val Ile Thr Gln Thr Met						
[4086]		675		680		685	
[4087]	Asn Gly Ile Arg Gly Asn Arg Asp Ser Asn Asn His Leu Thr Glu Lys						
[4088]		690		695		700	
[4089]	Lys Asn Trp Leu Ser Ala Pro Pro Leu Ile Glu Leu Ile Asp Ala Tyr						
[4090]		705		710		715	720
[4091]	Tyr Asn Leu Lys Lys Thr Phe Asn Asp Ser Gly Asp Gly Ile Lys Met						
[4092]		725		730		735	
[4093]	Leu Pro Lys Asp His Val Tyr Ala Glu Gly Glu Lys Gln Arg Cys Thr						
[4094]		740		745		750	
[4095]	Leu Arg Glu Glu Asn Phe Cys Lys Gly Ile Leu Glu Trp Arg Asp Asn						
[4096]		755		760		765	
[4097]	Val Lys Asp Tyr Phe Ile Lys Lys Leu Phe Ser Gln Ile Ala His Arg						
[4098]		770		775		780	
[4099]	Cys Tyr Glu Leu Gly Ile Gly Ile Val Ala Met Glu Asn Leu Asp Ile						
[4100]		785		790		795	800
[4101]	Met Gly Ser Ser Lys Asn Thr Lys Gln Ser Asn Arg Met Phe Asn Ile						
[4102]		805		810		815	
[4103]	Trp Pro Arg Gly Gln Met Lys Lys Ser Ala Glu Asp Ala Phe Ser Tyr						
[4104]		820		825		830	
[4105]	Met Gly Ile Leu Ile Gln Tyr Val Asp Glu Asn Gly Thr Ser Arg His						
[4106]		835		840		845	
[4107]	Asp Ala Asp Ser Gly Ile Tyr Gly Cys Arg Asp Gly Ala Asn Leu Trp						
[4108]		850		855		860	
[4109]	Leu Pro Asn Lys Lys Leu His Ala Asp Val Asn Ala Ser Arg Met Ile						
[4110]		865		870		875	880
[4111]	Ala Leu Arg Gly Leu Thr His His Thr Asn Leu Tyr Cys Arg Ser Leu						
[4112]		885		890		895	
[4113]	Thr Glu Ile Glu Asn Gly Lys Tyr Val Asn Thr Tyr Glu Leu Phe Asp						
[4114]		900		905		910	
[4115]	Thr Thr Lys Asn Asp Gln Ser Gly Ala Ala Lys Arg Leu Arg Gly Ala						

[4116]	915	920	925
[4117]	Glu Thr Leu Leu His Gly Tyr Ser Ala Thr Val Tyr Gln Ile His Thr		
[4118]	930	935	940
[4119]	Thr Asn Thr Gly Ala Gly Val Ala Leu Leu Pro Asp Leu Thr Ala Thr		
[4120]	945	950	955 960
[4121]	Asp Val Ile Lys Asn Lys Lys Ile Thr Ala Thr Lys Glu Asn Thr Ala		
[4122]	965	970	975
[4123]	Lys Tyr Tyr Lys Leu Asp Asn Thr Asn Thr Tyr Tyr Pro Trp Ser Val		
[4124]	980	985	990
[4125]	Cys Glu Lys Leu His Lys Asn Trp Lys Leu Ser Ser Arg Ala Asp Pro		
[4126]	995	1000	1005
[4127]	Lys Lys Lys Arg Lys Val		
[4128]	1010		
[4129]	<210> 83		
[4130]	<211> 885		
[4131]	<212> PRT		
[4132]	<213> 人工序列		
[4133]	<220>		
[4134]	<223> Cas12j.4-NLS融合蛋白的氨基酸序列		
[4135]	<400> 83		
[4136]	Met Lys Lys Lys Lys Asn Phe Ser Val Ser Ala Thr Gly Val Phe Ser		
[4137]	1 5 10 15		
[4138]	Phe Pro Thr Thr Glu Ala Lys Met Asp Phe Phe His Arg Phe Ile Glu		
[4139]	20 25 30		
[4140]	Leu Asn Gly Leu Ala Ala Glu Ile Glu Thr His Phe Leu Asn Leu Lys		
[4141]	35 40 45		
[4142]	Asn Asp Lys Asn Gly Glu Ser Val Tyr Asn Lys Val Leu Ser Asn Ser		
[4143]	50 55 60		
[4144]	Asn His Ser Arg Pro Phe Ser Thr Pro Leu Leu Gly Thr Met Thr Gly		
[4145]	65 70 75 80		
[4146]	Ser Thr Lys Val Thr Asp Lys Asn Ala Leu Tyr Gly Asn Asp Leu Asp		
[4147]	85 90 95		
[4148]	His Cys Arg Lys Lys Lys Ile Val Pro Phe Ser Ser Ser Ser Pro Leu		
[4149]	100 105 110		
[4150]	Ser Ser Gln Glu Lys Phe Phe Cys Ile Glu Ala Val Phe Arg Arg Ala		
[4151]	115 120 125		
[4152]	Lys Ser His Met Glu Cys Lys Lys Leu Phe Gln Asp Glu Thr Asn Arg		
[4153]	130 135 140		
[4154]	Met Asp Ser Gln Ile Asn Gly Ile Leu Asn Glu Leu Pro Tyr Gly Val		
[4155]	145 150 155 160		
[4156]	Glu Leu Ser Asn Met Leu Ser Glu Leu Ile Ala Ile Pro Phe Ala Ile		
[4157]	165 170 175		

[4158]	Gly Trp Lys Leu Glu Gly Tyr Leu Gly Gln Val Phe Phe Pro Ser Ile
[4159]	180 185 190
[4160]	Ala Glu Gly Leu Thr Pro Pro Lys Ser Ala Lys Ile Lys Gly Arg Arg
[4161]	195 200 205
[4162]	Arg Ser Ile Asp Tyr Ser Val Thr Asp Glu Ala Tyr Asp Ile Leu Met
[4163]	210 215 220
[4164]	Lys Tyr Ser Asn Leu His Ser Ser Phe Glu Thr Gly Leu Lys Met Ser
[4165]	225 230 235 240
[4166]	Asn Leu Phe Ser Ala Phe Tyr Lys Lys Ser Asn Arg Lys Asp Glu Ile
[4167]	245 250 255
[4168]	Gln Phe Thr Pro Ile Ser Met Glu Ser Arg Cys Asp Leu Leu Leu Gly
[4169]	260 265 270
[4170]	Lys Asn Phe Leu Lys Phe Asp Leu Lys Asn Cys Asp His Arg Ser Gly
[4171]	275 280 285
[4172]	Ser Leu Met Leu Thr Ile Asn Asp Lys Asn Arg Leu Asn Gly Asp Tyr
[4173]	290 295 300
[4174]	Glu Ile Arg Val Gly Ser Asp Lys Lys Asp Ser Tyr Leu Thr Gly Val
[4175]	305 310 315 320
[4176]	Asn Val Thr Asn Leu Gly Asp Asn Val Phe Asn Leu Asn Tyr Lys Val
[4177]	325 330 335
[4178]	Asn Gly Lys Arg Glu Tyr Asn Met Leu Leu Lys Glu Pro Ser Ile His
[4179]	340 345 350
[4180]	Ile Lys Met His Arg Met Arg Asp Asp Gly Asn Tyr Leu Ser Ser Asp
[4181]	355 360 365
[4182]	Phe Asp Phe Tyr Met Ile Phe Ser Met Ser Ser Glu Lys Asp Glu Glu
[4183]	370 375 380
[4184]	Lys Leu Ala Arg Ser Trp Asp Met Arg Ala Ala Met Ser Thr Ala Tyr
[4185]	385 390 395 400
[4186]	Gly Thr Asp Ile Lys Lys Tyr His Ser Ser Phe Pro Cys Arg Ile Leu
[4187]	405 410 415
[4188]	Ala Cys Asp Leu Gly Val Lys His Pro Tyr Ser Ala Ala Val Met Asp
[4189]	420 425 430
[4190]	Ile Gly Gln Leu Asn Glu Asn Gly Met Pro Val Ser Val Asp Lys Val
[4191]	435 440 445
[4192]	His Cys Met His Ser Glu Gly Val Ser Glu Ile Gly Gln Gly Tyr Asn
[4193]	450 455 460
[4194]	His Leu Ile Gln Lys Ile Leu Ala Leu Asn Tyr Ile Leu Ala Tyr Cys
[4195]	465 470 475 480
[4196]	Arg Glu Phe Val Ser Gly Thr Val Asp Asp Phe Asp Lys Ile Asp Tyr
[4197]	485 490 495
[4198]	Lys Leu Ser Gln Leu Ser Tyr Lys Gln Glu Asp Leu Leu Ile Asn Leu
[4199]	500 505 510

[4200]	Gln Glu Met Lys Asp His Phe Gly Asn Asp Met Gln Ala Trp Lys Lys
[4201]	515 520 525
[4202]	Ser Arg Thr Trp Val Val Ser Thr Leu Phe Phe Glu Leu Arg Gln Glu
[4203]	530 535 540
[4204]	Phe Asn Gln Leu Arg Asn Gln Arg Pro Gly Lys Lys Thr Val Ser Leu
[4205]	545 550 555 560
[4206]	Ala Asp Glu Phe Gln Tyr Ile Asp Met Arg Arg Lys Phe Ile Ser Leu
[4207]	565 570 575
[4208]	Ser Arg Ser Tyr Thr Asn Val Gly Arg Gln Ser Ser Lys His Arg His
[4209]	580 585 590
[4210]	Asp Ser Tyr Gln Thr His Tyr Asp Val Ile Asn Arg Cys Lys Lys Asn
[4211]	595 600 605
[4212]	Leu Leu Arg Asn Ile Cys Arg Arg Met Ile Asp Met Ala Val Gln Asn
[4213]	610 615 620
[4214]	Lys Cys Asp Ile Ile Val Val Glu Asp Leu Ser Phe Gln Leu Ser Ser
[4215]	625 630 635 640
[4216]	His Asn Ser Arg Arg Asp Asn Val Phe Asn Ala Leu Trp Ser Cys Lys
[4217]	645 650 655
[4218]	Ser Ile Lys Asn Met Leu Gly Ile Met Ala Glu Gln His Asn Ile Ile
[4219]	660 665 670
[4220]	Ile Ser Glu Val Asp Pro Asn His Thr Ser Lys Ile Asp Cys Glu Thr
[4221]	675 680 685
[4222]	Gly Asn Phe Gly Tyr Arg Tyr Ser Ser Asp Phe Tyr Ser Val Ile Asp
[4223]	690 695 700
[4224]	Gly Gln Leu Val Arg Arg His Ala Asp Glu Asn Ala Ala Ile Asn Ile
[4225]	705 710 715 720
[4226]	Gly Asn Arg Trp Ala Ser Arg His Thr Asp Leu Lys Ser Phe Asn Cys
[4227]	725 730 735
[4228]	Arg Gln Ile Ser Ile Asp Gly Arg Lys Val Ala Phe Pro Tyr Ala Lys
[4229]	740 745 750
[4230]	Gly Lys Arg Lys Ser Ala Leu Phe Gly Tyr Leu Phe Gly Asn Cys Lys
[4231]	755 760 765
[4232]	Thr Val Phe Val Ser Asp Asp Gly Asp Ser Tyr Thr Pro Ile Pro Tyr
[4233]	770 775 780
[4234]	Ser Lys Phe Arg Lys Ser Ile Ser Lys Asp Asp His Asp Val Val Asn
[4235]	785 790 795 800
[4236]	Tyr Leu His Asp Leu Thr Met Asn Lys Asn Val Ile Arg Val Glu Tyr
[4237]	805 810 815
[4238]	Asn Lys Ser Ile Lys Ser Ala Ser Val Glu Leu Tyr Leu Asn Asp Asp
[4239]	820 825 830
[4240]	Arg Val Ile Ser Arg Ser Leu Arg Asp Lys Glu Val Asp Ala Ile Glu
[4241]	835 840 845

[4242]	Lys Leu Val Ser Arg Gly Ser Leu Ile Asn Glu Ser Gly Pro Ser Leu
[4243]	850 855 860
[4244]	Glu His Asp Glu Val Lys Ser Val Thr His Ser Arg Ala Asp Pro Lys
[4245]	865 870 875 880
[4246]	Lys Lys Arg Lys Val
[4247]	885
[4248]	<210> 84
[4249]	<211> 881
[4250]	<212> PRT
[4251]	<213> 人工序列
[4252]	<220>
[4253]	<223> Cas12j.5-NLS融合蛋白的氨基酸序列
[4254]	<400> 84
[4255]	Met Lys Val His Glu Ile Pro Arg Ser Gln Leu Leu Lys Ile Lys Gln
[4256]	1 5 10 15
[4257]	Tyr Glu Gly Ser Phe Val Glu Trp Tyr Arg Asp Leu Gln Glu Asp Arg
[4258]	20 25 30
[4259]	Lys Lys Phe Ala Ser Leu Leu Phe Arg Trp Ala Ala Phe Gly Tyr Ala
[4260]	35 40 45
[4261]	Ala Arg Glu Asp Asp Gly Ala Thr Tyr Ile Ser Pro Ser Gln Ala Leu
[4262]	50 55 60
[4263]	Leu Glu Arg Arg Leu Leu Leu Gly Asp Ala Glu Asp Val Ala Ile Lys
[4264]	65 70 75 80
[4265]	Phe Leu Asp Val Leu Phe Lys Gly Gly Ala Pro Ser Ser Ser Cys Tyr
[4266]	85 90 95
[4267]	Ser Leu Phe Tyr Glu Asp Phe Ala Leu Arg Asp Lys Ala Lys Tyr Ser
[4268]	100 105 110
[4269]	Gly Ala Lys Arg Glu Phe Ile Glu Gly Leu Ala Thr Met Pro Leu Asp
[4270]	115 120 125
[4271]	Lys Ile Ile Glu Arg Ile Arg Gln Asp Glu Gln Leu Ser Lys Ile Pro
[4272]	130 135 140
[4273]	Ala Glu Glu Trp Leu Ile Leu Gly Ala Glu Tyr Ser Pro Glu Glu Ile
[4274]	145 150 155 160
[4275]	Trp Glu Gln Val Ala Pro Arg Ile Val Asn Val Asp Arg Ser Leu Gly
[4276]	165 170 175
[4277]	Lys Gln Leu Arg Glu Arg Leu Gly Ile Lys Cys Arg Arg Pro His Asp
[4278]	180 185 190
[4279]	Ala Gly Tyr Cys Lys Ile Leu Met Glu Val Val Ala Arg Gln Leu Arg
[4280]	195 200 205
[4281]	Ser His Asn Glu Thr Tyr His Glu Tyr Leu Asn Gln Thr His Glu Met
[4282]	210 215 220
[4283]	Lys Thr Lys Val Ala Asn Asn Leu Thr Asn Glu Phe Asp Leu Val Cys

[4284]	225	230	235	240
[4285]	Glu Phe Ala Glu Val Leu Glu Glu Lys Asn Tyr Gly Leu Gly Trp Tyr			
[4286]		245	250	255
[4287]	Val Leu Trp Gln Gly Val Lys Gln Ala Leu Lys Glu Gln Lys Lys Pro			
[4288]		260	265	270
[4289]	Thr Lys Ile Gln Ile Ala Val Asp Gln Leu Arg Gln Pro Lys Phe Ala			
[4290]		275	280	285
[4291]	Gly Leu Leu Thr Ala Lys Trp Arg Ala Leu Lys Gly Ala Tyr Asp Thr			
[4292]		290	295	300
[4293]	Trp Lys Leu Lys Lys Arg Leu Glu Lys Arg Lys Ala Phe Pro Tyr Met			
[4294]	305	310	315	320
[4295]	Pro Asn Trp Asp Asn Asp Tyr Gln Ile Pro Val Gly Leu Thr Gly Leu			
[4296]		325	330	335
[4297]	Gly Val Phe Thr Leu Glu Val Lys Arg Thr Glu Val Val Val Asp Leu			
[4298]		340	345	350
[4299]	Lys Glu His Gly Lys Leu Phe Cys Ser His Ser His Tyr Phe Gly Asp			
[4300]		355	360	365
[4301]	Leu Thr Ala Glu Lys His Pro Ser Arg Tyr His Leu Lys Phe Arg His			
[4302]		370	375	380
[4303]	Lys Leu Lys Leu Arg Lys Arg Asp Ser Arg Val Glu Pro Thr Ile Gly			
[4304]	385	390	395	400
[4305]	Pro Trp Ile Glu Ala Ala Leu Arg Glu Ile Thr Ile Gln Lys Lys Pro			
[4306]		405	410	415
[4307]	Asn Gly Val Phe Tyr Leu Gly Leu Pro Tyr Ala Leu Ser His Gly Ile			
[4308]		420	425	430
[4309]	Asp Asn Phe Gln Ile Ala Lys Arg Phe Phe Ser Ala Ala Lys Pro Asp			
[4310]		435	440	445
[4311]	Lys Glu Val Ile Asn Gly Leu Pro Ser Glu Met Val Val Gly Ala Ala			
[4312]		450	455	460
[4313]	Asp Leu Asn Leu Ser Asn Ile Val Ala Pro Val Lys Ala Arg Ile Gly			
[4314]	465	470	475	480
[4315]	Lys Gly Leu Glu Gly Pro Leu His Ala Leu Asp Tyr Gly Tyr Gly Glu			
[4316]		485	490	495
[4317]	Leu Ile Asp Gly Pro Lys Ile Leu Thr Pro Asp Gly Pro Arg Cys Gly			
[4318]		500	505	510
[4319]	Glu Leu Ile Ser Leu Lys Arg Asp Ile Val Glu Ile Lys Ser Ala Ile			
[4320]		515	520	525
[4321]	Lys Glu Phe Lys Ala Cys Gln Arg Glu Gly Leu Thr Met Ser Glu Glu			
[4322]		530	535	540
[4323]	Thr Thr Thr Trp Leu Ser Glu Val Glu Ser Pro Ser Asp Ser Pro Arg			
[4324]	545	550	555	560
[4325]	Cys Met Ile Gln Ser Arg Ile Ala Asp Thr Ser Arg Arg Leu Asn Ser			

[4326]		565		570		575
[4327]	Phe Lys Tyr Gln Met Asn Lys Glu Gly Tyr Gln Asp Leu Ala Glu Ala					
[4328]		580		585		590
[4329]	Leu Arg Leu Leu Asp Ala Met Asp Ser Tyr Asn Ser Leu Leu Glu Ser					
[4330]		595		600		605
[4331]	Tyr Gln Arg Met His Leu Ser Pro Gly Glu Gln Ser Pro Lys Glu Ala					
[4332]		610		615		620
[4333]	Lys Phe Asp Thr Lys Arg Ala Ser Phe Arg Asp Leu Leu Arg Arg Arg					
[4334]		625		630		635
[4335]	Val Ala His Thr Ile Val Glu Tyr Phe Asp Asp Cys Asp Ile Val Phe					
[4336]		645		650		655
[4337]	Phe Glu Asp Leu Asp Gly Pro Ser Asp Ser Asp Ser Arg Asn Asn Ala					
[4338]		660		665		670
[4339]	Leu Val Lys Leu Leu Ser Pro Arg Thr Leu Leu Leu Tyr Ile Arg Gln					
[4340]		675		680		685
[4341]	Ala Leu Glu Lys Arg Gly Ile Gly Met Val Glu Val Ala Lys Asp Gly					
[4342]		690		695		700
[4343]	Thr Ser Gln Asn Asn Pro Ile Ser Gly His Val Gly Trp Arg Asn Lys					
[4344]		705		710		715
[4345]	Gln Asn Lys Ser Glu Ile Tyr Phe Tyr Glu Asp Lys Glu Leu Leu Val					
[4346]		725		730		735
[4347]	Met Asp Ala Asp Glu Val Gly Ala Met Asn Ile Leu Cys Arg Gly Leu					
[4348]		740		745		750
[4349]	Asn His Ser Val Cys Pro Tyr Ser Phe Val Thr Lys Ala Pro Glu Lys					
[4350]		755		760		765
[4351]	Lys Asn Asp Glu Lys Lys Glu Gly Asp Tyr Gly Lys Arg Val Lys Arg					
[4352]		770		775		780
[4353]	Phe Leu Lys Asp Arg Tyr Gly Ser Ser Asn Val Arg Phe Leu Val Ala					
[4354]		785		790		795
[4355]	Ser Met Gly Phe Val Thr Val Thr Thr Lys Arg Pro Lys Asp Ala Leu					
[4356]		805		810		815
[4357]	Val Gly Lys Arg Leu Tyr Tyr His Gly Gly Glu Leu Val Thr His Asp					
[4358]		820		825		830
[4359]	Leu His Asn Arg Met Lys Asp Glu Ile Lys Tyr Leu Val Glu Lys Glu					
[4360]		835		840		845
[4361]	Val Leu Ala Arg Arg Val Ser Leu Ser Asp Ser Thr Ile Lys Ser Tyr					
[4362]		850		855		860
[4363]	Lys Ser Phe Ala His Val Ser Arg Ala Asp Pro Lys Lys Lys Arg Lys					
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[4365]	Val					
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 [4378] 35 40 45
 [4379] Asn Leu Leu Lys Leu Met Leu Ile Leu Arg Gly Glu Lys Tyr Thr Leu
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 [4382] 65 70 75 80
 [4383] Leu Cys Lys Glu His Asn Ile Lys Gly Ser Ile Cys Ser Leu Lys Glu
 [4384] 85 90 95
 [4385] Lys Ser Arg Lys Leu Tyr Glu Val Phe Ser Cys Tyr Ile Asp Lys Lys
 [4386] 100 105 110
 [4387] Gly Asn Leu Lys Thr Asn Ser Lys Ala Arg Ser Phe Ala Gly Val Leu
 [4388] 115 120 125
 [4389] Leu Asn Pro Lys Asp Val Lys Leu Pro Pro Gln Ile Asp Ser Ile Ser
 [4390] 130 135 140
 [4391] Ser Phe Val Val Glu Leu Arg Ala Lys Gly Val Leu Pro Ile Lys His
 [4392] 145 150 155 160
 [4393] Glu Gly Asn Tyr Leu Ser Gly His Pro Ser Leu Lys Tyr Ser Val Ala
 [4394] 165 170 175
 [4395] Gln Asn Val Leu Val Lys Leu Thr Ser Met Glu Lys Leu Gln Lys Ile
 [4396] 180 185 190
 [4397] Tyr Ser Asp Glu Lys Ala Gly Trp Glu Asn Ile Val Ser Glu Val Arg
 [4398] 195 200 205
 [4399] Ser Asp Leu Pro Lys Ile Glu Arg Tyr Glu Arg Met Leu Leu Ser Ile
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 [4401] Lys Ala Val Lys Glu Met Glu Lys Phe Gly Ile Asn Asn Tyr Arg His
 [4402] 225 230 235 240
 [4403] Leu Leu Asn Asn Trp Arg Asp Glu Val Asp Lys Asp Ser Gly Lys Val
 [4404] 245 250 255
 [4405] Leu Lys Gln Gly Met Arg Thr Tyr Phe Val Asn Met Leu Glu Ser Lys
 [4406] 260 265 270
 [4407] Lys Asp Tyr Arg Phe Glu Glu Ser Asp Arg Tyr Leu Phe Gly Tyr Ala
 [4408] 275 280 285
 [4409] Pro Glu Val Met Asn Leu Val Tyr His Asp Phe Arg Asp Leu Trp Gln

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[4411]	Gly Glu Asp Ile Ile Gly Ser Gln Ser Pro Glu Lys Lys Asp Arg Asp		
[4412]	305	310	315 320
[4413]	Tyr Val Asp Val Ile Phe Asn Tyr Phe Asn Trp Arg Lys Glu Ser Ile		
[4414]		325 330	335
[4415]	Asn Ile Ser Ser Phe Asp Ser Tyr Gly Lys Thr Ala Gln Ile Lys Leu		
[4416]		340 345	350
[4417]	Gly Asp Asn Tyr Val Pro Phe Ser Asn Phe Gln Tyr Asp Lys Ile Leu		
[4418]		355 360	365
[4419]	Asp Ala Trp Thr Leu Glu Ile Ala Asn Val Ser Gly Glu Gly Asp Asn		
[4420]	370	375	380
[4421]	His Lys Leu Val Ile Ala Arg Ser Pro Gln Phe Asp Ser His Ser Ser		
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[4423]	Val Lys Asp Ile Val Met Lys Asn Leu Lys Gly Lys Glu Ala Ser Lys		
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[4425]	Thr Thr Leu Glu Phe Arg Tyr Ser Gly Asp Ser Lys Lys Ser Thr Trp		
[4426]		420 425	430
[4427]	Tyr Arg Gly Thr Leu Lys Glu Pro Thr Leu Arg Tyr Ser Ser Ser Lys		
[4428]		435 440	445
[4429]	Asn Cys Leu Tyr Val Asp Phe Ala Leu Ser Asn His Ile Val Glu Gly		
[4430]	450	455	460
[4431]	Leu Ile Ser Asp Asn Leu Gly Ile Ser Asp Lys Met Tyr Lys Phe Arg		
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[4433]	Gly Glu Phe Met Lys Ala Ser Pro Ser Ser Gly Lys Gln Ser Asn Ser		
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[4435]	Ile Asn Leu Pro Ile Lys Lys Leu Arg Ala Met Gly Val Asp Phe Asn		
[4436]		500 505	510
[4437]	Leu Arg Arg Pro Phe Gln Ala Ser Ile Tyr Asp Val Glu Asn Lys Asn		
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[4439]	Gly Asn Leu Glu Phe Ser Phe Val Lys His Val Gln Ser Phe Ser Asn		
[4440]	530	535	540
[4441]	Glu Asn Asp Glu Glu Arg Ala Lys Glu Leu Leu Asn Ile Glu Arg Asn		
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[4443]	Ile Leu Ala Leu Lys Ile Leu Ile Trp Gln Thr Val Gly Tyr Val Thr		
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[4445]	Gly Lys Asn Asp Thr Ile Asp Gly Val Val Thr Arg Lys Asn Asn Ala		
[4446]		580 585	590
[4447]	Val Asp Ile Glu Lys Thr Leu Gly Ile Asn Met Lys Glu Tyr Met Ala		
[4448]		595 600	605
[4449]	Tyr Leu Asn Gln Phe Arg Ser Tyr Glu Asp Lys Asn Lys Ala Phe Met		
[4450]	610	615	620
[4451]	Asp Leu Arg Lys Arg Glu Tyr Ala Trp Ile Val Pro Pro Leu Ile Phe		

[4452]	625	630	635	640
[4453]	Gln Cys Arg Ser Arg Leu Ile Ser Phe Arg Ser Glu Tyr Phe Asn Thr			
[4454]		645	650	655
[4455]	Pro Lys Asp Glu Lys Ser His Tyr Cys Gln His Arg Asn Phe Val Asp			
[4456]		660	665	670
[4457]	Tyr Ser Thr Phe Leu Lys Lys Asn Val Val Lys Lys Met Met Glu Leu			
[4458]		675	680	685
[4459]	Arg Arg Ser Tyr Ser Thr Phe Gly Met Ser Ser Glu Gln Ser Ile Trp			
[4460]		690	695	700
[4461]	Val Thr Asn Asn Asp His Ala Lys Asp Gly Ser Lys Lys Asn Gly Asn			
[4462]	705	710	715	720
[4463]	Met Phe Asp Asp Asp Leu His Gln Trp Tyr Asn Gly Leu Val Arg Lys			
[4464]		725	730	735
[4465]	Cys Ser Ser Leu Ala Ser Ser Ile Ile Asn Val Ala Arg Asp Asn Gly			
[4466]		740	745	750
[4467]	Ala Ile Leu Val Phe Ile Glu Asp Leu Asp Cys His Pro Ser Ala Phe			
[4468]		755	760	765
[4469]	Asp Ser Glu Glu Asp Asn Ser Leu Lys Ser Ile Trp Gly Trp Gly Ser			
[4470]		770	775	780
[4471]	Ile Lys Ala Ser Leu Ala His Gln Ala Arg Lys His Asn Ile Ala Val			
[4472]	785	790	795	800
[4473]	Val Ala Asn Asp Pro His Leu Thr Ser Leu Val Ser Ser Thr Thr Gly			
[4474]		805	810	815
[4475]	Glu Leu Gly Ile Ala Lys Gly Arg Asp Val Leu Phe Phe Asp Ser Lys			
[4476]		820	825	830
[4477]	Gly Lys Leu Thr Ser Lys Val Asn Arg Asp Glu Asn Ala Ala Gln Asn			
[4478]		835	840	845
[4479]	Ile Ala Ile Arg Gly Phe Val Arg His Ser Asp Leu Arg Glu Phe Val			
[4480]		850	855	860
[4481]	Ala Glu Lys Ile Glu Glu Asn Arg Tyr Arg Val Val Val Asn Lys Thr			
[4482]	865	870	875	880
[4483]	His Lys Arg Lys Ala Gly Ala Ile Tyr Arg His Ile Gly Ser Thr Glu			
[4484]		885	890	895
[4485]	Cys Ile Met Ser Lys Gln Ala Asp Gly Ser Leu Lys Ile Asp Lys Thr			
[4486]		900	905	910
[4487]	Glu Leu Thr Pro Leu Glu Ile Lys Met Glu Lys Lys Asn Asp Lys Lys			
[4488]		915	920	925
[4489]	Met Tyr Val Ile Leu His Gly Lys Thr Trp Arg Leu Arg His Glu Leu			
[4490]		930	935	940
[4491]	Asn Glu Lys Leu Glu Lys Asp Leu Asp Asn His Leu Lys Ser Lys Ser			
[4492]	945	950	955	960
[4493]	Ser Val Ile Ser Ser Arg Ala Asp Pro Lys Lys Lys Arg Lys Val			

[4494]		965	970	975
[4495]	<210>	86		
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[4504]	Thr Leu Arg Gly Lys Val Tyr Asn His Asp Thr Ala Met Glu Ala Phe			
[4505]		20	25	30
[4506]	Ala Pro Val Met Lys Gly Met Val Pro Tyr Ala Asn Asn Leu Met Arg			
[4507]		35	40	45
[4508]	Ile Leu Leu Thr Leu Arg Leu Glu Lys Tyr Thr Leu Asp Gly Ile His			
[4509]		50	55	60
[4510]	His Thr Lys Glu Glu Val Glu Lys Asp Leu Arg Gly Leu Met Lys Glu			
[4511]		65	70	75
[4512]	Tyr Gly Ile Asn Leu Ser Phe Ala Lys Phe Ser Glu Met Ala Gly Glu			
[4513]		85	90	95
[4514]	Val Tyr Arg Val Phe Val Cys Tyr Val Asp Ala Lys Gly Lys Leu Lys			
[4515]		100	105	110
[4516]	Val Asn Gly Lys Ala Arg Gly Phe Ala Asn Val Phe Phe Ser Glu Asp			
[4517]		115	120	125
[4518]	Asp Ala Thr Ile Pro Glu Asn Cys Pro Ser Met Glu Leu Leu Arg Lys			
[4519]		130	135	140
[4520]	Lys Gly Met Phe Pro Ile Leu Val Asp Gly Lys Pro Ile Ser Ser Ile			
[4521]		145	150	155
[4522]	Ser Arg Glu Lys Thr Pro Leu Lys Tyr Ser Val Ala Gln Asp Val Leu			
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[4524]	Thr Lys Leu Thr Ser Met Glu Glu Ile Ser Lys Glu Tyr Glu Lys Ala			
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[4526]	Lys Thr Asp Trp Glu Asn Glu Cys Gln Lys Val Ile Ser Gln Leu Pro			
[4527]		195	200	205
[4528]	Leu Ile Gly Arg Tyr Glu Ala Leu Leu Thr Thr Ile Pro Leu Ile Pro			
[4529]		210	215	220
[4530]	Glu Met Arg Gly Phe Asp Gly Asp Asn Tyr Arg Lys Met Leu Asn Arg			
[4531]		225	230	235
[4532]	Trp Arg Asp Tyr Val Asn Glu Asp Gly Glu Leu Val Arg Gly Gly Met			
[4533]		245	250	255
[4534]	Lys Thr Tyr Phe Leu Asp Leu Leu Ser Lys Asp Thr Ser His Lys Phe			
[4535]		260	265	270

[4536]	Asn Glu Glu Glu Arg Tyr Leu Phe Gly Tyr Cys Pro Glu Phe Met Asn
[4537]	275 280 285
[4538]	Leu Ile Tyr His Asp Phe Arg Asp Leu Trp Ser Lys Glu Asp Ile Ile
[4539]	290 295 300
[4540]	Gly Ser Gln Arg Lys Gly Lys Gly Leu Lys Gly Lys Asp Tyr Val Asp
[4541]	305 310 315 320
[4542]	Val Ile Phe Asn Cys Phe His Trp Arg Arg Glu Ser Ile Asn Ile Ser
[4543]	325 330 335
[4544]	Ser Phe Gly Asn Asn Asp Lys Val Met Asn Ile His Leu Gly Asp Asn
[4545]	340 345 350
[4546]	Phe Val Pro Phe Glu Leu Lys Ser Gln Asn Gly Ile Trp Glu Val His
[4547]	355 360 365
[4548]	Val Gln Asn Leu His Gly Gln Asn Asp Pro His Arg Val Ile Val Cys
[4549]	370 375 380
[4550]	Arg Cys Pro Gln Phe Asn Glu Asp Ser Ser Met Lys Met Val His Pro
[4551]	385 390 395 400
[4552]	Leu Ala Lys Asn Gly Glu Glu Ser Asp Lys Glu Asn Ile Glu Phe Arg
[4553]	405 410 415
[4554]	Tyr Ser Gly Asp Ser Lys Arg Glu Thr Trp Tyr Thr Gly Leu Leu Lys
[4555]	420 425 430
[4556]	Glu Pro Thr Leu Arg Tyr Asp Val Glu Arg Lys Ser Leu Tyr Val Asp
[4557]	435 440 445
[4558]	Phe Ile Leu Ser Asn His Arg Val Glu Gly Val Val Thr Asn Glu Tyr
[4559]	450 455 460
[4560]	Leu Lys Asp Pro Arg Asp Leu Phe Gly Val Arg Gly Tyr Phe Leu Ser
[4561]	465 470 475 480
[4562]	Ser Ser Val Ser Asn Pro Arg Gln Lys Asp Lys Thr Ser Leu Pro Asp
[4563]	485 490 495
[4564]	Gly Lys Phe Asn Val Met Gly Val Asp Leu Gly Leu Lys Cys Pro Tyr
[4565]	500 505 510
[4566]	Glu Cys Ala Ile Tyr Gly Ile Thr Val Lys Asn Gly Lys Met Gln His
[4567]	515 520 525
[4568]	Lys Trp Ser His Asn Val Ser Ala Glu Asp Asn Asn Asn Val Ser Glu
[4569]	530 535 540
[4570]	Arg Leu Ala Asn Leu Lys Lys Ile Asp Glu Lys Ile Leu Ala Thr Gln
[4571]	545 550 555 560
[4572]	Val Leu Ile Ser Leu Thr Lys Met Cys Val Val Lys Asp Glu Glu Ile
[4573]	565 570 575
[4574]	Pro Asp Ser Tyr Thr Leu Arg Glu His Arg Val Asp Ile Ala Lys Ser
[4575]	580 585 590
[4576]	Leu Asp Leu Asp Met Asp Lys Tyr Arg Arg Tyr Val Glu Lys Cys Lys
[4577]	595 600 605

[4578]	Lys Asn Pro Asp Lys Ile Gln Ala Leu Lys Asp Ile Arg Lys Ser Glu
[4579]	610 615 620
[4580]	Asn Asn Trp Ile Val Ala Glu Lys Ile Asn Glu Ile Arg Ser Leu Ile
[4581]	625 630 635 640
[4582]	Ser Glu Ile Arg Ser Glu Tyr Tyr Ala Ser Lys Asp Lys Arg Asn Tyr
[4583]	645 650 655
[4584]	Cys Arg Asn Leu Asn Gly Val Asp Leu Ser Val Phe Leu Lys Lys Lys
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[4586]	Val Val Lys Asn Trp Ile Ser Leu Leu Arg Ser Phe Ser Thr Phe Gly
[4587]	675 680 685
[4588]	Met Thr Pro Gln Glu Ser Ala Tyr Ile Arg Lys Asp Phe Ala Lys Asn
[4589]	690 695 700
[4590]	Leu Ser Lys Trp Tyr Lys Gly Leu Val Arg Lys Cys Gly Ser Ile Ala
[4591]	705 710 715 720
[4592]	Ala His Ile Val Asn Ile Ala Arg Asp Asn Lys Val Met Val Ile Phe
[4593]	725 730 735
[4594]	Ile Glu Asp Leu Asp Ala Arg Thr Ser Ala Phe Asp Ser Lys Glu Asp
[4595]	740 745 750
[4596]	Asn Glu Leu Lys Ile Leu Trp Gly Trp Gly Glu Ile Lys Lys Trp Ile
[4597]	755 760 765
[4598]	Gly His Gln Ala Arg Lys His Asn Ile Ala Val Val Ala Val Asp Pro
[4599]	770 775 780
[4600]	His Leu Thr Ser Leu Val Asn His Glu Ser Gly Leu Leu Gly Ile Ala
[4601]	785 790 795 800
[4602]	Gly Ser Gly Asn Asp Arg Asn Ile Tyr Thr Phe Gln Lys Asn Lys Lys
[4603]	805 810 815
[4604]	Tyr Val Val Ile Asn Arg Asp Asn Asn Ala Ala His Asn Ile Ala Leu
[4605]	820 825 830
[4606]	Arg Gly Leu Ser Lys His Thr Asp Ile Arg Glu Phe Tyr Val Glu Gln
[4607]	835 840 845
[4608]	Ile Asp Val Asp His Tyr Arg Leu Met Tyr Gly Pro Glu Ala Glu Asn
[4609]	850 855 860
[4610]	Gly Lys Arg Arg Ser Gly Ala Ile Tyr Lys His Ile Gly Ser Thr Glu
[4611]	865 870 875 880
[4612]	Cys Val Phe Ser Lys Gln Lys Asn Gly Thr Leu Lys Val Glu Lys Thr
[4613]	885 890 895
[4614]	Ser Leu Thr Lys Asp Glu Lys Glu Met Pro Lys Ile Asn Gly Lys Gly
[4615]	900 905 910
[4616]	Val Tyr Ala Ile Leu His Gly Asn Glu Trp Arg Leu Arg His Glu Leu
[4617]	915 920 925
[4618]	Asn Glu Glu Leu Gly Ala Lys Leu Asp Gly Ile Ser Val Lys Arg Val
[4619]	930 935 940

[4620] Val Ser Glu Pro Asn Lys Val Lys Thr Ser Leu Val Lys Gly Ser Val
 [4621] 945 950 955 960
 [4622] Arg Ala Ser Arg Ala Asp Pro Lys Lys Lys Arg Lys Val
 [4623] 965 970
 [4624] <210> 87
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 [4633] Glu Asn Lys Met Tyr Pro Asp Lys Asp Thr Asp Phe Pro Val Asn Ser
 [4634] 20 25 30
 [4635] Gln Phe Ser Arg Ser Ile Ser Ile Arg Ala Asn Val Asp Pro Lys Asp
 [4636] 35 40 45
 [4637] Leu Leu Val Leu Lys Arg Thr Phe Glu Glu Thr Thr Lys Ile Ser Asp
 [4638] 50 55 60
 [4639] Glu Leu Leu Ser Thr Leu Leu Met Leu Arg Gly Lys Asp Tyr Cys Leu
 [4640] 65 70 75 80
 [4641] Asp Asn Val Val Cys Lys Gly Glu Glu Val Leu Glu Asn Leu Tyr Lys
 [4642] 85 90 95
 [4643] Lys Leu Ser Lys Asn Ala Thr Val Asn Arg Asp Lys Phe Ile Ser Thr
 [4644] 100 105 110
 [4645] Ala Lys Ala Phe Tyr Glu Tyr Phe His Gly Cys Ser Tyr His Lys Gly
 [4646] 115 120 125
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 [4653] His Ala Lys Ile Lys Gly Phe Glu Lys Ile Asp Lys Glu Tyr Gln Ala
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 [4656] 195 200 205
 [4657] Leu Asn Arg Tyr Gly Glu Met Leu Lys Gly Leu Ser Asp Leu Gly Thr
 [4658] 210 215 220
 [4659] Met Gly Asn Phe Asn Gly Lys Lys Tyr Asp Arg Phe Met Gly His Trp
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 [4661] Arg Asn Glu Gln Lys Ile Pro Asp His Ile Ser Met Leu Asp Phe Phe

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[4663]	Arg Lys Ile Tyr Gln Glu Lys Gly Lys Ser His Arg Phe Thr Ala Ile					
[4664]		260		265		270
[4665]	Asp Asn Phe Thr Tyr Gly Tyr Glu Ser Glu Phe Met Asn His Ile Tyr					
[4666]		275		280		285
[4667]	Leu Asn Phe Ser Asp Leu Trp Leu Lys Glu Asp Val Ile Gly Asp Glu					
[4668]		290		295		300
[4669]	Glu Tyr Val Ser Leu Ile Arg Gly Ala Tyr His Trp Gln Lys Asp Val					
[4670]		305		310		315
[4671]	Val Gly Ile Ala Ser Phe Ser Gly Tyr Asn Lys Tyr Glu Lys Leu Phe					
[4672]		325		330		335
[4673]	Met Gly Asp Asn Lys Ile Asn Tyr Ala Leu Asp Phe Ser Asn Lys Asp					
[4674]		340		345		350
[4675]	Gln Trp Leu Met Lys Phe Asn Asn Val Ile Ser Lys Glu Pro Glu Thr					
[4676]		355		360		365
[4677]	Ile Thr Leu Arg Leu Cys Lys Asn Gly Tyr Phe Asn Asn Leu Ser Val					
[4678]		370		375		380
[4679]	Leu Glu Lys Asn Asp Glu Asn Gly Arg Tyr Lys Ile Arg Phe Ser Thr					
[4680]		385		390		395
[4681]	Glu Lys Gln Gly Lys Tyr Phe Tyr Glu Ala Phe Ile Arg Glu Pro Phe					
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[4683]	Leu Arg Tyr Asn Lys Asp Asn Asp Lys Ile Tyr Val His Phe Cys Leu					
[4684]		420		425		430
[4685]	Ser Glu Glu Ile Lys Glu Asn Cys Pro Asn His Leu Asp Thr Arg Ser					
[4686]		435		440		445
[4687]	Asp Lys Tyr Leu Phe Lys Ser Ala Leu Leu Thr Asn Ser Arg Gln Lys					
[4688]		450		455		460
[4689]	Leu Gly Lys Leu His Tyr Arg Asp Phe His Ile Val Gly Val Asp Leu					
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[4691]	Gly Ile Asn Pro Val Ala Lys Ile Thr Val Cys Lys Val His Val Asp					
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[4693]	Lys Asn Glu Asn Leu Lys Ile Thr Lys Ile Ile Thr Glu Glu Thr Arg					
[4694]		500		505		510
[4695]	Lys Asn Ile Asp Thr Asn Tyr Leu Asp Gln Leu Asn Leu Leu Tyr Lys					
[4696]		515		520		525
[4697]	Lys Ile Val Ser Leu Lys Arg Leu Ile Arg Ala Thr Val Ala Phe Lys					
[4698]		530		535		540
[4699]	Lys Asp Gly Glu Glu Ile Pro Lys Met Phe Lys Met Gly Lys Lys Ser					
[4700]		545		550		555
[4701]	Pro Tyr Phe Leu Asn Trp Thr Glu Val Leu Asn Val Asn Tyr Asp Asp					
[4702]		565		570		575
[4703]	Tyr Ile Lys Glu Ile Ser Thr Phe Ser Val Asp Arg Leu Ser Gly Leu					

[4704]		580		585		590										
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[4710]	625			630		635		640								
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[4713]	Ile	Gly	Leu	Gln	Lys	Met	Phe	Ser	Gly	Gly	Gly	Lys	Asp	Val	Ala	Lys
[4714]				660		665		670								
[4715]	Lys	Asp	Tyr	Leu	Tyr	Leu	Arg	Gly	Leu	Arg	Lys	His	Ile	Gly	Asn	Tyr
[4716]				675		680		685								
[4717]	Thr	Ala	Ser	Ala	Ile	Val	Ser	Ile	Ala	Gln	Lys	Tyr	Asn	Ala	Val	Phe
[4718]				690		695		700								
[4719]	Ile	Phe	Ile	Glu	Asp	Leu	Asp	Leu	Lys	Ile	Ser	Gly	Met	Asn	Gly	Lys
[4720]	705			710		715		720								
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[4722]				725		730		735								
[4723]	Arg	Leu	Ser	Glu	Lys	Ala	Glu	Lys	Phe	Gly	Ile	Gly	Ile	Val	Pro	Val
[4724]				740		745		750								
[4725]	Asn	Pro	Glu	Leu	Thr	Ser	Gln	Met	Asp	Arg	Glu	Thr	Phe	Leu	Leu	Gly
[4726]				755		760		765								
[4727]	Tyr	Arg	Asn	Pro	Thr	Asn	Lys	Lys	Glu	Leu	Tyr	Val	Lys	Arg	Asp	Asp
[4728]				770		775		780								
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[4730]	785			790		795		800								
[4731]	Leu	Arg	Gly	Leu	Gly	His	His	Ala	Asn	Leu	Ile	Gln	Phe	Arg	Ala	Asp
[4732]				805		810		815								
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[4734]				820		825		830								
[4735]	Gln	Gly	Ala	Leu	Tyr	Gly	Tyr	Leu	Asn	Ser	Thr	Ala	Val	Leu	Phe	Lys
[4736]				835		840		845								
[4737]	Asp	Lys	Gly	Asp	Gly	Val	Leu	Thr	Ile	His	Lys	Ser	Lys	Leu	Thr	Lys
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 [4762] Asp Lys Glu Phe Asp Lys Ile Ile Ser Lys Leu Gly Leu Ser Lys Lys
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 [4766] Val Lys Leu Gln Asn Gly Arg Ser Leu Ala Asn Ile Leu Phe Glu Thr
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 [4768] Lys Gly Thr Thr Leu Ile Gly Cys Gly Lys Asp Lys Lys Gly Glu Lys
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 [4774] Asp Tyr Lys Tyr Leu Ile Cys Tyr Leu Val His Asn Val Leu Ser Ser
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 [4778] Ser Lys Leu Ser Asn Ser Asn Leu Pro Gln Leu Glu Arg Met Ser Glu
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 [4780] Phe Leu Asn Gly Ile Asn His Leu Gly Asn Ile Ile Gly Trp Asn Gly
 [4781] 210 215 220
 [4782] Lys Lys Tyr Ile Gly Phe Ile Lys Lys Trp Thr Asp Glu Glu Ser Ser
 [4783] 225 230 235 240
 [4784] Met Tyr Asp Phe Phe Val Gln Lys Leu Gln Asp Asn Pro Lys Tyr Lys
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 [4786] Phe Gly Lys Lys Asp Gln Phe Leu Tyr Gly Tyr Glu Pro Glu Phe Leu
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[4788]	Asn Tyr Leu Phe His Asp Phe Arg Asp Leu Trp His Pro Asp Asn Leu
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[4790]	Ile Gly Lys Asp Glu Tyr Val Asp Leu Ile Ser Gly Lys Asn Asn Thr
[4791]	290 295 300
[4792]	Asp Ala Glu Thr Ala Asn Lys Gly Ala Tyr His Trp Leu Lys Asp Phe
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[4794]	Ile Asn Ile Ser Ser Phe Asp Ala Tyr Gly Lys Met Ala Thr Ile Gly
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[4812]	Phe Ile Gly Lys Asn Phe Val Gly Leu Ala Ile Asp Arg Gly Ile Asn
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[4820]	Val Gly Ile Lys Ser Leu Val Trp Asn Thr Val Lys Tyr Arg Thr Gly
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[4824]	Asp Leu Phe Glu Met Phe Asp Ile Asp Tyr Asn Asn Tyr Leu Lys Glu
[4825]	565 570 575
[4826]	Val Asn Asn Leu Pro Tyr Asp Pro Asn Ser Glu Arg Ser Ile Ile Gln
[4827]	580 585 590
[4828]	Thr Trp Val Ser Ser Pro Trp Lys Val Lys Asp Leu Val Lys Asp Ala
[4829]	595 600 605

[4830]	Lys Asn Arg Met Val Gln Ile Lys Thr Gln Tyr His Asn Ala Lys Asp
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[4832]	Lys Glu Lys Tyr Ile Thr Thr Gln Asn Arg Ala Gly Phe Tyr Asp Phe
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[4836]	Ser Gly Gly Gln Lys Asp Ile Cys Lys Asn Asn Glu Glu Tyr Arg Arg
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[4839]	675 680 685
[4840]	Leu Ala Arg Lys Phe Asn Val Asp Cys Ile Phe Leu Glu Asp Leu Asp
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[4853]	785 790 795 800
[4854]	Ile Asp Ile Arg Glu Phe Phe Ala Glu Lys Ile Glu Val Ser Gly Lys
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[4856]	Thr Leu Tyr Arg Ile Ser Asn Lys Leu Gly Lys Gln Arg Met Gly Ser
[4857]	820 825 830
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[4862]	Leu Ala Pro Arg Ile Ala Glu Lys Lys Ser Thr Tyr Leu Ile Met Asn
[4863]	865 870 875 880
[4864]	Gly Ser Lys Trp Met Phe Arg His Glu Ala Lys Lys Ile Val Glu Thr
[4865]	885 890 895
[4866]	Tyr Lys Asp Arg Tyr Cys Ala Asn His Lys Val Ala Ser Lys Asp Gly
[4867]	900 905 910
[4868]	Ser Arg Ala Asp Pro Lys Lys Lys Arg Lys Val
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 [4881] Arg Asn Gly Phe Ser Asp Ile Gly Val Asp Pro Ser Leu Val Ser Tyr
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[4919]	Ser Lys Asn Cys Trp Glu Ala Glu Arg Val Lys Leu Tyr Glu Gln Asp		
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[4923]	Pro Ile Glu Lys Met Lys Leu Arg Thr Phe Lys Thr Ile Val Lys Lys		
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[4925]	Leu Asp Lys Glu Phe Gly Lys Arg Gly Asp Lys Thr Pro Ser Ile His		
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[4931]	Asn Lys Lys Gln Phe Tyr Lys Leu Cys Lys Asp His Asp Gly Lys Arg		
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[4933]	Thr Phe Ala Lys Val Val Glu Glu Ser Tyr His Trp Gly Lys Asn Ser		
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[4935]	Ile Asn Val Ser Thr Phe Gln Asn Ser Thr Ser Ile Leu Leu Gly Gly		
[4936]	465	470	475 480
[4937]	Asn Tyr Leu Asn Tyr Ser Met Ser Ile Glu Gly Glu Gly Leu Val Ile		
[4938]		485	490 495
[4939]	Lys Phe Asp Asn Pro Leu Ser Gly Lys Glu Val His Phe Val Val Cys		
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[4941]	Asn Asn Lys Tyr Leu Ser Asp Leu Glu Ile Leu Ser Gly Asn Pro Asn		
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[4943]	Arg Lys Asp Asn Asn Tyr Thr Ile Ser Tyr Ser Thr Gly Gly Lys Ala		
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[4945]	Arg Phe Ile Ala Lys Ser Lys Glu Pro Arg Ile Phe Phe Asn Arg Lys		
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[4947]	Thr Lys Lys Trp Glu Ile Ala Phe Gln Leu Ser Asp Val Ser Pro Leu		
[4948]		565	570 575
[4949]	Asn Gly Lys Phe Gly Lys Gln Gly Glu Phe Leu Ser Asn Leu Arg Lys		
[4950]		580	585 590
[4951]	Phe Val Tyr Asn His Val Ala Lys Ser Pro Ser Lys Leu Asn Ile Ser		
[4952]		595	600 605
[4953]	Asp Asn Asn Cys Arg Ala Val Ala Tyr Asp Leu Gly Ile Arg Asn Val		
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[4955]	Gly Ala Trp Ser Ser Phe Asp Phe Ser Tyr Lys Asp Gly Val Leu Gly		

[4956]	625	630	635	640
[4957]	Gly Tyr Lys Tyr Leu Thr Ser Gly Ser Leu Arg Ser Lys Ser Glu Ser			
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[4959]	Ser Glu Met Asp Gln Gly Tyr Tyr Phe Val Leu Asn Leu Lys Lys Ile			
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[4961]	Val Lys Leu Ile Pro Val Val Lys Lys Ser Ile Ile Asp Asp Pro Glu			
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[4963]	Leu Lys Arg Gln Phe Ile Gly Val Leu Asn Glu Asn Gly Asn Thr Val			
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[4965]	Gly Leu Gly Asn Ile Gly Lys Leu Asp Ile Ala Ser Arg Lys Ala Val			
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[4967]	Gln Ser Phe His Asn Cys Ile Gln Gln Ile Asn Tyr Tyr Val Asp Thr			
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[4969]	Tyr Ala Asp His Ile Asp Lys Ile Ser Ala Lys Asp Phe Val Asp Asp			
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[4971]	Ile Asp Gly Ile Lys Val Leu Asp Glu Asp Asp Pro Tyr Val Val Lys			
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[4973]	Ile Leu Ser His Leu Pro Glu Asp Val Glu Gly Asn Gln Asp Asp Ile			
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[4975]	Leu Asn Ile Ser Leu Leu Lys Trp Lys Thr Ser Asn Ala Gln Phe Val			
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[4978]		805	810	815
[4979]	Glu Asn Leu Asp Asn Ile Arg Gly Lys Lys Thr Gln Val Val Thr Gln			
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[4981]	Lys Thr Phe His Lys Ile Lys Phe Ala Lys Ala Leu Leu Ser Leu Met			
[4982]		835	840	845
[4983]	Lys Ser Trp Ser Ser Ile Gly Thr Val Arg Val Val Lys Thr Asp Gln			
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[4985]	Ile Tyr Gly Lys Lys Ile Trp Asp Tyr Ile Asn Gly Leu Arg Arg Asn			
[4986]	865	870	875	880
[4987]	Val Leu Thr Tyr Leu Ser Ser Ala Ile Val Asn Asn Ala Leu Asp Leu			
[4988]		885	890	895
[4989]	Gly Ala His Met Ile Ile Leu Glu Asp Leu Asp Ser Ser Val Ser Lys			
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[4991]	Tyr Arg Glu Lys Asp Lys Asn Ala Ile Gln Ser Leu Trp Gly Ser Gly			
[4992]		915	920	925
[4993]	Glu Leu Lys Lys Arg Ile Glu Glu Lys Ala Glu Lys His Arg Val Val			
[4994]		930	935	940
[4995]	Val Gln Tyr Val Ser Pro Tyr Leu Thr Ser Gln Leu Asp Asn Glu Thr			
[4996]	945	950	955	960
[4997]	Lys Asp Ile Gly Tyr Arg Lys Gly Gly Arg Leu Tyr Val Val Arg Asn			

[4998]		965		970		975
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[5001]	Gly Glu Arg Phe Phe Asp Arg Asp Leu Ile Gln Thr Leu Ser Gly Val					
[5002]		995		1000		1005
[5003]	Val Val Glu Asp Gln Ser Thr Val Tyr Ile Leu Gln Lys Arg Asn					
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[5005]	Val Ser Ser Asp Asn Arg Lys Arg Phe Tyr Lys Lys Phe Leu Glu					
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[5007]	Asp Val Gly Gly Lys Ser Lys Lys Asp Ala Val Leu Lys Met Gly					
[5008]		1040		1045		1050
[5009]	Asp His Gly Glu Leu Glu Val Glu Arg Leu Ile Asp Gly Lys Lys					
[5010]		1055		1060		1065
[5011]	Leu Asp Ile Asp Gly Lys Lys Ile Leu Val Asp Gly Glu Lys Val					
[5012]		1070		1075		1080
[5013]	Pro Phe Arg Asn Thr Ser Val Tyr Tyr Ser Pro Lys Lys Lys Lys					
[5014]		1085		1090		1095
[5015]	Trp Val Ser Lys Glu Leu Arg Cys Asn His Ile Lys Leu Thr Val					
[5016]		1100		1105		1110
[5017]	Glu Glu Gln Asp Ile Lys Ser Arg Ala Asp Pro Lys Lys Lys Arg					
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[5027]	<400> 90					
[5028]	Met Asn Asn Tyr Asp Asn Tyr Leu Ser Asp Tyr Leu Ala Met Leu Pro					
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[5030]	His Thr Lys Arg Thr Glu Ile Lys Lys Thr Ala Ser Lys Ile Ser Arg					
[5031]		20		25		30
[5032]	Lys Leu Asn Gln Lys Glu Val Lys Lys Gln Ile Glu Arg Ser Glu Tyr					
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[5034]	Ile Arg Ser Asn Cys Gly Tyr Ile Asn Ile Glu Arg Pro Gln Lys Ser					
[5035]		50		55		60
[5036]	Leu Ser Phe Leu Ser Tyr Ser Thr Ile Lys Ser Ala Cys Met Ser Val					
[5037]	65	70		75		80
[5038]	Asn Phe Arg Ala Phe Gln Asn Pro Ile Asn Asp Tyr Glu Thr Ala Ile					
[5039]		85		90		95

[5040]	Cys Asn Gly Ile Asn Glu Cys Glu Arg Phe Phe Tyr Gln Gln Ile Asp
[5041]	100 105 110
[5042]	Ser Ile Tyr Met Ser Gln Ile Ile Glu Gln Leu Phe Asp Phe Tyr Ile
[5043]	115 120 125
[5044]	Ala Ser Arg Gln His Asp Met Phe Ile Asn Asn Thr Val Val Pro Tyr
[5045]	130 135 140
[5046]	Asp Val Asn Lys Leu Lys Ser Tyr Tyr Thr Ala Asn Glu Lys Tyr Ser
[5047]	145 150 155 160
[5048]	Phe Glu Gln Phe Cys Asp Asp Ile Lys Glu Phe Thr Asn Lys Gly Phe
[5049]	165 170 175
[5050]	Thr Ser Gly Gly Val Ser Cys Ile Leu Asn Leu Phe Tyr Lys Gly Ser
[5051]	180 185 190
[5052]	Val Lys Asp Ser Lys Asn Lys Lys Asp Tyr Ile Lys Ser Val Lys Arg
[5053]	195 200 205
[5054]	Leu Glu Thr Asn Gly Leu Phe Lys Lys Leu Asn Ile Phe Glu Lys Asn
[5055]	210 215 220
[5056]	Gly Ile Ser Lys Tyr Phe Ala Ala Ser Thr Leu Ser Thr Phe Phe Ala
[5057]	225 230 235 240
[5058]	Thr Ile Ser Ser Trp Lys Lys Gln Asn Asp Asp Trp Thr Gly Val Ala
[5059]	245 250 255
[5060]	Lys Asp Gly Thr Ser Leu Leu Ser Lys Leu Glu Asn Lys Thr Ile Thr
[5061]	260 265 270
[5062]	Leu Gln Ser Ile Ile Lys His His Arg Val Ile Asn Glu Leu Ala Val
[5063]	275 280 285
[5064]	Leu Ile Val Lys Ala Tyr Lys Asp Pro Val Lys Thr Leu Asn Asn Leu
[5065]	290 295 300
[5066]	Phe Glu Glu Arg Ser Asp Asn Asn Asn Asp Phe Lys Tyr Thr Cys Ser
[5067]	305 310 315 320
[5068]	Asp Asp Glu Asp Lys Tyr Pro Met Tyr Ile Lys Arg Glu Ile Ala Glu
[5069]	325 330 335
[5070]	Phe Val Lys Lys His Lys Thr Val Trp Glu Glu Ile Arg Tyr Phe Asp
[5071]	340 345 350
[5072]	Glu Ser Asp Thr Lys Lys Lys Lys Arg Asp Lys Lys Glu Ser Ser Ser
[5073]	355 360 365
[5074]	Asp Asp Lys Ser Tyr Leu Cys Cys Gly Asp Ser Trp Asp Tyr Leu Lys
[5075]	370 375 380
[5076]	Thr Trp Val Arg Leu Tyr Gly Glu Tyr Tyr Phe Phe Asp Asn Ala Leu
[5077]	385 390 395 400
[5078]	Asn Gln Phe Leu Arg Lys Pro Ser Ala Ser Met His Leu Tyr Thr Ser
[5079]	405 410 415
[5080]	Leu Asp Trp Ile Asn Lys Lys Thr Ile Cys Ile Val Gly Ala Asn Tyr
[5081]	420 425 430

[5082]	Tyr Lys Ile Gly Lys Val Glu Val Val Glu Arg Asn Asn Gln Arg Phe
[5083]	435 440 445
[5084]	Leu Leu Val Tyr Val Ser Val Pro Glu Met Glu Asn Tyr Ile Ile Ile
[5085]	450 455 460
[5086]	Pro Leu Gln Leu Asn Lys Tyr Phe Gly Asn Phe Gln Cys Lys Ile Phe
[5087]	465 470 475 480
[5088]	Glu Gly Arg Leu Gln Ala Ile Phe Lys Arg Tyr Ala Asn Phe Asn Ala
[5089]	485 490 495
[5090]	Leu Lys Asn Asn Lys Pro Gln Pro Ser Pro Asn Ile Ser Val Arg Ile
[5091]	500 505 510
[5092]	Asn Glu Phe His Phe Ala Leu Arg Ser Tyr Arg Lys Gln Gln Ile Ser
[5093]	515 520 525
[5094]	Ala Glu Asp Phe Ser Lys Gly Arg Phe Ser Leu Ile Ser Lys Ile Gly
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[5102]	Pro Met Ile Leu Met Gly Val Asp Phe Gly Tyr Ser Pro Leu Ala Ser
[5103]	595 600 605
[5104]	Tyr Asn Ile Lys Pro Leu Gln Thr Gly Lys Pro Ala Thr Asp Trp Val
[5105]	610 615 620
[5106]	Lys Asn Leu His Gly Asn Phe Leu Cys Gln Asn Val Ser Leu Gly Glu
[5107]	625 630 635 640
[5108]	Thr Ile Thr Glu Gly Glu Ile Gly Asp Val Pro Thr Asp Thr Tyr Thr
[5109]	645 650 655
[5110]	Ser Ser Asn Glu Ile Tyr Ser Ile Ala Thr Leu Thr Phe Arg Asn Ala
[5111]	660 665 670
[5112]	Asp Gly Lys Leu Glu Asn Arg Ser Phe Ser Arg Phe Tyr His Glu Leu
[5113]	675 680 685
[5114]	Asn Asn Thr Leu Asn Ile Ile Glu Gln Ile Lys Gly Thr Phe Asn Phe
[5115]	690 695 700
[5116]	Ile His Ser Ile Asn Thr Gln Phe Lys Glu Ile Lys Ala Leu Lys Thr
[5117]	705 710 715 720
[5118]	Thr Glu Glu Phe Ser Ser Tyr Val Ser Thr Leu Thr Trp Asp Gln Phe
[5119]	725 730 735
[5120]	Ile Glu Asp Ser Arg Lys Thr Ala Arg Tyr Ser Lys Tyr Trp Ile His
[5121]	740 745 750
[5122]	Ile Ile Asn Glu Asn Pro Lys Arg Arg Thr Ile Ala Thr Leu Asn Glu
[5123]	755 760 765

[5124]	Thr Leu Lys Leu Val Asp Glu Lys His Arg Phe Thr Val Thr Ile Gln
[5125]	770 775 780
[5126]	Glu Ile Phe Asp Leu Val Lys Tyr Cys Gln Gln His Gly Tyr Tyr Pro
[5127]	785 790 795 800
[5128]	Lys Ser Asn Val Met Ser Lys Leu Arg Asn Leu Ala Ile Lys Leu Ile
[5129]	805 810 815
[5130]	Asn Asp Leu Ile Arg Tyr Gln Lys Ile Gly Ile His Ser Cys Tyr Leu
[5131]	820 825 830
[5132]	Asp Phe Cys Val Leu Ile Lys Asn His Ile Ala Leu Leu Asn Ser Ser
[5133]	835 840 845
[5134]	Thr Ala Phe Ile Ile Asn Phe Ser Arg Asn Lys Glu Asn Ile Ile Arg
[5135]	850 855 860
[5136]	Asn Asn Thr Ser Lys Ile His Ser Leu Trp Val Tyr Arg Asp Asn Phe
[5137]	865 870 875 880
[5138]	Arg Arg Gln Met Ile Lys Asn Leu Cys Ser Gln Ile Leu Lys Ile Ala
[5139]	885 890 895
[5140]	Ala Lys Asn Lys Val His Ile Val Val Val Glu Lys Leu Asn Asn Met
[5141]	900 905 910
[5142]	Arg Thr Asn Asn Arg Asn Asn Glu Asp Lys Asn Asn Met Ile Asp Leu
[5143]	915 920 925
[5144]	Leu Ala Thr Gly Gln Phe Arg Lys Gln Leu Ser Asp Gln Ala Lys Trp
[5145]	930 935 940
[5146]	Tyr Gly Ile Ala Val Val Asp Thr Ala Glu Tyr Asn Thr Ser Lys Val
[5147]	945 950 955 960
[5148]	Asp Phe Met Thr Gly Glu Tyr Gly Tyr Arg Asp Glu Asn Asn Lys Arg
[5149]	965 970 975
[5150]	His Phe Tyr Cys Arg Lys Gln Asp Lys Thr Val Leu Leu Asp Cys Asp
[5151]	980 985 990
[5152]	Lys Lys Ala Ser Glu Asn Ile Leu Leu Ala Phe Val Thr Gln Ser Leu
[5153]	995 1000 1005
[5154]	Leu Leu Asn His Leu Lys Val Leu Ile Thr Glu Asp Gly Lys Thr
[5155]	1010 1015 1020
[5156]	Ala Val Ile Asp Leu Ser Glu Arg Thr Thr Glu Pro Gln Lys Ile
[5157]	1025 1030 1035
[5158]	Arg Ser Lys Ile Trp Thr Asn Ser Asp Val Gln Lys Ile Ile Phe
[5159]	1040 1045 1050
[5160]	Cys Lys Gln Glu Asn Gly Ser Tyr Val Leu Lys Lys Gly Ser Thr
[5161]	1055 1060 1065
[5162]	Asp Ile Lys Glu Lys Met His Lys Ala Val Leu His Arg His Gly
[5163]	1070 1075 1080
[5164]	Ser Leu Trp Tyr Asp Tyr Leu Asn His Lys Asn Met Ile Glu Asp
[5165]	1085 1090 1095

[5166]	Ile Lys Asn Leu His Leu Ser Asn Cys Ser Leu Thr Thr Ser Thr
[5167]	1100 1105 1110
[5168]	Asn Ser Asp Val Ile Asn Ser His Ser Gly Ser Ser Arg Ser Leu
[5169]	1115 1120 1125
[5170]	Asp Lys Thr Lys Thr Tyr Ala Ser Arg Ala Asp Pro Lys Lys Lys
[5171]	1130 1135 1140
[5172]	Arg Lys Val
[5173]	1145
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[5181]	Met Ala Ser Ser Asp Ala Gln Lys Phe Pro Gln Thr His Asn Lys Val
[5182]	1 5 10 15
[5183]	Met Ser Phe Arg Leu Thr Ala Ser Asn Ile Gly Ser Val Leu Ser Leu
[5184]	20 25 30
[5185]	His Ser Asn Leu His Asp Ala Ala Glu Ile Gly Ile Asn Glu Cys Arg
[5186]	35 40 45
[5187]	Trp Trp Ile Gly Asp Gly Glu Ile Tyr Glu Arg Asp Pro Ala Cys Arg
[5188]	50 55 60
[5189]	Ser Ile Lys Lys Gly Asn Asp Ile Arg Thr Val Thr Ser Glu Lys Ile
[5190]	65 70 75 80
[5191]	Lys Glu Leu Trp Thr Lys His Thr Asp His Ser Val Pro Leu Val Asp
[5192]	85 90 95
[5193]	Phe Ile Asp Met Leu Lys Phe Val Ala Gln Cys Ala Ile Tyr Gly Asp
[5194]	100 105 110
[5195]	Ser Arg Ala Leu Ala Ser Thr Leu Phe Gly Lys Ser Lys Ala Glu Thr
[5196]	115 120 125
[5197]	Arg Gly Val Ser Thr Glu Asp Met Thr Val Ile Arg Ala Trp Ile Ala
[5198]	130 135 140
[5199]	Glu Thr Asp Ala Val Leu Ala Ser Gly Leu Ser Pro Lys Lys Lys Lys
[5200]	145 150 155 160
[5201]	Lys Lys Glu Lys Glu Ala Gly Lys Lys Glu Arg Lys Pro Asp Val Lys
[5202]	165 170 175
[5203]	Met Glu Met Cys Arg Arg Ile Arg Cys Thr Met Val Gln Cys Gly Tyr
[5204]	180 185 190
[5205]	Phe Arg Arg Phe Pro Phe Glu Ala Lys Ile Asp Asn Gly Gly Glu Arg
[5206]	195 200 205
[5207]	Gly Lys Met Asp Ser Glu Leu Ser Tyr Val Ser Ala Arg Asn Leu Leu

[5208]	210	215	220
[5209]	Arg Cys Leu Ser Thr Trp Arg Ala Ser Ser Val Met Arg Arg Asp Ser		
[5210]	225	230	235 240
[5211]	Tyr Leu Ile Glu Glu Glu Arg Ile Lys Glu Ala Glu Ser Lys Met Thr		
[5212]		245	250 255
[5213]	Pro Glu Ile Ile Asp Gly Leu Arg Arg Leu Tyr Arg Tyr Cys Ala Val		
[5214]		260	265 270
[5215]	Asp His Asp Phe Leu Lys Trp Phe Gly Gly Arg Ile Ile Arg His Ile		
[5216]		275	280 285
[5217]	Asp Ser Cys Leu Ala Pro Ala Ile Ala Gly Asn Thr Gly Arg Pro Thr		
[5218]	290	295	300
[5219]	Gly Gly Glu Ser Phe Thr Val Ile Tyr Asp Arg Arg Lys Lys Arg Asp		
[5220]	305	310	315 320
[5221]	Val Lys Ile Thr Tyr Ser Val Pro Glu Glu Ile Tyr Gly Tyr Leu Ser		
[5222]		325	330 335
[5223]	Ser His Pro Glu Leu Val Ala Ile Gly Lys Asp Gly Met Thr Pro Ile		
[5224]		340	345 350
[5225]	Ser Arg His Ala Asp Tyr Leu Glu Met Ile Ala Ser His Glu Lys His		
[5226]		355	360 365
[5227]	Arg Trp Tyr Ala Thr Phe Pro Thr Val Gly Lys Glu Asp Gly Tyr Arg		
[5228]	370	375	380
[5229]	Thr Ser Val Leu Leu Gly Lys Asn Tyr Leu Thr Tyr Asp Leu Ser Tyr		
[5230]	385	390	395 400
[5231]	Asp Gly Glu Ser Val Pro Asp Lys Lys Ile Asn Val Ile Ser Lys Gly		
[5232]		405	410 415
[5233]	Gln Pro Val Cys Leu Asp Leu His Asp Gly Arg Arg Val Ser Ser Leu		
[5234]		420	425 430
[5235]	Tyr Leu Thr Val Gly Glu Ser Ala Ala Tyr Asp Ile Ala Val Arg Lys		
[5236]		435	440 445
[5237]	Asn Lys Arg His His Gly Lys Pro Ala Asp Tyr Cys Arg Met Arg Val		
[5238]	450	455	460
[5239]	His Leu Thr Gln Glu Arg Glu Asp Lys Thr Tyr Asn Asp Pro Tyr Phe		
[5240]	465	470	475 480
[5241]	Ser Asn Met Glu Ile Trp Arg Ala Gly Asp Gln Val Tyr Ala Ile Glu		
[5242]		485	490 495
[5243]	Phe Asp Arg His Gly Ala Arg Tyr Thr Ala Ile Val Lys Glu Pro Ser		
[5244]		500	505 510
[5245]	Val Glu Tyr Arg Asn Lys Lys Leu Tyr Leu Arg Val Asn Met Val Leu		
[5246]		515	520 525
[5247]	Asp Ser Pro Ser Arg Gln Asp Asp Lys Asp Met Tyr Tyr Ala Tyr Met		
[5248]	530	535	540
[5249]	Thr Ala Tyr Pro Ser Ser Asn Pro Pro Val Glu Thr Ser Asp Asn Lys		

[5250]	545	550	555	560
[5251]	Lys Arg Phe Glu Arg Leu Gly Pro Gly Arg Arg Ala Ile Gly Gly Ile			
[5252]		565	570	575
[5253]	Asp Ile Gly Ile Gly Arg Pro Tyr Val Ala Val Val Ala Ser Tyr Glu			
[5254]		580	585	590
[5255]	Val Gly Pro Ala Gly Thr Glu Gln Lys Phe Gln Ile Glu Asp Arg Leu			
[5256]		595	600	605
[5257]	Ile Glu Asp Asp Gly Ser Ser Pro Tyr Asp Ser Leu Tyr Asn Asp Phe			
[5258]		610	615	620
[5259]	Leu Thr Asp Ile Arg Thr Val Ser Arg Ile Ile Glu Ala Ala Lys Lys			
[5260]		625	630	635
[5261]	Ile Ser Glu Gly Asp Leu Glu Asp Ile Pro Ser Asp Met Ser Val Asp			
[5262]		645	650	655
[5263]	Glu Asp Gly Ser Ile Ala Ala Thr Met Lys Arg Met Ser Ala Arg Ile			
[5264]		660	665	670
[5265]	Ala Glu Arg His His Leu Tyr Gly Glu Arg Lys Ser Glu Ala Tyr Ala			
[5266]		675	680	685
[5267]	Thr Phe Leu Lys Met Asn His Lys Gln Arg Leu Asp Ile Leu Leu Thr			
[5268]		690	695	700
[5269]	Gln Lys Ala Ser Asn Ala Thr Leu Lys Gln Leu Val Glu Glu Asp Pro			
[5270]		705	710	715
[5271]	Ser Phe Leu Pro Arg Ile Cys Val Tyr Tyr Val Ile Ser Val Glu Arg			
[5272]		725	730	735
[5273]	Glu Leu Lys Asn Lys His Arg Asn Ala Tyr Leu Asp Gly Leu Thr Val			
[5274]		740	745	750
[5275]	Asp Glu Lys Tyr Ser Gly Glu Thr Lys Arg Gly Tyr Ala Gln Lys Arg			
[5276]		755	760	765
[5277]	Leu Asn Ser Met Leu Arg Ala Tyr Ser Ala Leu Gly Glu Glu Glu Thr			
[5278]		770	775	780
[5279]	Asp Glu Val Arg Thr Phe Ser Thr Arg Ser Glu Lys Val Arg Asn Met			
[5280]		785	790	795
[5281]	Ala Lys Asn Ala Ile Lys Arg Asn Ala Arg Lys Leu Val Asn Phe Tyr			
[5282]		805	810	815
[5283]	Val Gly Lys Gly Ile Arg Thr Ile Val Ala Glu Asp Thr Asp Pro Thr			
[5284]		820	825	830
[5285]	Lys Ser Arg Asn Asp Gly Lys Lys Ser Asn Arg Ile Lys Ala Ala Trp			
[5286]		835	840	845
[5287]	Ser Pro Lys Gln Phe Leu Ala Ala Val Lys Asn Ala Ala Gln Trp His			
[5288]		850	855	860
[5289]	Gly Leu Glu Ile Ala Glu Val Asp Pro Arg Met Thr Ser Gln Val His			
[5290]		865	870	875
[5291]	Pro Glu Thr Gly Leu Ile Gly Tyr Arg Asp Gly Asp Thr Leu His Cys			

[5292]		885		890		895
[5293]	Pro Asp Gly Ser Lys Ile Asp Ala Asp Val Ala Gly Ala Ala Asn Val					
[5294]		900		905		910
[5295]	Cys Arg Val Phe Ala Gly Arg Gly Leu Trp Arg Phe Ser Ile Asn Thr					
[5296]		915		920		925
[5297]	Asn Ile Asp Ile Ser Asn Lys Asp Glu Lys Lys Arg Leu Arg Ala Tyr					
[5298]		930		935		940
[5299]	Ile Val His His Phe Gly Ser Glu Ser Asn Trp Glu Lys Phe Arg Lys					
[5300]		945		950		955
[5301]	Gln Tyr Pro Ser Gly Thr Thr Leu Tyr Leu His Gly Arg Glu Trp Leu					
[5302]		965		970		975
[5303]	Thr Ala Glu Glu His Lys Ser Ala Ile Asp Arg Ile Arg Asp Asp Val					
[5304]		980		985		990
[5305]	Gly Arg Asp Ala Glu Asn Asp His Val Ala Ile Val Thr Ala Ala Glu					
[5306]		995		1000		1005
[5307]	Lys Val Glu Ile Phe Ser Arg Ala Asp Pro Lys Lys Lys Arg Lys					
[5308]		1010		1015		1020
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[5316]	<400> 92					
[5317]	Met Ser His Asp Leu Lys Pro Gln Arg Leu Ile Arg Ser Asn Ile Thr					
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[5319]	Lys Thr His Ser Asp Gln Asn Ala Lys Gln Val Ala Glu Glu Val Lys					
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[5321]	Lys Glu His Leu Asn Tyr Leu Leu Ile Lys Asn Glu Met Leu Ile Ser					
[5322]		35		40		45
[5323]	Ile Val Pro Glu Ala Lys Asp Asp Asp Gly Asn Asp Ile Asp Phe Lys					
[5324]		50		55		60
[5325]	Lys Gln Leu Lys Ser Leu Tyr Lys Glu Thr Asp Gln Ser Val Ser Phe					
[5326]		65		70		75
[5327]	Ser Val Phe Cys Gln Met Met Lys Phe Arg Asn Ile Ala Leu Leu Tyr					
[5328]		85		90		95
[5329]	Ala Lys Gly Gln Ser Arg Trp Ala Val Ser Ser Tyr Phe Thr Gly Asn					
[5330]		100		105		110
[5331]	Arg Arg Lys Asp Asp Tyr Ala Lys Asp Leu Ser Leu Leu Asp Glu Ala					
[5332]		115		120		125
[5333]	Ile Glu Leu Leu Glu Cys Lys Arg Arg Lys Lys Ala Glu Glu Glu Asn					

[5334]	130	135	140
[5335]	Glu Glu Glu Asn Glu Thr Pro Lys Lys Lys Glu Asp Asn Pro Ser Asn		
[5336]	145	150	155 160
[5337]	Ile Ser Glu Glu Gln Ile Met Lys Leu Phe Tyr Ala Val Asn Lys Lys		
[5338]		165 170	175
[5339]	Leu Lys Glu Ile Gly Tyr Leu Asp Arg Tyr Ser His Ile Glu Lys Gln		
[5340]		180 185	190
[5341]	Glu Gln Tyr Ala Ile Ile Gly Val Thr Ser Arg Thr Val Lys Ala Trp		
[5342]		195 200	205
[5343]	Asp Tyr Ala Asn Phe Ala Thr Arg Asn His Tyr Gln Ser Val Gln Asn		
[5344]	210	215	220
[5345]	Glu Tyr Gln Lys Lys Leu Lys Ala Leu Pro Gly Thr Lys Lys Asp Lys		
[5346]	225	230	235 240
[5347]	Val Cys Leu Glu Lys Phe Phe Asp His Leu Asn Glu Asn Asn Ile Ala		
[5348]		245 250	255
[5349]	Ala Asp Trp Asp Lys Trp Arg Leu Lys Lys His Ile Leu Gln Cys Ile		
[5350]		260 265	270
[5351]	Ile Pro Ala Ala Lys Ile Gly Leu Lys Glu Leu Lys Gln Ser Phe Tyr		
[5352]		275 280	285
[5353]	Val Asp Asn Lys Gly Asn Lys His Asn Tyr Phe Val Asn Gly Leu Tyr		
[5354]	290	295	300
[5355]	Glu Glu Ile Leu Lys Arg Pro Phe Leu Tyr Ser Ala Glu Asp Pro Glu		
[5356]	305	310	315 320
[5357]	Glu Ser Ile Leu Tyr Leu Gly Val Glu Val Ala Ser Leu His Ser Lys		
[5358]		325 330	335
[5359]	Leu Asn His Leu Arg Ser Glu Ala Arg Phe Ser Phe Glu Thr Pro Asp		
[5360]		340 345	350
[5361]	Asp Ile Cys Lys Tyr Met Thr Ile Cys Gly Asp Asn Tyr His Asn Phe		
[5362]		355 360	365
[5363]	Thr Met Ser Ala Ile Gly Glu Asp Val Glu Asp Ile Glu Val Glu Val		
[5364]	370	375	380
[5365]	Tyr Asp Tyr Asn His Ser Lys Lys Tyr Glu Thr Met Arg Phe Ile Asn		
[5366]	385	390	395 400
[5367]	Gly Lys Arg Thr Thr Asp Leu Ser Leu Asn Phe Lys Gly Ile Pro Val		
[5368]		405 410	415
[5369]	Arg Leu Cys Leu Glu Gly Lys Arg Asn Asn Ser Tyr Phe Ala Asp Ala		
[5370]		420 425	430
[5371]	Ile Val Trp Glu Leu Asp Asn Lys Asp Lys Thr Gly Tyr Leu Ile Glu		
[5372]		435 440	445
[5373]	Tyr Gly Lys Ser Asn Asn Arg Leu Tyr Met Leu Val Lys Glu Pro Leu		
[5374]	450	455	460
[5375]	Ile Gly Cys Arg Arg Lys Phe Gly Lys Asp Val Leu Phe Val Ser Leu		

[5376]	465	470	475	480
[5377]	Ser Gly Thr Leu Val Asn Lys Tyr Ile Glu Asp Asp Ile Val Ser Ala			
[5378]		485	490	495
[5379]	Arg Tyr Leu Met Gln Thr Ala Ala Pro Ile Phe Lys Thr Ser Arg Ala			
[5380]		500	505	510
[5381]	Lys Lys Gln Asp Lys Ile Gly Asp Lys Trp Phe Glu His Cys Gln Gly			
[5382]		515	520	525
[5383]	Ser Thr Ile Lys Ile Ala Gly Ile Asp Ile Gly Ile Asn Pro Ile Ala			
[5384]		530	535	540
[5385]	Ala Ile Thr Val Ala Asn Val Thr Phe Asp Arg Ala Leu Gly Asn Lys			
[5386]		545	550	555
[5387]	Ile Lys Asn Gln Lys Gln Ile Val Ile Asp Cys Tyr Ala Glu Asp Tyr			
[5388]		565	570	575
[5389]	Lys Ile Asp Pro Val Val Val Lys Arg Met Glu Asp Ile Arg His Ile			
[5390]		580	585	590
[5391]	Lys Tyr Thr Ile Asn Ser Trp Tyr His Leu Ala Asp Cys Cys Arg Leu			
[5392]		595	600	605
[5393]	Lys Ala Ala Asn Lys Glu Tyr Val Val Asn Glu Arg Lys Gln Gly Phe			
[5394]		610	615	620
[5395]	Phe Arg Glu Asn Ile Glu Tyr Leu Lys Glu Val Ala Lys Lys Ala Ile			
[5396]		625	630	635
[5397]	Thr Glu Ser Asp Gln Gln Ile Lys Glu Gln Lys Ala Ala Leu Lys Arg			
[5398]		645	650	655
[5399]	Phe Asp Gly Glu Lys Lys Lys Glu Ile Gln Ala Thr Ile Asn Gly Phe			
[5400]		660	665	670
[5401]	Asn Leu Lys Ile Lys Ile Leu Lys Lys Phe Val Arg Gln Ser Ala Lys			
[5402]		675	680	685
[5403]	Lys Ile Phe Asp Ser Thr Leu Glu Thr Leu Glu Lys Tyr Asp Asn Asn			
[5404]		690	695	700
[5405]	Ile Glu Gln Ala Lys Arg Asp Arg Glu Phe Gly Leu Lys Ile Ile Tyr			
[5406]		705	710	715
[5407]	Asp Leu Ile Ile Lys Tyr Tyr Lys Arg Ser Lys Lys Glu Arg Glu Met			
[5408]		725	730	735
[5409]	Asn Gln Arg Ile Tyr Val Asp Asp Tyr Asn Gln Glu Glu Ile Asp Thr			
[5410]		740	745	750
[5411]	Glu Arg Thr Lys Lys Ile Arg Lys Glu Thr Ile Thr Phe Cys Asp Asn			
[5412]		755	760	765
[5413]	Asp Trp Asn Ser Leu Thr Lys Arg Ile His Asp Leu Glu Lys Lys Met			
[5414]		770	775	780
[5415]	Lys Lys Ile Gly Ile Ser Glu Pro Gly Arg Val Glu Gln Glu Ile Asn			
[5416]		785	790	800
[5417]	Asp Arg Asp Tyr Tyr Asn Asn Ile Gln Asp Asn Thr Lys Lys Arg Gln			

[5418]		805		810		815
[5419]	Ala Lys Ile Ile Val Asp Ala Leu Lys Glu Glu Gly Val Ser Ile Ile					
[5420]		820		825		830
[5421]	Val Val Glu Asp Leu Thr Gly Gly Gly Ser Glu Asn Thr Lys Glu Ile					
[5422]		835		840		845
[5423]	Asn Lys Ser Phe Asp Ala Phe Ala Pro Ile Arg Phe Leu Asn Ala Leu					
[5424]		850		855		860
[5425]	Lys Asn Cys Ala Glu Thr Asn Gly Ile Gln Val Thr Glu Val Leu Ser					
[5426]		865		870		875
[5427]	Pro Met Ser Ser Lys Met Val Pro Ser Thr Gly Glu Ile Gly His Arg					
[5428]		885		890		895
[5429]	Asp Lys Arg Asp Lys Gln Leu Tyr Tyr Lys Asp Gly Glu Glu Leu Lys					
[5430]		900		905		910
[5431]	Ser Ile Asp Gly Asp Ile Ser Ala Ser Glu Ile Leu Leu Arg Arg Gly					
[5432]		915		920		925
[5433]	Val Ser Arg His Thr Glu Leu Ile Gly Thr Met Asn Val Glu Asp Val					
[5434]		930		935		940
[5435]	Leu Asp Lys Asn Asn Asn Lys Asn Lys Cys Ile Lys Gly Tyr Val Cys					
[5436]		945		950		955
[5437]	Asn Arg Trp Gly Asn Ile Gln Asn Phe Glu Lys Ile Leu Lys Glu Lys					
[5438]		965		970		975
[5439]	Gly Ile Gly Glu Arg Glu Ile Ile Tyr Leu His Gly Asp Lys Ile Leu					
[5440]		980		985		990
[5441]	Thr Met Asp Glu Lys Arg Thr Leu Gln Ala Ser Ile Arg Lys Glu Leu					
[5442]		995		1000		1005
[5443]	Lys Glu Met Arg Glu Arg Glu Ser Gly Glu Glu Asn Ala Gly Thr					
[5444]		1010		1015		1020
[5445]	Ala Arg Lys Lys Ser Lys Pro Lys Lys Lys Lys Lys Ile Lys Arg					
[5446]		1025		1030		1035
[5447]	Asn Asn Asp Gln Asp Leu Ser Asn Asn Arg Pro Ala Ala Ser Ser					
[5448]		1040		1045		1050
[5449]	Arg Ala Asp Pro Lys Lys Lys Arg Lys Val					
[5450]		1055		1060		
[5451]	<210> 93					
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[5457]	<400> 93					
[5458]	Met Lys Glu Asn Lys Met Lys Glu Asn Gly Ser Met Thr Thr His Ser					
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[5460]	Lys Val Ile Ala Leu Lys Met Lys Ser Glu Asn Val Glu Phe Asp Thr
[5461]	20 25 30
[5462]	Phe Tyr Lys Glu Ser Phe Glu Leu Phe Lys Gln Phe Thr Asn Glu Phe
[5463]	35 40 45
[5464]	Val Ala Trp Gly Asn Asp Glu Ile Tyr Gln Tyr Gly Ser Ser Lys Arg
[5465]	50 55 60
[5466]	Lys Lys Asp Asp Gln Lys Ile Ser Leu Ile Pro Val Ile Glu Asp Ile
[5467]	65 70 75 80
[5468]	Tyr Lys Ser Val Glu Lys Lys Ala Thr Ala Glu Gly Ile Ser Lys Thr
[5469]	85 90 95
[5470]	Asp Phe Arg Ala Val Leu Lys Tyr Leu Tyr His Gln Ile Ile Asn Val
[5471]	100 105 110
[5472]	Gly Asn Ser Gly Arg Ser Tyr Gly Thr Ser Leu Phe Gly Gly Cys Glu
[5473]	115 120 125
[5474]	Val Lys Glu Lys Leu Ser Lys Gln Asp Ile Ser Asn Ile Val Glu Cys
[5475]	130 135 140
[5476]	Val Lys Glu Leu Glu Leu Cys Lys Ser Lys Gln Glu Glu Ser Asp Ala
[5477]	145 150 155 160
[5478]	Tyr Asp Lys Ile Leu Leu Lys Glu Lys Ile Thr His Ile Val Lys Ser
[5479]	165 170 175
[5480]	Gly Glu Thr Ala Gly Asp Ile Thr Lys Lys Tyr Asn Gln Ala Thr Thr
[5481]	180 185 190
[5482]	Gly Arg Lys Thr Ser Ser Lys Gly Phe Phe Asp Lys Ser Thr Lys Thr
[5483]	195 200 205
[5484]	Glu Val Lys Tyr Lys Asp Ile Lys Asp Asp Thr Leu Leu Gln Asp Gly
[5485]	210 215 220
[5486]	Ser Thr Ile Phe Ile Lys Ser Ser Val Asp Leu Phe Val Lys Lys Val
[5487]	225 230 235 240
[5488]	Cys Asn Thr Leu Arg Glu Ile Asn Phe Phe Asp Arg Leu Pro Phe Lys
[5489]	245 250 255
[5490]	Asn Asn His Ser Asn Asn Tyr Gly Leu Leu Phe Ser Met Leu Ser Gln
[5491]	260 265 270
[5492]	Ile Glu Ser Trp Lys Thr Ile Ser Glu Thr Thr Lys Lys Ser His Glu
[5493]	275 280 285
[5494]	Glu His Gly Glu Lys Ile Ala Ser Met Val Lys Lys Leu Asp Leu Thr
[5495]	290 295 300
[5496]	Gln Thr Glu Leu Met Lys Asp Phe Ala Ala Phe Cys Ile Glu Asn Asn
[5497]	305 310 315 320
[5498]	Ile Thr Lys Lys Phe Asp His Lys Phe Lys Arg His Met Glu Asp Cys
[5499]	325 330 335
[5500]	Val Ile Pro Ser Phe Lys Asn Gly Lys Ile Pro Asp Lys Leu Phe Tyr
[5501]	340 345 350

[5502]	Phe Asn Ile Ile Leu Ala Lys Lys Thr Asp Glu Gln Ile Asp Tyr Ser
[5503]	355 360 365
[5504]	Leu Ser Ser Glu Phe Tyr Thr Lys Leu Phe Ser Met Pro Asn Leu Trp
[5505]	370 375 380
[5506]	Gln Glu Glu Glu Ala Phe Ile Val Lys Asn Ile Asn Leu Ile Glu Glu
[5507]	385 390 395 400
[5508]	Ile Thr Ile Phe Asn Lys Arg Arg Asn Tyr Ala Cys Cys Pro Leu Ile
[5509]	405 410 415
[5510]	Lys Glu Lys Glu Tyr Asp Arg Phe Gln Ile Gln Leu Asn Glu Thr Asn
[5511]	420 425 430
[5512]	Phe Leu Lys Phe Gln Phe Asp Pro Lys Asn Val Val Asn Ile Asp Glu
[5513]	435 440 445
[5514]	Asn Thr Thr Glu Ala Thr Val Gly Phe Asp Glu Lys Leu Lys Leu Val
[5515]	450 455 460
[5516]	Val Cys Ala Asp Lys Lys Tyr Ala Phe Ser Ile Phe Thr Gln Cys Lys
[5517]	465 470 475 480
[5518]	Tyr His Gly Asn Lys His Lys Pro Asn Thr Tyr Phe Asn Asn Leu Lys
[5519]	485 490 495
[5520]	Ile Ile Lys Val Ile Glu Ser Lys Ser Asn Ser Val Lys Ser Met Lys
[5521]	500 505 510
[5522]	Tyr Thr Phe Glu Phe Thr Lys Arg Asn Glu Leu Lys Arg Ala Glu Ile
[5523]	515 520 525
[5524]	Lys Gln Pro Ser Ile Val Tyr Lys Asn Asn Asn Tyr Tyr Ile Arg Ile
[5525]	530 535 540
[5526]	Asn Met Asn Val Ile Leu Asp Ala Asp Gln Thr Ser Tyr Lys Ile Ile
[5527]	545 550 555 560
[5528]	Asn Asn Asn Gln Thr Ala Ser Leu Pro Ser Tyr Phe Gln Ser Ser Leu
[5529]	565 570 575
[5530]	Pro Phe Glu Asn Asn Arg Gly Lys Ile His Asp Lys Gly Ile Val His
[5531]	580 585 590
[5532]	Trp Glu Lys Ile Lys Asn Arg Lys Ile Ile Ala Met Gly Val Asp Leu
[5533]	595 600 605
[5534]	Gly Val Arg Arg Pro Phe Ser Tyr Ala Ile Gly Asn Phe Thr Leu Asn
[5535]	610 615 620
[5536]	Lys Asp Ile Leu Asp Lys Asn Asp Val Asn Ile Val Ala Ser Gly Phe
[5537]	625 630 635 640
[5538]	Asn Leu Cys Ser Asp Ser Asp Val Tyr Phe Gln Val Phe Asn Gln Ile
[5539]	645 650 655
[5540]	Lys Thr Leu Ala Lys Phe Ile Gly Lys Leu Lys Ser His Asn Lys Gly
[5541]	660 665 670
[5542]	Leu Lys Val Asp Phe Glu Lys Asp Lys Lys Tyr Ile Phe Asp Leu Val
[5543]	675 680 685

[5544]	Asn Asp Ala Lys Ala Tyr Phe Lys Asp Met Ser Ala Lys Arg Ile Asn
[5545]	690 695 700
[5546]	Asp Thr Lys Asp Asn Ile Ser Asn Thr Val Thr Asn Lys Glu Arg Ile
[5547]	705 710 715 720
[5548]	Tyr Gly Ser Phe Val Ser Glu Ser Ala Glu Ser Ala Ile Gln Cys Ala
[5549]	725 730 735
[5550]	Ile Asp Arg Ser Glu Lys Glu Ser Gly Leu Thr Leu Lys Lys Asp Ile
[5551]	740 745 750
[5552]	Ser Trp Leu Val Asn Val Leu Ser Lys Tyr Leu Glu Arg Lys Phe Lys
[5553]	755 760 765
[5554]	Glu Val Lys Asn Asn Arg Lys Tyr Thr Asn Val Asn Lys Cys Asp Asn
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[5556]	Cys Phe Asn Trp Leu Arg Val Ile Glu Asn Ile Lys Arg Leu Lys Arg
[5557]	785 790 795 800
[5558]	Ser Ile Ser Tyr Leu Gly Glu Asp Leu Gln Lys Asn Pro Glu Leu Lys
[5559]	805 810 815
[5560]	Ile Glu Leu Lys Asn Leu Asn Glu Tyr Gly Asn Asn Val Lys Ser Asp
[5561]	820 825 830
[5562]	Phe Leu Lys Gln Ile Ala Ser Asn Ile Ile Lys Val Ala Ile Glu His
[5563]	835 840 845
[5564]	Lys Cys Asp Ile Val Phe Ile Glu Lys Leu Gly Lys Ala Asp Ser Arg
[5565]	850 855 860
[5566]	Ser Arg Lys Leu Asn Glu Met Phe Ser Phe Trp Ser Pro Lys Ala Ile
[5567]	865 870 875 880
[5568]	Lys Lys Ala Ile Glu Asn Ala Ala Ser Trp His Gly Ile Pro Val Val
[5569]	885 890 895
[5570]	Glu Val Asp Pro Ser Cys Thr Ser Lys Val His Tyr Glu Thr Asn Leu
[5571]	900 905 910
[5572]	Phe Gly His Arg Ile Gly Asn Asp Leu Tyr Tyr Val Glu Asp Gln Cys
[5573]	915 920 925
[5574]	Leu Lys Lys Val Asp Ala Asp Ile Asn Ala Ala Lys Gln Ile Leu Val
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[5576]	Arg Gly Ala Thr Arg His Gly Asn Ile Ser Ser Ile Asn Ile Lys Tyr
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[5578]	Leu Gln Ala Lys Ile Ala Glu Leu Asn Ser Glu Ala Asn Ser Glu Glu
[5579]	965 970 975
[5580]	Asp Lys Glu Glu Ile Lys Gln Gly Gly Lys Arg Ile Gln Gly Phe Leu
[5581]	980 985 990
[5582]	Trp Lys Lys Tyr Gly Asn Ile Thr Asn Ile Thr Asn Gln Leu Thr Ala
[5583]	995 1000 1005
[5584]	Ala His Lys Glu Arg Glu Ser Lys Phe Asp Tyr Ile Tyr Leu His
[5585]	1010 1015 1020

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[5604]	35 40 45
[5605]	Glu Met Phe Phe Leu Met Arg Gly Gln Pro Tyr Arg Arg Arg Ile Gly
[5606]	50 55 60
[5607]	Ser Glu Glu Lys Gln Val Thr Gln Glu His Ile Asp Ala Arg Leu Arg
[5608]	65 70 75 80
[5609]	Val Leu Val Gly Asp Tyr Ser Leu Asn Glu Val Lys Pro Leu Leu Arg
[5610]	85 90 95
[5611]	Gln Leu Tyr Asp Gly Ile Lys Ala Lys Gln Asn Tyr Ala Pro Thr His
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[5613]	Phe Val Arg Phe Phe Ile Gln Pro Thr Lys Gly Ala Ile Asp Lys Lys
[5614]	115 120 125
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[5617]	Met Gly Val Leu Pro Ile Leu Pro Leu Ser Pro Gly Phe Lys Phe Trp
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[5619]	Thr Ala Ala Met Met Met Ala Cys Ser Arg Met Asn Ser Trp Glu Ala
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[5623]	Glu Asn Tyr Lys Lys Glu Ile Arg Phe Glu Asp Leu Cys Glu Glu Trp
[5624]	195 200 205
[5625]	Ser Leu Phe Ser Asp Trp Leu Thr Glu Ala Glu Ser Asp Asn Glu Gly
[5626]	210 215 220
[5627]	Gly Cys Lys Phe Lys Leu Thr Pro Arg Phe Leu Gln Arg Trp Glu Arg

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[5630]		245	250	255
[5631]	Leu Gly Pro Val Met Glu Ala Leu Ala Gly Asp Lys Tyr Arg Gln Leu			
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[5633]	Trp Asp Asn Gly Glu Glu Arg Asp Tyr Ile Thr Glu Leu Gly Asp Leu			
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[5635]	Val Thr Ser Gln Arg Lys Ala Val Arg Leu Ser Arg Asp Ser Ala Val			
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[5637]	Thr Phe Pro Asp Glu Glu Leu Ser Pro Val Gly Thr Glu Phe Gly His			
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[5639]	Asn Tyr Met Ser Phe Ser Ile Asp Gln Glu Asn Ser His Leu Val Thr			
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[5647]	Gly Asp Phe Lys Glu Ala Thr Val Arg Asn Arg Arg Ser Leu Lys Thr			
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[5661]	Pro Asp Asp Leu Gly Tyr Ala Val Cys Leu Glu Ser Trp Val Asp Gly			
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[5663]	Val Glu Lys Asn His Lys Val Ala Gln Glu Met Lys Asp Trp Arg Arg			
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[5665]	Glu Cys Leu Ala Ala Gln Arg Leu Ile His Tyr Ala Lys Phe Leu Lys			
[5666]		530	535	540
[5667]	Lys Arg Asp Lys Asn Glu Glu Ile Asp Tyr Lys His Glu Glu Ser Leu			
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[5669]	Glu Thr Ile Ala Gly Leu Leu Gly Ile Glu Ile Asp Pro Glu Gln Ile			

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[5671]	Ile Asp Val Pro	Leu Lys Leu Leu Asp Leu Val Gly Gln Ala Ile Gly				
[5672]		580		585		590
[5673]	Ala Leu Arg Lys Lys Tyr Leu Val Leu Lys Lys Asn Glu Val Arg Gln					
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[5675]	Gly Arg Ile Thr Ser Glu Leu Phe Leu Trp Pro Glu Cys Val Asp Thr					
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[5677]	Tyr Ile Arg Leu Leu Lys Ser Trp Thr Tyr Lys Asp Lys Lys Pro Tyr					
[5678]		625		630		635
[5679]	Gln Lys Gly Glu Thr Asn Lys Asp Ala Phe Lys Lys Leu Lys Gly Tyr					
[5680]		645		650		655
[5681]	Leu Ala Arg Leu Arg Lys Asp Leu Ala Pro Lys Tyr Ala Ala Val Ile					
[5682]		660		665		670
[5683]	Ala Asp Ala Ala Ile Arg His Lys Val His Val Val Val Ala Glu Asn					
[5684]		675		680		685
[5685]	Leu Glu Gln Phe Gly Leu Ser Met Lys Asn Glu Lys Asp Leu Asn Arg					
[5686]		690		695		700
[5687]	Val Leu Ala His Trp Ser His Gln Lys Ile Trp Ser Met Val Glu Glu					
[5688]		705		710		715
[5689]	Gln Leu Arg Pro Tyr Gly Ile Met Val Val Tyr Val Asp Pro Arg His					
[5690]		725		730		735
[5691]	Thr Ser Lys Leu Asp Phe Ala Thr Asp Glu Phe Gly Gly Arg Cys Phe					
[5692]		740		745		750
[5693]	Thr Ser Leu Tyr Val Met Arg Asp Gly Lys Lys Thr Thr Thr Asp Thr					
[5694]		755		760		765
[5695]	Glu Lys Asn Ala Ser Gln Asn Ile Pro Lys Lys Phe Leu Thr Arg His					
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[5697]	Arg Asn Val Ser Trp Leu Leu Ala Tyr Ala Val Asp Leu Ser Asp Ser					
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[5699]	Gln Lys Lys Lys Leu Gly Ile Gly Asp Glu Lys Val Trp Leu Pro Asn					
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[5701]	Met Gly Leu Met Ile Ser Gly Ala Leu Lys Ala Lys His Gly Lys Asn					
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[5703]	Ser Ala Leu Leu Val Glu Asp Gly Glu Asn Tyr Arg Leu Leu Pro Ile					
[5704]		835		840		845
[5705]	Thr Ala Ala Gln Ala Lys Lys Phe Val Val Lys Arg Lys Lys Glu Glu					
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[5707]	Phe Tyr Arg His Gly Glu Ile Trp Leu Thr Lys Glu Ala His Lys Ala					
[5708]		865		870		875
[5709]	Arg Ile Glu Tyr Leu Phe Pro Glu Ser Lys Lys Gly Arg Lys Ser Ser					
[5710]		885		890		895
[5711]	Arg Ala Asp Pro Lys Lys Lys Arg Lys Val					

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[5726]	Thr Ile Leu Ser Glu Pro Ala Leu His Phe Gln Phe Val Asn Gly Ile			
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[5728]	Leu Pro Val Lys Asn Gly Pro Gly Ala Asp Asp Ser Ser Trp Arg His			
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[5730]	Ser Glu Asn Cys Tyr Ser Met Leu Phe Glu Lys Asn Ser Lys Ser Gly			
[5731]		85	90	95
[5732]	Lys Ser Asp Gly Lys Val Arg Gln Val Arg Glu Leu Lys Val Ala Leu			
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[5734]	Phe Gly Lys Lys Glu Lys Gly Lys Gly Ile Val Gly Lys Lys Thr Trp			
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[5750]	Ser Ala Tyr Gln Gln Phe Lys Gln Phe Ala Asp Glu Leu Leu Thr Gln			
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[5752]	Glu Gly Tyr Arg Ile Ser Gly Arg Val Leu Arg Ala Val Glu Lys Lys			
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[5758]	Ala Phe Asn Lys Lys Lys Ala Asp Ala Arg Val Thr Val Cys Ser Glu
[5759]	305 310 315 320
[5760]	Thr Ser Pro Leu Gln Phe Pro Phe Gly Met Thr Gly Asn Gly Tyr Pro
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[5762]	Phe Thr Leu Ser Ala Cys Glu Gly Arg Ile Asn Ala Thr Ile His Phe
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[5764]	Pro Gly Gly Asp Leu Pro Leu Arg Leu Arg Lys Ser Lys Tyr Phe Gln
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[5766]	Asn Pro Glu Ile Leu Pro Val Lys Asp Gly Phe Gln Ile Thr Phe Thr
[5767]	370 375 380
[5768]	Arg Gly Lys Thr Pro Leu Val Gly Thr Ile Lys Glu Pro Ser Leu Leu
[5769]	385 390 395 400
[5770]	Lys Lys Asn Asn His Tyr Tyr Leu Ser Leu Arg Val Asn Val Pro Ser
[5771]	405 410 415
[5772]	Val Lys Ile Pro Lys Glu Val Arg Asp Thr Arg Ala Tyr Tyr Ser Ser
[5773]	420 425 430
[5774]	Ala Val Gly Gly Asp Glu Thr Thr Pro Val Pro Val Lys Ala Val Ala
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[5776]	Ile Asp Leu Gly Val Thr Thr Leu Ala Asp Tyr Ser Ile Ile Asp Thr
[5777]	450 455 460
[5778]	Cys Leu Pro Gly Asp Cys Lys Val Phe Gly Gly Glu Thr Ala Ala Phe
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[5780]	Thr Ala His Gly Lys Ile Gly Gln Cys Ala Asn Lys Ser Leu Arg Asp
[5781]	485 490 495
[5782]	Arg Leu Tyr Lys Asn Thr Glu Glu Ala Leu Phe Leu Gly Lys Phe Ile
[5783]	500 505 510
[5784]	Arg Leu Ser Lys Lys Leu Arg Asp Gly Glu Gly Leu Asn Arg Trp Glu
[5785]	515 520 525
[5786]	Val Glu Lys Leu Pro Gly Tyr Ala Glu Arg Leu Gly Ile Thr Gln His
[5787]	530 535 540
[5788]	Leu Asp Asn Ala Tyr Thr Arg Lys Asp Glu Ile Ala Arg Lys Phe Lys
[5789]	545 550 555 560
[5790]	Gln Ile Lys Gly Asn Phe Asp Lys Leu Val Ser Glu Phe Ala Leu Arg
[5791]	565 570 575
[5792]	Asp His Pro Ser Lys Lys Gly Glu Ser Trp Glu Thr Ile Ser Ala Glu
[5793]	580 585 590
[5794]	Thr Ile Gln Val Leu Ala Ala Leu Lys Arg Ile Gln Ser Leu Leu Lys
[5795]	595 600 605

[5796]	Ser Trp Thr Tyr Tyr Ser Trp Thr Ala Glu Asp Tyr Val Leu Ala Leu
[5797]	610 615 620
[5798]	Thr Ala Asp Gly Pro Val Cys Ile Asp Gly Glu His Val Lys Ala Val
[5799]	625 630 635 640
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[5804]	Gln Leu Ala Thr Gly Val Lys His Arg Asn His Pro Val Asn Phe Leu
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[5812]	Lys Asp Ala Tyr Glu Asn Tyr Arg Trp Asn Leu Phe Ala Pro Ala Thr
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[5814]	Ile Val Lys Lys Leu Glu Ala Ala Leu Leu Glu Val Gly Ile Ala Met
[5815]	755 760 765
[5816]	Ala Gln Val Asp Pro Arg His Thr Ser Gln Ile Ala Pro Thr Gly Ala
[5817]	770 775 780
[5818]	Phe Gly Phe Arg Asp His Ala Phe Leu Tyr Tyr Gln Asp Asp Gly Leu
[5819]	785 790 795 800
[5820]	Cys Arg Ile Asp Ala Asn Thr Asn Ala Ser Met Arg Ile Ala Glu Arg
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[5824]	Gly Glu Thr Glu Tyr Leu Ile Pro Glu Ser Ala Ser Lys Arg Leu Asn
[5825]	835 840 845
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[5827]	850 855 860
[5828]	Asn Cys Ser Gly Phe Val Leu Glu Gly Leu Thr Lys Lys Gln Tyr Ala
[5829]	865 870 875 880
[5830]	Lys Leu Ala Glu Thr Ala Gly Lys Lys Glu Ser Phe Tyr Gln Tyr Asp
[5831]	885 890 895
[5832]	Asp Arg Trp Phe Asp Lys Gly His His Phe Ala Cys Arg Ala Thr Leu
[5833]	900 905 910
[5834]	Glu Asn Lys Val Gln Val Cys Leu Asn Gly Gly Gly Arg Ile Lys Asp
[5835]	915 920 925
[5836]	Thr Thr Pro Asp Phe Asn Pro Lys Ser Leu Leu Arg Ser Asp Leu Gln
[5837]	930 935 940

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[5883]	Val Glu Gly Lys Asp Ala Asp Tyr Gln Pro Val Leu Ser Leu Leu Thr					
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[5885]	Gln Tyr Pro Asp Leu Gln Gly Asp Phe Glu Glu Leu Gly Arg Val Tyr					
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[5887]	Leu Ala Glu Ala Glu Tyr Leu Arg Lys Lys Val Asp Ala Arg Val Thr					
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[5889]	Val Cys Asp Ala Glu Thr Ser Pro Leu Gln Phe Pro Phe Gly Leu Thr					
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[5891]	Gly Asn Gly Tyr Ser Ile Thr Leu Thr Val Val Lys Gly Gln Ile Ala					
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[5893]	Ala Thr Leu His Leu Pro Gly Gly Asp Ile Thr Pro Arg Leu Arg Arg					
[5894]		355		360		365
[5895]	Ser Lys Tyr Phe Gln Asn Pro Glu Ile Ala Pro Val Lys Asp Gly Lys					
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[5897]	Gly Lys Val Asn Gly Phe Gln Ile Ser Phe Lys Arg Gly Lys Thr Pro					
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[5911]	Arg Cys Gly Ser Lys Ser Leu Arg Asp Gly Leu Tyr Lys Asn Thr Glu					
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[5913]	Ala Gly Tyr Phe Leu Ala Lys Tyr Ile Arg Leu Ser Lys Asn Leu Arg					
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[5915]	Gly Gly Val Gly Leu Asn Lys Leu Glu Lys Glu Lys Leu Leu Glu His					
[5916]		530		535		540
[5917]	Val Glu Arg Leu Gly Ile Glu His Cys Ala Asp Asp Phe Ala Arg Lys					
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[5919]	Asp Glu Ile His Arg Lys Phe Ser Glu Ile Lys Ser Lys Leu Glu Lys					
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[5921]	Ser Ile Ser Glu Phe Ala Leu Arg Asp Arg Pro Asp Lys Lys Gly Ala					

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[5927]	Ala Glu Asp Tyr Val Leu Ala Leu Thr Gly Glu Gly Arg Thr Arg Val		
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[5929]	Ser Asp Glu His Val Glu Ser Val Val Lys Thr Gly Arg Arg Gln Phe		
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[5931]	Ala Pro Cys Gly Lys Ala Ala Leu Leu Arg Leu Leu Glu Lys Gly Lys		
[5932]	660	665	670
[5933]	Ile Val Glu Val Cys Pro Gly Gln Phe Gln Leu Ala Glu Gly Val Asp		
[5934]	675	680	685
[5935]	Tyr Lys Arg His Pro Thr Glu Phe Leu Ala Ala His Ile Arg His Phe		
[5936]	690	695	700
[5937]	Asn Gly Leu Arg Arg Asp Leu Thr Asn Lys Leu Val Arg Ala Ile Val		
[5938]	705	710	715 720
[5939]	Glu Lys Ala Gln Gln His Arg Val Gln Ile Val Ile Val Glu Asp Phe		
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[5941]	Gly Ile Pro Asp Ile Glu Gly Arg Ile Met Asp His Tyr Asp Asn Tyr		
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[5943]	Arg Trp Asn Leu Phe Ala Pro Ala Lys Val Ile Glu Lys Leu Glu Glu		
[5944]	755	760	765
[5945]	Ala Leu Ser Glu Val Gly Ile Ala Met Ala Glu Val Asp Pro Arg His		
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[5947]	Thr Ser Gln Leu Ala Pro Thr Gly Asp Phe Gly Phe Arg Asp His Glu		
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[5949]	Asn Leu Tyr Phe Trp Glu Lys Gly Leu Cys Arg Thr Asp Ala Asn Thr		
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[5951]	Asn Ala Ser Met Arg Ile Ala Glu Arg Phe Phe Thr Arg His Ser Val		
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[5953]	Leu Ser Gln Leu Arg Ala Val Lys Ile Ser Glu Thr Glu Phe Leu Ile		
[5954]	835	840	845
[5955]	Pro Val Ser Thr Gly Lys Arg Glu Asn Ala Phe Ile Lys Ser Gln Thr		
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[5957]	Gly Lys Leu Phe Ala Lys Leu Val Ala Asp Ser Asn Gly Phe Val Met		
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[5959]	Val Gly Leu Thr Glu Lys Gln His Gly Ala Thr Val Thr Val Gly Lys		
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[5961]	Lys Val Ser Phe Tyr Asn His Ala Gly Arg Trp Leu Gly Lys Ala His		
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[5963]	His Ile Ala His Arg Asp Arg Ile Lys Asn Glu Val Asn Gln Val Leu		

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[5967]	Lys Thr Ser Arg Ala Asp Pro Lys Lys Lys Arg Lys Val		
[5968]	945	950	955
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[5986]	His Glu Gly Val Lys Tyr Leu Lys Gln Tyr Tyr Glu Ser Cys Val Asp		
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[5988]	Ala Gly Lys Pro Ala Lys Tyr Ala Ala Asn Met Phe Leu Thr Lys Thr		
[5989]	100	105	110
[5990]	Ile Ser Gly Thr Asn Pro Leu Gln Cys His Thr Ala Val Tyr Lys Leu		
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[5992]	Tyr Lys Lys Val Gln Ala Lys Gln Ile Thr Lys Lys Glu Phe Ile Asp		
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[5994]	Lys Leu Tyr Ser Lys Thr Lys Lys Lys Lys Ser Leu Lys Pro Ala Tyr		
[5995]	145	150	155
[5996]	Lys Val Phe Thr Glu Asn Glu His Ile Glu Phe Tyr His Lys Val Arg		
[5997]	165	170	175
[5998]	Ser Gly Lys Leu Pro Ala Ser Glu Val Arg Leu Glu Glu Ser Arg Arg		
[5999]	180	185	190
[6000]	Ala Pro Asp Val Gly Leu Glu Val Gly Leu Leu Leu Arg Glu Leu Gly		
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[6004]	Leu Ala Trp Thr Ile Ala Ile Arg Trp Leu Lys Asn Trp Asn Glu Asn		
[6005]	225	230	235
			240

[6006]	Asn Lys Asn Thr Ala Lys Glu Lys Ala Lys Gln Lys Ala Ile Val Asp
[6007]	245 250 255
[6008]	Lys Leu Arg Thr Ser Leu Asp Gln Lys Glu Val Asp Leu Phe Glu Glu
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[6010]	Phe Ala Glu Glu Cys Ser Gln Glu Gln Phe Gly Ile Arg Glu Gly Phe
[6011]	275 280 285
[6012]	Val Lys Ala Lys Lys Arg Leu Lys Ser Phe Pro Lys Gly Ile Glu Lys
[6013]	290 295 300
[6014]	Ser Ser Tyr Lys Glu Gly Met Arg Ile Leu Val Gln Asn Lys His Gly
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[6016]	Ser Ile Trp Asp Asn Phe Glu Asn Leu Ala Tyr His His Ile Ala Leu
[6017]	325 330 335
[6018]	Asn Glu Tyr Asn Arg Leu Arg Asp Glu Ala Ser Phe Ser Phe Pro Asp
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[6020]	Pro Ile Tyr His Pro Ile Arg Ala Glu Phe Gly Leu Thr Ser Leu Pro
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[6024]	Asn Leu Pro Asp Gly Pro Leu Met Met Leu Gly Lys Lys Ser Arg Tyr
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[6030]	Lys Ser Ile Thr Val Val Phe Ala Lys Ser Ser Ile Tyr Val Gly Leu
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[6032]	Pro Tyr Arg Pro Ile Ser Ile Pro Ile Pro Gln Ala Val Thr Asn Ser
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[6034]	Thr Tyr Tyr Phe Lys Lys Asn Leu Ser Ser Thr Ser Lys Phe Asp Lys
[6035]	465 470 475 480
[6036]	Asp Val Phe Met Gly Leu Thr Ala Val Ser Val Asp Leu Gly Leu Asn
[6037]	485 490 495
[6038]	Pro Val Phe Ser Met Ser Ala Cys Arg Leu Asp Glu Met Lys Ala Asp
[6039]	500 505 510
[6040]	Glu His Tyr Ser Cys Glu Val Pro Gly Phe Gly Trp Ala Asn Gln Ile
[6041]	515 520 525
[6042]	Trp Ser Lys Arg Ala Gly Gly Val Trp Asn Arg Ser Phe Arg Asp Lys
[6043]	530 535 540
[6044]	Ile Arg Gly Phe Val Pro Gly Asn Leu Ser Asp Arg Ile Phe Cys Cys
[6045]	545 550 555 560
[6046]	Lys Lys Ser Ile Ile Val Ser Lys Lys Leu Arg Asp Glu Lys Pro Leu
[6047]	565 570 575

[6048]	Thr Gln Tyr Glu Glu Glu Asn Phe Glu Arg Trp Met Gln Val Val Gly
[6049]	580 585 590
[6050]	Val Asp Pro Asn Glu Asp His Tyr Lys Gln Leu Arg Ile Ala Ile Arg
[6051]	595 600 605
[6052]	Asp Ile Lys Thr Glu Tyr Glu Thr Val Arg Ser Glu Phe Ala Leu Arg
[6053]	610 615 620
[6054]	Asp His Pro Asn Asn Ser Asn Lys Thr Thr Glu Asn Ile Cys Thr Glu
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[6056]	Cys Phe Asp Met Leu Phe Val Ile Lys Asn Leu Ile Ser Leu Leu Lys
[6057]	645 650 655
[6058]	Ser Trp Asn Arg Trp His Arg Thr Thr Gly Asp Ile Glu Glu Arg Gly
[6059]	660 665 670
[6060]	Lys Asp Pro Asn Glu Cys Ser Thr Tyr Trp Arg His Tyr Asn Gly Leu
[6061]	675 680 685
[6062]	Lys Thr Asp Leu Leu Lys Lys Leu Thr Asn Ile Leu Ile Glu Ser Ala
[6063]	690 695 700
[6064]	Lys Ser Ile Gly Ala His Ile Ile Ile Leu Glu Asp Leu Thr Leu Ser
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[6066]	Gln Arg Ser Ser Arg Ser Arg Arg Glu Asn Ser Leu Val Ala Ile Phe
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[6070]	Gly Ile Leu Val Tyr Leu Glu Asp Pro Arg His Ser Ser Gln Ile Ser
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[6072]	Ile Val Thr Asn Glu Phe Gly Tyr Arg Pro Lys Glu Asp Lys Ala Lys
[6073]	770 775 780
[6074]	Leu Tyr Phe Met Asp Glu Glu Thr Val Cys Val Thr Asn Cys Asp Asp
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[6076]	Ser Ala Ala Leu Met Leu Gln Gln Ser Phe Trp Ser Arg His Lys Asp
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[6080]	Ser Ser Glu Asp Lys Asp Gly Thr Lys Met Arg Leu Arg Ser Tyr Leu
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[6083]	850 855 860
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[6086]	Glu Phe Gly Lys Asp Glu Tyr Phe Tyr Arg His Gly Glu Gln Trp Phe
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[6088]	Thr Ala Asp Ala His Phe Asp Lys Leu Arg Glu Phe Gly Asn Gln Val
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[6090]	Phe Leu Thr Pro Gln Ser Gln Ile Lys Arg Ile Asn Leu Gln Val Glu
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[6092]	Gly Thr Ser Arg Ala Asp Pro Lys Lys Lys Arg Lys Val
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[6105]	Met Arg Leu Leu Leu Ala Met Arg Arg Glu Pro Tyr Ser Leu Arg Met
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[6108]	50 55 60
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[6111]	Arg Ser Phe Ala Leu Ala Val Leu His Gln Asp Asn Pro Lys Lys Arg
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[6113]	Ala Phe Leu Glu Ser Glu Asn Cys Val Ser Ile Leu Cys Leu Glu Lys
[6114]	100 105 110
[6115]	Ser Ala Ser Gly Thr Arg Tyr Tyr Lys Arg Pro Gly Tyr Gln Leu Leu
[6116]	115 120 125
[6117]	Lys Lys Ala Ile Glu Glu Glu Trp Gly Trp Asp Lys Phe Glu Ala Ser
[6118]	130 135 140
[6119]	Leu Leu Asp Glu Arg Thr Gly Glu Val Ala Glu Lys Phe Ala Ala Leu
[6120]	145 150 155 160
[6121]	Ser Met Glu Asp Trp Arg Arg Phe Phe Ala Ala Arg Asp Pro Asp Asp
[6122]	165 170 175
[6123]	Leu Gly Arg Glu Leu Leu Lys Thr Asp Thr Arg Glu Gly Met Ala Ala
[6124]	180 185 190
[6125]	Ala Leu Arg Leu Arg Glu Arg Gly Val Phe Pro Val Ser Val Pro Glu
[6126]	195 200 205
[6127]	His Leu Asp Leu Asp Ser Leu Lys Ala Ala Met Ala Ser Ala Ala Glu
[6128]	210 215 220
[6129]	Arg Leu Lys Ser Trp Leu Ala Cys Asn Gln Arg Ala Val Asp Glu Lys
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[6131]	Ser Glu Leu Arg Lys Arg Phe Glu Glu Ala Leu Asp Gly Val Asp Pro

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[6133]	Glu Lys Tyr Ala Leu Phe Glu Lys Phe Ala Ala Glu Leu Gln Gln Ala					
[6134]		260		265		270
[6135]	Asp Tyr Asn Val Thr Lys Lys Leu Val Leu Ala Val Ser Ala Lys Phe					
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[6137]	Pro Ala Thr Glu Pro Ser Glu Phe Lys Arg Gly Val Glu Ile Leu Lys					
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[6139]	Glu Asp Gly Tyr Lys Pro Leu Trp Glu Asp Phe Arg Glu Leu Gly Phe					
[6140]		305		310		315
[6141]	Val Tyr Leu Ala Glu Arg Lys Trp Glu Arg Arg Arg Gly Gly Ala Ala					
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[6149]	Val Pro Ser Arg Tyr Phe Trp Asn Pro Ser Val Gly Arg Thr Thr Ser					
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[6153]	Tyr Val Gly Glu Val Lys Glu Ile Gly Leu Val Arg Gln Arg Gly Arg					
[6154]		420		425		430
[6155]	Tyr Tyr Phe Phe Ile Asp Tyr Asn Phe Asp Pro Glu Glu Val Ser Asp					
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[6157]	Glu Thr Lys Val Gly Arg Ala Phe Phe Arg Ala Pro Leu Asn Glu Ser					
[6158]		450		455		460
[6159]	Arg Pro Lys Pro Lys Asp Lys Leu Thr Val Met Gly Ile Asp Leu Gly					
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[6161]	Ile Asn Pro Ala Phe Ala Phe Ala Val Cys Thr Leu Gly Glu Cys Gln					
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[6163]	Asp Gly Ile Arg Ser Pro Val Ala Lys Met Glu Asp Val Ser Phe Asp					
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[6165]	Ser Thr Gly Leu Arg Gly Gly Ile Gly Ser Gln Lys Leu His Arg Glu					
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[6167]	Met His Asn Leu Ser Asp Arg Cys Phe Tyr Gly Ala Arg Tyr Ile Arg					
[6168]		530		535		540
[6169]	Leu Ser Lys Lys Leu Arg Asp Arg Gly Ala Leu Asn Asp Ile Glu Ala					
[6170]		545		550		555
[6171]	Arg Leu Leu Glu Glu Lys Tyr Ile Pro Gly Phe Arg Ile Val His Ile					
[6172]		565		570		575
[6173]	Glu Asp Ala Asp Glu Arg Arg Arg Thr Val Gly Arg Thr Val Lys Glu					

[6174]	580	585	590
[6175]	Ile Lys Gln Glu Tyr Lys Arg Ile Arg His Gln Phe Tyr Leu Arg Tyr		
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[6177]	His Thr Ser Lys Arg Asp Arg Thr Glu Leu Ile Ser Ala Glu Tyr Phe		
[6178]	610	615	620
[6179]	Arg Met Leu Phe Leu Val Lys Asn Leu Arg Asn Leu Leu Lys Ser Trp		
[6180]	625	630	635
[6181]	Asn Arg Tyr His Trp Thr Thr Gly Asp Arg Glu Arg Arg Gly Gly Asn		
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[6183]	Pro Asp Glu Leu Lys Ser Tyr Val Arg Tyr Tyr Asn Asn Leu Arg Met		
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[6185]	Asp Thr Leu Lys Lys Leu Thr Cys Ala Ile Val Arg Thr Ala Lys Glu		
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[6187]	His Gly Ala Thr Leu Val Ala Met Glu Asn Ile Gln Arg Val Asp Arg		
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[6189]	Asp Asp Glu Val Lys Arg Arg Lys Glu Asn Ser Leu Leu Ser Leu Trp		
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[6191]	Ala Pro Gly Met Val Leu Glu Arg Val Glu Gln Glu Leu Lys Asn Glu		
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[6193]	Gly Ile Leu Ala Trp Glu Val Asp Pro Arg His Thr Ser Gln Thr Ser		
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[6195]	Cys Ile Thr Asp Glu Phe Gly Tyr Arg Ser Leu Val Ala Lys Asp Thr		
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[6197]	Phe Tyr Phe Glu Gln Asp Arg Lys Ile His Arg Ile Asp Ala Asp Val		
[6198]	770	775	780
[6199]	Asn Ala Ala Ile Asn Ile Ala Arg Arg Phe Leu Thr Arg Tyr Arg Ser		
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[6203]	Asn Val Thr Arg Gln His Glu Arg Ala Tyr Leu Glu Leu Gln Thr Gly		
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[6205]	Ala Pro Ala Ala Thr Leu Asn Pro Thr Ala Glu Ala Ser Tyr Glu Leu		
[6206]	835	840	845
[6207]	Val Gly Leu Ser Pro Glu Glu Glu Glu Leu Ala Gln Thr Arg Ile Lys		
[6208]	850	855	860
[6209]	Arg Lys Lys Arg Glu Pro Phe Tyr Arg His Glu Gly Val Trp Leu Thr		
[6210]	865	870	875
[6211]	Arg Glu Lys His Arg Glu Gln Val His Glu Leu Arg Asn Gln Val Leu		
[6212]	885	890	895
[6213]	Ala Leu Gly Asn Ala Lys Ile Pro Glu Ile Arg Thr Ser Arg Ala Asp		
[6214]	900	905	910
[6215]	Pro Lys Lys Lys Arg Lys Val		

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[6260]	Glu Lys Gly Arg Arg Arg Asn Ile Leu Phe Asp Phe Thr Val Glu Lys
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[6262]	Cys Gly Asp Asn Tyr Leu Phe Arg Tyr Asp Glu Asn Gly Lys Arg Pro
[6263]	305 310 315 320
[6264]	Arg Ala Gly Val Val Lys Glu Pro Arg Phe Cys Trp Arg Arg Lys Gly
[6265]	325 330 335
[6266]	Asn Ser Val Glu Leu Tyr Leu Ala Met Pro Ile Asn Ile Glu Asn Ser
[6267]	340 345 350
[6268]	Met Arg Asn Ile Phe Val Gly Lys Gln Lys Ser Gly Lys His Ser Ala
[6269]	355 360 365
[6270]	Phe Thr Arg Gln Trp Pro Lys Glu Val Glu Gly Leu Asp Glu Leu Arg
[6271]	370 375 380
[6272]	Asp Ala Val Val Leu Gly Val Asp Ile Gly Ile Asn Arg Ala Ala Phe
[6273]	385 390 395 400
[6274]	Cys Ala Ala Leu Lys Thr Ser Arg Phe Glu Asn Gly Leu Pro Ala Asp
[6275]	405 410 415
[6276]	Val Gln Val Met Asp Thr Thr Cys Asp Ala Leu Thr Glu Lys Gly Gln
[6277]	420 425 430
[6278]	Glu Tyr Arg Gln Leu Arg Lys Asp Ala Thr Cys Leu Ala Trp Leu Ile
[6279]	435 440 445
[6280]	Arg Thr Thr Arg Arg Phe Lys Ala Asp Pro Gly Asn Lys His Asn Gln
[6281]	450 455 460
[6282]	Ile Lys Glu Lys Asp Val Glu Arg Phe Asp Ser Ala Asp Gly Ala Tyr
[6283]	465 470 475 480
[6284]	Arg Arg Tyr Met Asp Ala Ile Ala Glu Met Pro Ser Asp Pro Leu Gln
[6285]	485 490 495
[6286]	Val Trp Glu Ala Ala Arg Ile Thr Gly Tyr Gly Glu Trp Ala Lys Glu
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[6288]	Ile Phe Ala Arg Phe Asn His Tyr Lys His Glu His Ala Cys Cys Ala
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[6290]	Val Ser Leu Ser Leu Ser Asp Arg Leu Val Trp Cys Arg Leu Ile Asp
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[6292]	Arg Ile Leu Ser Leu Lys Lys Cys Leu His Phe Gly Gly Tyr Glu Ser
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[6294]	Lys His Arg Lys Gly Phe Cys Lys Ser Leu Tyr Arg Leu Arg His Asn
[6295]	565 570 575
[6296]	Ala Arg Asn Asp Val Arg Lys Lys Leu Ala Arg Phe Ile Val Asp Ala
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[6298]	Ala Val Asp Ala Gly Ala Ser Val Ile Ala Met Glu Lys Leu Pro Ser
[6299]	595 600 605

[6300]	Ser Gly Gly Lys Gln Ser Lys Asp Asp Asn Arg Ile Trp Asp Leu Met
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[6302]	Ala Pro Asn Thr Leu Ala Thr Thr Val Cys Leu Met Ala Lys Val Glu
[6303]	625 630 635 640
[6304]	Gly Ile Gly Phe Val Gln Val Asp Pro Glu Phe Thr Ser Gln Trp Val
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[6306]	Phe Glu Gln Arg Val Ile Gly Asp Arg Glu Gly Arg Ile Val Ser Cys
[6307]	660 665 670
[6308]	Leu Asp Ala Glu Gly Val Arg Arg Asp Tyr Asp Ala Asp Glu Asn Ala
[6309]	675 680 685
[6310]	Ala Lys Asn Ile Ala Trp Leu Ala Leu Thr Arg Glu Ala Glu Pro Phe
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[6312]	Cys Met Ala Phe Glu Lys Arg Asn Gly Val Val Glu Pro Lys Gly Leu
[6313]	705 710 715 720
[6314]	Arg Phe Asp Ile Pro Glu Glu Pro Thr Arg Glu Gln Asp Glu Ser Asp
[6315]	725 730 735
[6316]	Gln Asp Phe Lys Lys Arg Leu Glu Glu Arg Asp Lys Leu Ile Glu Arg
[6317]	740 745 750
[6318]	Leu Gln Ala Lys Ala Asp Arg Met Gln Ala Ile Val Gln Arg Leu Phe
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[6320]	Gly Asp Arg Arg Pro Trp Asp Ala Phe Ala Asp Arg Ile Pro Glu Gly
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[6322]	Lys Ser Lys Arg Leu Phe Arg His Arg Asp Gly Leu Val Leu Asn Lys
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[6324]	Pro Phe Lys Gly Leu Cys Gly Ser Glu Asn Ser Glu Gln Lys Ala Ser
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[6327]	820 825 830
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[6337]	Gln Asp Cys Ile Lys Thr Ile Ser Ala Lys Cys Leu Leu Thr Arg Ala
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[6339]	Gln Ile Asp Glu Leu Arg Ala Lys Tyr Asp Ala Val Leu Asp Thr Met
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[6341]	Arg Pro Leu Ile Arg Leu Ile Leu Ala Gly Tyr Glu Gly Arg Asp Asp

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[6343]	Gly Ile Tyr Glu Glu Ile Ala Pro Glu Met Ser Lys Lys Lys Phe Phe		
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[6345]	Glu Ala Ala Thr Glu Trp Arg Glu Ser Ile Val Lys Asn Ala Ser Pro		
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[6347]	Arg Ala Met Lys Ala Ser Val Phe Gly Asp Lys Glu Pro Cys Lys Ser		
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[6377]	Glu Asn Gly Leu Arg Pro Arg Val Ala Glu Leu Lys Glu Pro Arg Leu		
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[6381]	Val Lys Met His Val Lys Ser Pro Glu Met Phe Ala Gly Asp His Leu		
[6382]		370	375 380
[6383]	Ala Phe Ser Arg Tyr Trp Pro Lys Glu Val Glu Gly Leu Asp Ser Asp		

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[6385]	Thr Lys Ile Thr Ala Leu Gly Val Asp Val Gly Ile Ile Arg Ser Ala			
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[6387]	Tyr Cys Val Ala Val Thr Ala Glu Arg Phe Val Asp Gly Leu Pro Thr			
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[6389]	Glu Met Thr Val Gly Lys Ala Ser Phe Asp Ala Gln Thr Glu Lys Gly			
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[6391]	Arg Glu Tyr Phe Glu Leu Gly Arg Arg Ala Thr Met Leu Gly Trp Leu			
[6392]		450	455	460
[6393]	Ile Lys Thr Thr Arg Arg Tyr Lys Lys Asp Pro Lys Asn Glu His Asn			
[6394]		465	470	475
[6395]	Gln Ile Lys Glu Ser Asp Val Ala Ala Phe Asp Gly Ser Pro Gly Ala			
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[6397]	Phe Glu His Tyr Ile Leu Ala Val Asp Glu Met Ser Asp Asp Pro Leu			
[6398]		500	505	510
[6399]	Asp Val Trp Gly His Ala Asn Ile Thr Gly Tyr Gly Lys Trp Thr Lys			
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[6401]	Gln Ile Phe Lys Glu Phe Asn Gln Leu Lys Arg Glu Arg Ala Glu Gly			
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[6403]	Gln Val Glu Pro Asn Met Thr Asp Asp Leu Thr Trp Cys Ser Leu Ile			
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[6405]	Asp Tyr Ile Ile Ser Leu Lys Lys Thr Leu His Phe Gly Gly Tyr Glu			
[6406]		565	570	575
[6407]	Thr Lys Glu Arg Glu Ser Phe Cys Pro Ala Leu Tyr Asn Glu Arg Ala			
[6408]		580	585	590
[6409]	Asn Cys Arg Asp Val Val Arg Lys Arg Leu Ala Arg Tyr Val Val Glu			
[6410]		595	600	605
[6411]	Arg Ala Ile Ala Ala Glu Ala Gln Val Ile Ser Val Glu Asn Leu Ser			
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[6413]	Lys Cys Arg Arg Asp Asp Lys Arg Lys Asn Arg Val Trp Asp Leu Met			
[6414]		625	630	635
[6415]	Ser Gln Gln Ser Trp Ile Gly Val Leu Thr Asn Met Ala Arg Met Glu			
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[6417]	Asn Ile Ala Val Val Ser Val Asn Pro Asp Leu Thr Ser Gln Trp Val			
[6418]		660	665	670
[6419]	Glu Gln Cys Gly Ala Ile Gly Asp Arg Lys Ala Arg Thr Ile Ala Cys			
[6420]		675	680	685
[6421]	Arg Asp Val Asn Gly Lys Phe Val Ser Leu Asp Ala Asp Leu Asn Ala			
[6422]		690	695	700
[6423]	Ala Tyr Asn Ile Ala Ser Arg Ala Leu Thr Arg His Ala Glu Pro Phe			
[6424]		705	710	715
[6425]	Ser Ile Thr Phe Lys Lys Lys Asp Gly Ile Leu Glu Gln Lys Asp Val			

[6426]		725		730		735
[6427]	Cys Phe Asp Pro Gly Val Ile Pro Val Leu Glu Lys Asn Glu Asn Glu					
[6428]		740		745		750
[6429]	Glu Lys Phe Arg Glu Arg Val Glu Lys Tyr Glu Lys Ser Leu Val Ile					
[6430]		755		760		765
[6431]	Lys Gln Glu Arg Ala Val Arg Trp Arg Ala Ile Leu Gln His Leu Phe					
[6432]		770		775		780
[6433]	Gly Asn Glu Arg Pro Trp Asp Glu Phe Thr Asp Glu Val Lys Glu Gly					
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[6435]	Arg His Val Ser Leu Tyr Arg His His Gly Lys Leu Val Arg Thr Lys					
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[6437]	Gln Tyr Ala Gly Leu Val Lys Glu Ala Asn Asn Glu Leu Val Pro Val					
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[6439]	Cys Ala Val Ala Arg Ser Arg Ala Asp Pro Lys Lys Lys Arg Lys Val					
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[6450]	Asp Thr Glu Ser Pro Ala Ala Asp Thr Gln Val Arg Val His Trp Leu					
[6451]		20		25		30
[6452]	Ala Ala Ser His Arg Ala Ser Pro Gly Leu Gln Gln Val Lys Glu Met					
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[6816]	Leu Phe Asn Tyr Phe Thr Ser Val Ala Ser Gly Ile Lys Asp Lys Val	
[6817]		50 55 60
[6818]	Tyr Asn Leu Gln Ser Asp Glu Lys Thr Ala Pro Ile Phe Asn Asp Tyr	
[6819]		65 70 75 80
[6820]	Val Lys Gln Pro Gln Arg Gly Arg Ser Ala Ala Thr Thr Leu Phe Thr	
[6821]		85 90 95
[6822]	Lys Leu Asp Ala Glu Lys Thr Tyr Thr Ser Gln His Ser Phe Pro Gly	
[6823]		100 105 110
[6824]	Lys Trp Arg Asp Ser Gly Ile Phe Pro Leu Tyr Asn Lys Glu Ser Glu	
[6825]		115 120 125
[6826]	Lys Tyr Asp Leu Ser Thr His Gly Tyr His Tyr Ser Ala Asn Ala Glu	
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[6828]	Ile His Thr Gln Leu Asp Ser His Asp Glu Cys Asn Lys Glu Cys Glu	
[6829]		145 150 155 160
[6830]	Lys Glu Tyr Ala Ala Leu Arg Asp Glu Val Asn Asn Tyr Lys Tyr Glu	
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[6832]	Phe Thr Leu Gln Phe Lys Ala Glu Asn Ala Glu Lys Phe Tyr Asn Phe	
[6833]		180 185 190
[6834]	Val Glu Lys Leu Thr Leu Met Gly Trp Arg Tyr Asp Ala Thr Phe Arg	
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[6836]	Ser Phe Phe Glu Leu His Met His Pro Lys Leu Lys Thr Gly Glu Thr	
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[6838]	Thr Tyr Arg Ala Thr Tyr Lys Leu Pro Ser Gly Lys Ser Lys Arg Tyr	
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[6843]		260 265 270
[6844]	Asn Asn Leu Leu Ser Arg Lys Lys Asp Lys Ala Asn Tyr Ser Ser Thr	
[6845]		275 280 285

[6846]	Ser Leu Ile Lys Ser Gln Ile Arg Leu Tyr Leu Gly Asn Asn Gly Val
[6847]	290 295 300
[6848]	Pro Phe Thr Ala Arg Glu His Asp Gly Arg Ile Tyr Phe Ser Phe Arg
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[6850]	Leu Pro Ala Ile Asn Gly Glu Lys Gly Arg Met Val Glu Ile Pro Cys
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[6852]	Ser Tyr Lys Lys Val Phe Asn Gly Lys Ala Arg Lys Ser Cys Tyr Leu
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[6854]	Gly Gly Leu Thr Ile Glu Lys Thr Asp Ala Gly Lys His Ile Phe Lys
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[6856]	Tyr Ser Val Asn Asn Lys Lys Pro Gln Val Ala Glu Leu Asn Glu Cys
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[6858]	Phe Leu Arg Leu Val Val Arg Asn Arg Glu Tyr Phe Asn Asn Val Val
[6859]	385 390 395 400
[6860]	Ala Gly Lys Ile Thr Asp Ile Asn Thr Asp His Phe Asp Phe Tyr Val
[6861]	405 410 415
[6862]	Asp Leu Pro Leu Asn Val Lys Glu Asp Pro Ile His Asp Leu Ser Ser
[6863]	420 425 430
[6864]	Thr Glu Val Phe Gly Lys Asn Gly Leu Arg Ser Tyr Tyr Ser Ser Ala
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[6866]	Tyr Pro Glu Ile Lys Asn Leu Gly Ser Gln Ile Glu Thr Gly Lys Asn
[6867]	450 455 460
[6868]	Leu Thr Cys Pro Ile Thr Lys Thr His Asn Ile Met Gly Ile Asp Leu
[6869]	465 470 475 480
[6870]	Gly Gln Arg Asn Pro Phe Ala Tyr Cys Ile Lys Asp Asn Thr Gly Lys
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[6872]	Leu Ile Ala Gln Gly His Met Asp Gly Ser Lys Asn Glu Thr Tyr Lys
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[6874]	Lys Tyr Ile Asn Phe Gly Lys Glu Ser Thr Ser Val Ser His Leu Ile
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[6876]	Lys Glu Thr Arg Ser Tyr Leu His Gly Asp Pro Glu Ala Ile Ser Lys
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[6878]	Glu Leu Tyr Asn Glu Val Ala Gly Phe Cys Asn Asn Pro Val Ser Tyr
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[6884]	Asn Trp Ile Gly Arg Asp Trp Leu Trp Tyr Ile Ser Lys Gln Tyr Lys
[6885]	595 600 605
[6886]	Lys His Asn Glu Asn Arg Met Gln Asp Ala Asp Trp Arg Gln Thr Leu
[6887]	610 615 620

[6888]	Tyr Trp Ile Asp Ser Leu Tyr Arg Tyr Ile Asp Val Met Lys Ser Phe
[6889]	625 630 635 640
[6890]	His Asn Phe Gly Ser Phe Tyr Asp Lys Asn Leu Lys Lys Lys Val Asn
[6891]	645 650 655
[6892]	Gly Thr Val Val Gly Phe Cys Lys Thr Val His Asp Gln Ile Asn Asn
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[6902]	Ser Phe Asn Val Ala Leu Ile Glu Ile Asp Glu Arg Asn Thr Ser Gln
[6903]	740 745 750
[6904]	Val Cys Lys Glu Asn Trp Ser Tyr Arg Glu Ala Asp Asp Leu Tyr Tyr
[6905]	755 760 765
[6906]	Val Thr Asp Gly Glu Ser His Lys Val His Ala Asp Glu Asn Ala Ala
[6907]	770 775 780
[6908]	Asn Asn Ile Val Asp Arg Cys Ile Ser Arg His Thr Asn Met Phe Ser
[6909]	785 790 795 800
[6910]	Leu His Met Val Asn Pro Lys Asp Asp Tyr Tyr Val Pro Thr Cys Ile
[6911]	805 810 815
[6912]	Trp Asp Thr Thr Glu Glu Ser Gly Lys Arg Val Arg Gly Phe Leu Thr
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[6914]	Lys Leu Tyr Lys Asn Ser Asp Val Val Phe Thr Lys Lys Gly Asp Lys
[6915]	835 840 845
[6916]	Leu Val Lys Ser Lys Thr Ser Val Lys Glu Leu Lys Lys Leu Val Gly
[6917]	850 855 860
[6918]	Lys Thr Lys Glu Lys Arg Gly Gln Tyr Trp Tyr Arg Phe Glu Gly Lys
[6919]	865 870 875 880
[6920]	Ser Trp Ile Asn Glu Ala Asp Arg Asp Thr Ile Ile Leu Asn Ala Lys
[6921]	885 890 895
[6922]	Lys Ile Ser Arg Glu Arg Asp Asn Gly Glu Gln Ser Thr Asp Thr Arg
[6923]	900 905 910
[6924]	Ser Gln Asn Val Thr Val Ser Val Leu Asp Val Cys Glu Thr Ala Glu
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[6926]	Lys Lys Lys Leu Val Leu Val
[6927]	930 935

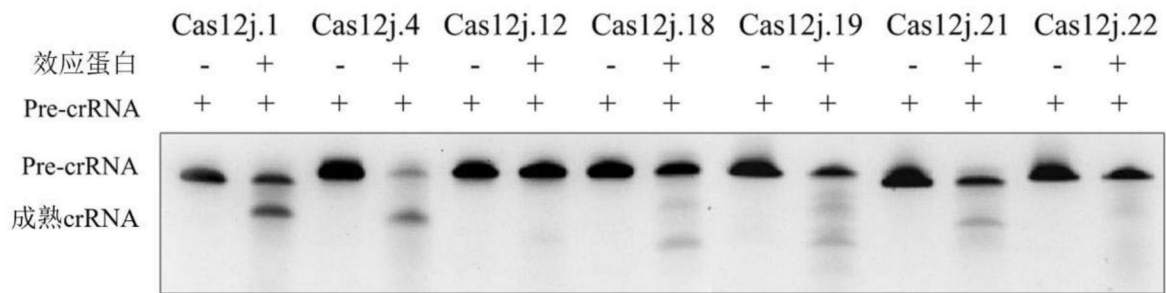


图1

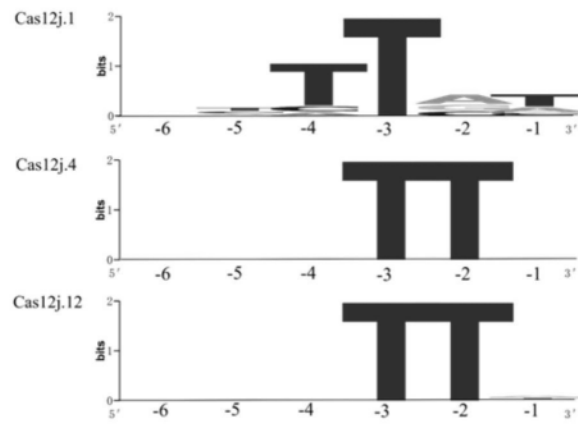


图2A

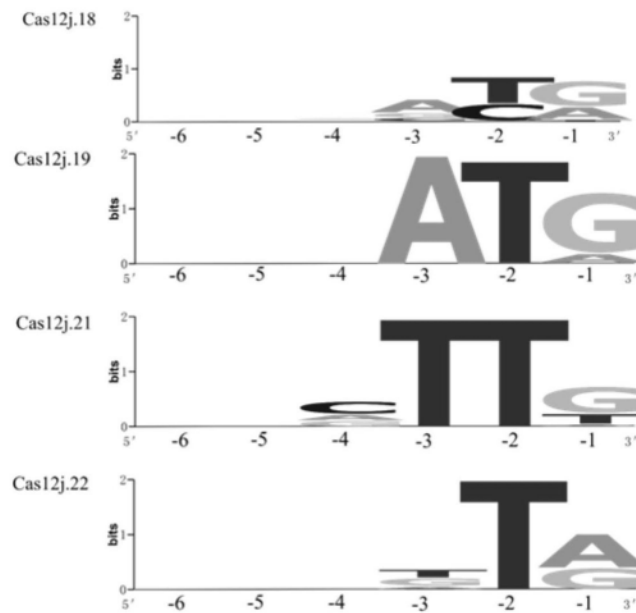


图2B

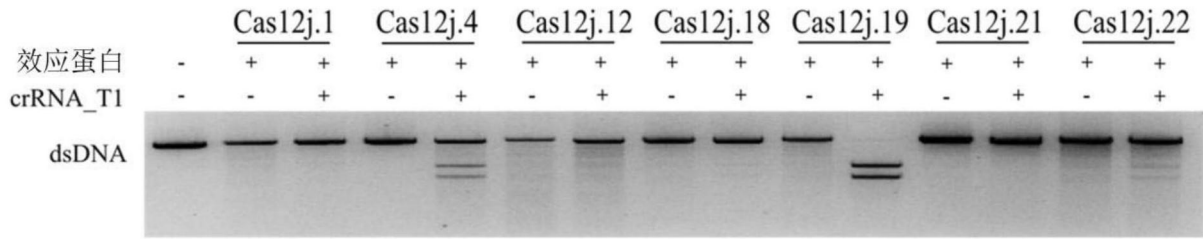


图3

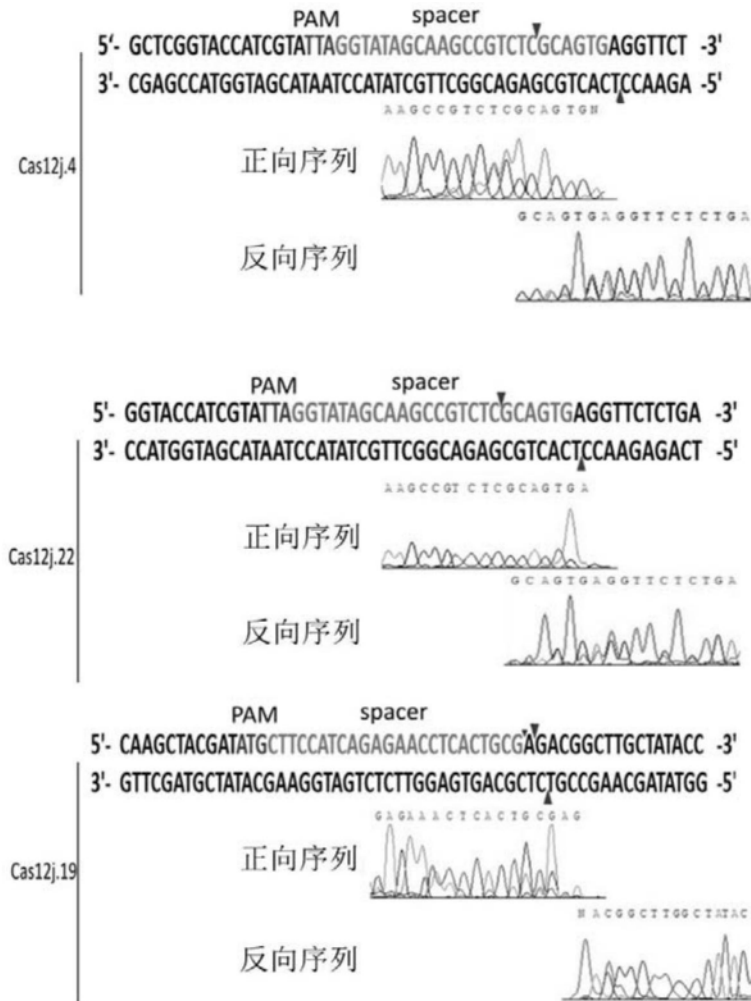


图4

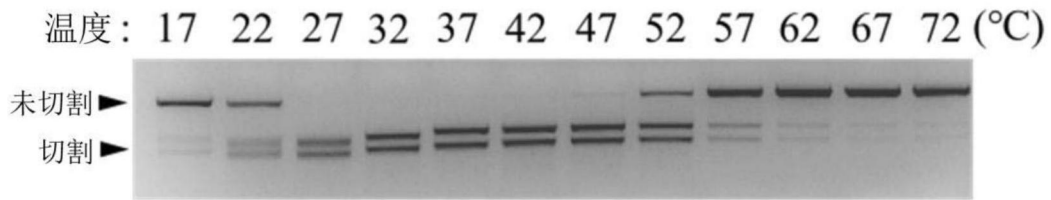


图5

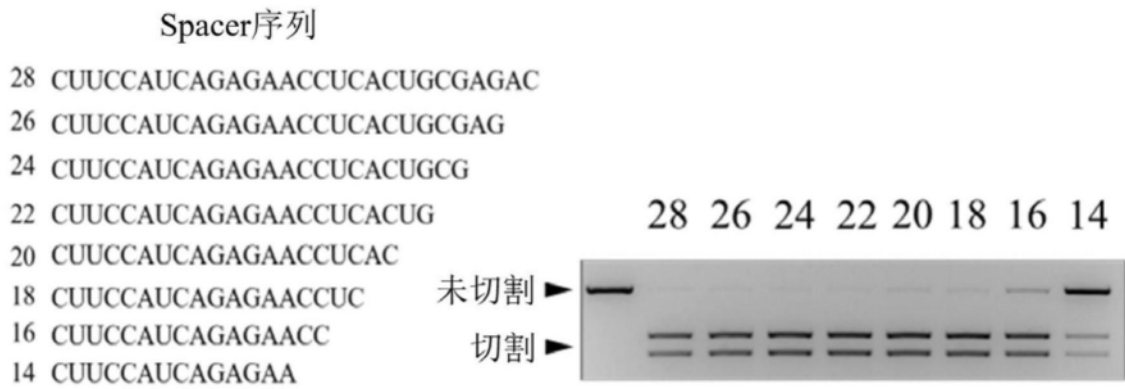


图6

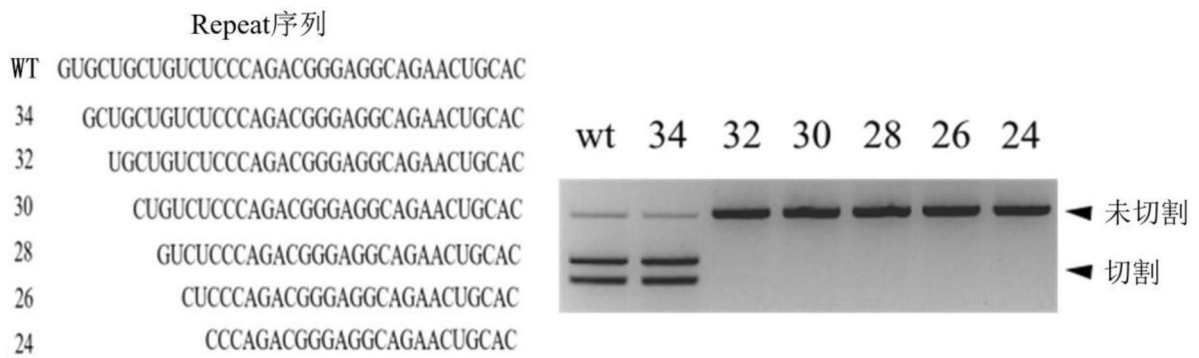


图7

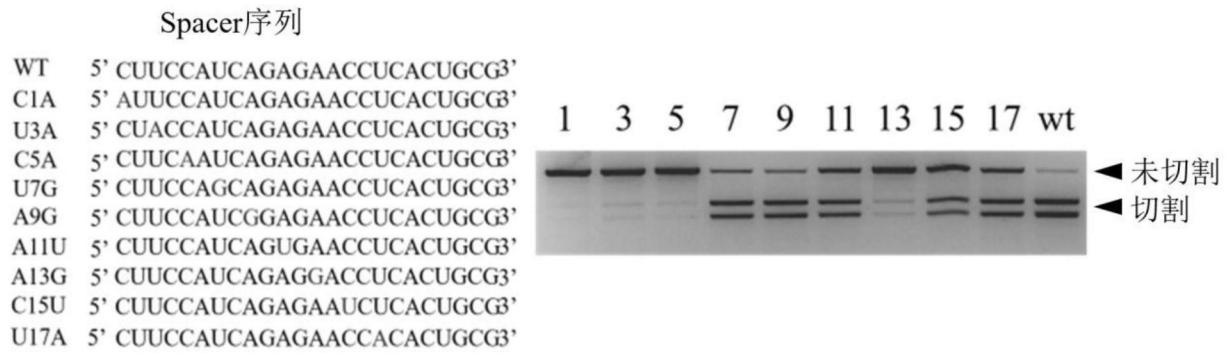


图8