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(54) 发明名称

来那度胺或泊利度胺与经抗CD38抗体灭毒
的干扰素 α 构筑体的组合, 和其用途

(57) 摘要

癌症治疗方法, 其包括向癌症患者投与经抗
CD38抗体灭毒的人类IFN α -2b构筑体和来那度
胺(lenalidomide)或泊利度胺(pomalidomide)。
可根据这些方法治疗的肿瘤包括有包含表达CD-
38的肿瘤细胞的肿瘤, 包括B细胞淋巴瘤、多发性
骨髓瘤、非霍奇金淋巴瘤(non-Hodgkin's
lymphoma)、慢性骨髓性白血病、慢性淋巴细胞性
白血病和急性淋巴细胞白血病。

1. (i) 包括特异地结合到CD38, 包含如SEQ ID NO:27所示的重链可变区氨基酸序列和如SEQ ID NO:29所示的轻链可变区氨基酸序列, 并且融合到经灭毒的干扰素 α -2b的抗体的构筑体和(ii) 来那度胺(lenalidomide)或泊利度胺(pomalidomide)在制备通过同步地或依序地实质上同时授予(i)和(ii)用于治疗B细胞淋巴瘤、多发性骨髓瘤、早期多发性骨髓瘤、前多发性骨髓瘤、瓦尔登斯特伦氏巨球蛋白血症(Waldenström'smacroglobulinemia)、非霍奇金淋巴瘤(non-Hodgkin'slymphoma)、慢性骨髓性白血病、慢性淋巴细胞性白血病或急性淋巴细胞白血病中的任一种的药物组合中的用途。

2. 根据权利要求1所述的用途, 其中所述抗体包含人类IgG4恒定区。

3. 根据权利要求2所述的用途, 其中根据EU编号系统, 所述人类IgG4恒定区包含位置228处的脯氨酸。

4. 根据权利要求3所述的用途, 其中根据EU编号系统, 所述人类IgG4恒定区进一步包含所述恒定区的位置252处的酪氨酸、位置254处的苏氨酸和位置256处的谷氨酸。

5. 根据权利要求1所述的用途, 其中所述抗体包含人类IgG1恒定区。

6. 根据权利要求5所述的用途, 其中根据EU编号系统, 所述人类IgG1恒定区包含所述恒定区的位置252处的酪氨酸、位置254处的苏氨酸和位置256处的谷氨酸。

7. 根据权利要求1所述的用途, 其中所述经灭毒的干扰素 α -2b包含SEQ ID NO:3、SEQ ID NO:5、SEQ ID NO:6、SEQ ID NO:7、SEQ ID NO:212或SEQ ID NO:213的氨基酸序列。

8. 根据权利要求7所述的用途, 其中所述经灭毒的干扰素 α -2b包含SEQ ID NO:212或SEQ ID NO:213的氨基酸序列。

9. 根据权利要求7所述的用途, 其中所述经灭毒的干扰素 α -2b包含SEQ ID NO:212的氨基酸序列。

10. 根据权利要求1所述的用途, 其中所述构筑体包含抗体, 所述抗体包含有包含SEQ ID NO:27的氨基酸序列的重链可变区、包含SEQ ID NO:29的氨基酸序列的轻链可变区和人类IgG4恒定区, 并且所述经灭毒的干扰素 α -2b包含SEQ ID NO:212的氨基酸序列。

11. 根据权利要求10所述的用途, 其中根据EU编号系统, 所述人类IgG4恒定区进一步包含所述恒定区的位置252处的酪氨酸、位置254处的苏氨酸和位置256处的谷氨酸。

12. 根据权利要求1到11中任一权利要求所述的用途, 其中所述药物组合包含来那度胺和所述构筑体, 其用于治疗B细胞淋巴瘤、多发性骨髓瘤、早期多发性骨髓瘤、前多发性骨髓瘤、瓦尔登斯特伦氏巨球蛋白血症、非霍奇金淋巴瘤、慢性骨髓性白血病、慢性淋巴细胞性白血病或急性淋巴细胞白血病中的任一种。

13. 根据权利要求1到11中任一权利要求所述的用途, 其中所述药物组合包含泊利度胺和所述构筑体, 其用于治疗B细胞淋巴瘤、多发性骨髓瘤、早期多发性骨髓瘤、前多发性骨髓瘤、瓦尔登斯特伦氏巨球蛋白血症、非霍奇金淋巴瘤、慢性骨髓性白血病、慢性淋巴细胞性白血病或急性淋巴细胞白血病中的任一种。

来那度胺或泊利度胺与经抗CD38抗体灭毒的干扰素 α 构筑体的组合,和其用途

[0001] 相关申请案的交叉引用

[0002] 本申请案要求2014年5月1日申请的美国临时申请案第61/986,913号的优先权,其以全文引用的方式并入本文中并且用于所有目的。

[0003] 对序列表的参考

[0004] 本申请案包括在2015年5月1日创建的以文本文件SEQ_LST_Lenalidomide_ST25.TXT形式电子提交的序列表,其尺寸为284,000字节。序列表以引用的方式并入本文中。

技术领域

[0005] 本发明大体上涉及癌症治疗领域。更确切地说,本发明涉及癌症疗法,其协同组合来那度胺或泊利度胺与经抗CD38抗体灭毒的干扰素 α -2b构筑体。所述组合疗法与由单独的来那度胺、泊利度胺或构筑体的投药呈现的肿瘤生长抑制或延缓相比实质上增强肿瘤生长抑制或延缓。另外,所述组合疗法可克服来那度胺抗性或泊利度胺抗性。

背景技术

[0006] 在整个本说明书中引用许多公开案,包括专利案、公开的专利申请案、技术论文、学术论文以及基因或蛋白质寄存编号。这些材料中的每一个皆以全文引用的方式并入本文中并且用于所有目的。

[0007] CD38是46kDa II型跨膜糖蛋白,其涉及跨膜信号传导和细胞粘着。其也称为环状ADP核糖水解酶,因为其可视细胞外pH值而将 NAD^+ 和 NADP^+ 转型成cADPR、ADPR和NAADP。这些产物诱导细胞内的 Ca^{2+} 移动,其可引起酪氨酸磷酸化和细胞活化。CD38还是可与配位体CD31相互作用的受体。通过CD31进行的受体活化引起细胞内事件,包括 Ca^{2+} 移动、细胞活化、增殖、分化和迁移。

[0008] CD38在多发性骨髓瘤细胞,在大多数情况下,T-和B-谱系急性成淋巴细胞白血病(ALL)、一些急性骨髓细胞性白血病、滤泡中心细胞淋巴瘤和T成淋巴细胞淋巴瘤的表面大量表达。CD38还在B-谱系慢性淋巴母细胞白血病(B-CLL)细胞上表达。在一些情况下,呈现CD38+纯系的B-CLL患者的特征是具有不利的临床病程以及疾病的更晚期状态、对化学疗法的不良反应和更短的存活时间。

[0009] 干扰素并且尤其IFN- α 能够增加细胞凋亡并且减少某些癌细胞的增殖。IFN- α 已由FDA批准用于治疗若干癌症,包括黑素瘤、肾细胞癌、B细胞淋巴瘤、多发性骨髓瘤、慢性骨髓性白血病(CML)和毛状细胞白血病。IFN- α 对肿瘤细胞的直接作用是通过IFN- α 直接结合到这些细胞上的I型IFN受体并且刺激细胞凋亡、末端分化和/或降低增殖来介导。此外,IFN- α 对非癌细胞的间接作用包括IFN- α 刺激免疫系统的能力,其可通过引起免疫系统排斥肿瘤来产生额外的抗癌作用。IFN- α 还呈现抑制肿瘤血管生成的能力,并且因此可通过代谢饥饿来抑制肿瘤生长。

[0010] IFN- α 的直接抗肿瘤活性是由癌细胞表面上I型干扰素受体介导,所述受体在受刺激时起始多种信号转导路径,引起减少的增殖和/或诱导末端分化或细胞凋亡。然而,I型干扰素受体也存在于大部分非癌性细胞上。非癌性细胞上由IFN- α 引起的I型受体的活化引起大量促炎性细胞因子和趋化因子的表达,引起不合需要的全身性毒性。这类毒性可引起严重的流感类症状,其阻止投与个体的IFN- α 对癌细胞发挥最大抗增殖性和促凋亡活性。

[0011] 通常,IFN可靶向癌细胞,例如通过使其与靶向抗体或其靶向片段连接。尽管这种方法可引起针对癌细胞的IFN的活性增加,但其并未完全解决IFN对健康细胞的不合需要的活性问题。使IFN- α 与IgG的重链的C端融合可例如延长IFN α 的半衰期,其可延长不合需要的不良事件。因此,需要改良干扰素的全身性毒性概况,同时保留其一种或多种抗肿瘤作用。

[0012] 来那度胺和泊利度胺皆为小分子免疫调节剂,以及抗多发性骨髓瘤药物撒利多胺(thalidomide)的衍生物。来那度胺和泊利度胺皆用于治疗 and 维持某些癌症,包括多发性骨髓瘤和淋巴瘤。在许多情况下,最初对来那度胺或泊利度胺具有敏感性的肿瘤变得对这些药剂具有抗性或耐性。在其它情况下,肿瘤不对来那度胺或泊利度胺疗法起反应。在本领域中,需要克服来那度胺或泊利度胺抗性 or 增强来那度胺或泊利度胺活性,并且潜在地提供疗法,从而使非反应性患者可对来那度胺或泊利度胺疗法起反应。

发明内容

[0013] 本发明披露用于治疗肿瘤的方法。所述方法可包含向具有肿瘤的个体投与可有效治疗肿瘤的量的经抗CD38抗体灭毒的IFN α -2b构筑体和可有效治疗肿瘤的量的来那度胺。所述方法可包含向具有肿瘤的个体投与可有效治疗肿瘤的量的经抗CD38抗体灭毒的IFN α -2b构筑体和可有效治疗肿瘤的量的泊利度胺。构筑体可增强来那度胺的抗肿瘤活性或可增强泊利度胺的抗肿瘤活性,和/或来那度胺或泊利度胺可增强构筑体的抗肿瘤活性。有效量优选是当与单独投与来那度胺或泊利度胺或构筑体之后的肿瘤生长相比时,两种药剂协同作用以实质上抑制和/或延缓肿瘤生长的量。投药可消除现有肿瘤,和/或抑制肿瘤重新产生。个体可以是任何哺乳动物,优选是灵长类动物,并且最优选是人类。优选的是,构筑体的量和来那度胺或泊利度胺的量足以使构筑体和来那度胺或泊利度胺在其治疗作用方面发挥协同作用。构筑体和来那度胺或泊利度胺中的每一个可包含于组合物中,所述组合物包含医药学上可接受的载剂,但构筑体和来那度胺或泊利度胺可包含在单独的组合物中。构筑体和来那度胺或泊利度胺可实质上同时投与,或可依序投与。投药可静脉内(例如构筑体)或经口(例如来那度胺或泊利度胺)进行,并且可按照医疗人员的指导进行。相信构筑体在循环中的留存时间长于来那度胺或泊利度胺,使得治疗疗程可包含与构筑体的投药相比更频繁地投与来那度胺或泊利度胺。根据这类方法,构筑体可包含本文中描述或例示的任何抗CD38抗体和任何经灭毒的干扰素 α -2b分子。

[0014] 肿瘤将包含表达CD-38的肿瘤细胞。肿瘤可包含B细胞淋巴瘤、多发性骨髓瘤、早期多发性骨髓瘤、前多发性骨髓瘤、瓦尔登斯特伦氏巨球蛋白血症(Waldenström's macroglobulinemia)、非霍奇金淋巴瘤(non-Hodgkin's lymphoma)、慢性骨髓性白血病、慢性淋巴细胞性白血病或急性淋巴细胞白血病。任何这类肿瘤可对单独的来那度胺或泊利度胺具有敏感性或对单独的来那度胺或泊利度胺具有抗性,使得组合疗法对个体产生治疗效益。尤其优选多发性骨髓瘤。本发明还提供经抗CD38抗体灭毒的IFN α -2b构筑体和来那度胺

或泊利度胺作为组合疗法在治疗以下疾病中的用途：B细胞淋巴瘤、多发性骨髓瘤、早期多发性骨髓瘤、前多发性骨髓瘤、瓦尔登斯特伦氏巨球蛋白血症、非霍奇金淋巴瘤、慢性髓性白血病、慢性淋巴细胞性白血病或急性淋巴细胞白血病。

[0015] 经抗CD38抗体灭毒的IFN α -2b构筑体优选是融合蛋白，其包含有包含重链和轻链的抗CD38抗体部分，和经灭毒的IFN α -2b部分，优选其中抗CD38抗体重链的C端与经灭毒的IFN α -2b的N端通过肽键直接融合。在一些方面中，抗CD38抗体重链的C端与经灭毒的IFN α -2b的N端通过具有五个或更多个氨基酸的连接肽融合，并且因此，构筑体进一步包含连接肽。

[0016] 构筑体的抗CD38抗体部分可包含有包含SEQ ID NO:17的氨基酸序列的重链可变区和包含SEQ ID NO:21的氨基酸序列的轻链可变区，任选的限制条件为SEQ ID NO:17不包括SEQ ID NO:24的氨基酸序列并且SEQ ID NO:21不包括SEQ ID NO:25的氨基酸序列。重链可变区和轻链可变区配对可选自本发明的表1-4中的任一个中阐述的配对。

[0017] 在一些方面中，构筑体的抗CD38抗体部分包含有包含SEQ ID NO:18的氨基酸序列的重链可变区和包含SEQ ID NO:22的氨基酸序列的轻链可变区。抗CD38抗体可包含有包含SEQ ID NO:19的氨基酸序列的重链可变区和包含SEQ ID NO:21的氨基酸序列的轻链可变区，任选的限制条件为SEQ ID NO:21不包括SEQ ID NO:25的氨基酸序列。在一些方面中，构筑体的抗CD38抗体部分包含有包含SEQ ID NO:20的氨基酸序列的重链可变区和包含SEQ ID NO:23的氨基酸序列的轻链可变区。

[0018] 构筑体的抗CD38抗体部分可包含有包含SEQ ID NO:26、SEQ ID NO:27或SEQ ID NO:28的氨基酸序列的重链可变区。构筑体的抗CD38抗体部分可包含有包含SEQ ID NO:29或SEQ ID NO:30的氨基酸序列的轻链可变区。SEQ ID NO:26、27或28中的任一个可与SEQ ID NO:29或30中的任一个配对。在尤其优选方面中，构筑体的抗CD38抗体部分可包含有包含SEQ ID NO:27的氨基酸序列的重链可变区和包含SEQ ID NO:29的氨基酸序列的轻链可变区。

[0019] 在一些方面中，经抗CD38抗体灭毒的干扰素 α -2b构筑体包含经抗CD38抗体重链灭毒的糖基化干扰素 α -2b融合多肽，其包含SEQ ID NO:216的氨基酸序列，和抗CD38抗体轻链，其包含有包含SEQ ID NO:29的氨基酸序列的可变区。在一些方面中，轻链具有SEQ ID NO:217的氨基酸序列（可变区和恒定区）。

[0020] 构筑体的抗CD38抗体部分可包含人类IgG1恒定区。在一些优选方面中，构筑体的抗CD38抗体部分可包含人类IgG4恒定区。优选的是，抗体包含IgG4恒定区或IgG1恒定区，其经工程改造以消除FcR结合，从而避免抗体介导的效应功能，相信这可在避免非特异性Fc受体介导的抗体结合和对非抗体靶向细胞的后续IFN介导的毒性方面具有优势。

[0021] 根据EU编号系统，人类IgG1恒定区可任选地包含位置252处的酪氨酸、位置254处的苏氨酸和位置256处的谷氨酸。根据EU编号系统，人类IgG4恒定区可任选地包含位置228处的脯氨酸，并且根据EU编号系统，任选地进一步包含位置252处的酪氨酸、位置254处的苏氨酸和位置256处的谷氨酸。构筑体的抗CD38抗体部分可包含Fab。

[0022] 构筑体的经灭毒的干扰素 α -2b部分可以是经灭毒的人类干扰素 α -2b。构筑体的经灭毒的干扰素 α -2b部分可包含SEQ ID NO:3、SEQ ID NO:5、SEQ ID NO:6、SEQ ID NO:7、SEQ ID NO:8、SEQ ID NO:211、SEQ ID NO:212、SEQ ID NO:213、SEQ ID NO:214或SEQ ID NO:

215中的任一个的氨基酸序列。构筑体的经灭毒的干扰素 α -2b部分可包括23个氨基酸N端截短(SEQ ID NO:4)。构筑体的经灭毒的干扰素 α -2b部分优选包括具有A145D取代(SEQ ID NO:5)或A145G取代(SEQ ID NO:7)的23个氨基酸N端截短。构筑体的经灭毒的干扰素 α -2b部分可糖基化,例如在位置106处具有氨基酸缺失或取代(其优选是T106A取代)的经截短(23个氨基酸N端截短)的人类干扰素 α -2b,但可包含其它适合的取代以移除糖基化位点(SEQ ID NO:214)。在一些优选方面中,构筑体的经灭毒的干扰素 α -2b部分包括T106A取代和A145D取代(SEQ ID NO:212)或A145G取代(SEQ ID NO:213)。在一些方面中,构筑体的经灭毒的干扰素 α -2b部分包括T106的缺失(SEQ ID NO:215)。

[0023] 在所述方法的尤其优选方面中,所述方法用于治疗人类个体中的多发性骨髓瘤。在一些方面中,所述方法包含向个体投与来那度胺和经抗CD38抗体灭毒的IFN α -2b构筑体,其包含有包含SEQ ID NO:27的氨基酸序列的重链可变区和包含SEQ ID NO:29的氨基酸序列的轻链可变区,以及IgG4恒定区,并且包含有包含SEQ ID NO:212或SEQ ID NO:213的氨基酸序列的经灭毒的IFN α -2b分子。在一些方面中,所述方法包含向个体投与泊利度胺和经抗CD38抗体灭毒的IFN α -2b构筑体,其包含有包含SEQ ID NO:27的氨基酸序列的重链可变区和包含SEQ ID NO:29的氨基酸序列的轻链可变区,以及IgG4恒定区,并且包含有包含SEQ ID NO:212或SEQ ID NO:213的氨基酸序列的经灭毒的IFN α -2b分子。

[0024] 还提供来那度胺或泊利度胺与经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合,其用于治疗B细胞淋巴瘤、多发性骨髓瘤、早期多发性骨髓瘤、前多发性骨髓瘤、瓦尔登斯特伦氏巨球蛋白血症、非霍奇金淋巴瘤、慢性骨髓性白血病、慢性淋巴细胞性白血病或急性淋巴细胞白血病中的任一种。还提供来那度胺与经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合,其用于治疗B细胞淋巴瘤、多发性骨髓瘤、早期多发性骨髓瘤、前多发性骨髓瘤、瓦尔登斯特伦氏巨球蛋白血症、非霍奇金淋巴瘤、慢性骨髓性白血病、慢性淋巴细胞性白血病或急性淋巴细胞白血病中的任一种。还提供泊利度胺与经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合,其用于治疗B细胞淋巴瘤、多发性骨髓瘤、早期多发性骨髓瘤、前多发性骨髓瘤、瓦尔登斯特伦氏巨球蛋白血症、非霍奇金淋巴瘤、慢性骨髓性白血病、慢性淋巴细胞性白血病或急性淋巴细胞白血病中的任一种。

附图说明

[0025] 图1展示在用媒介对照物、自由的未经灭毒的干扰素 α 2b (IFN- α)、包括与单独经灭毒的干扰素 α 2b (145D) 融合的抗CD38抗体的构筑体、单独来那度胺、自由的未经灭毒的干扰素- α 和来那度胺的组合或经抗CD38灭毒的干扰素 α 融合构筑体与来那度胺的组合治疗之后,具有多发性骨髓瘤异种移植的SCID小鼠中呈时间函数形式的中位肿瘤体积。以产生次最大肿瘤抑制的剂量投与经抗CD38抗体灭毒的干扰素 α 融合构筑体。以0.5mg/kg的剂量投与野生型干扰素,其与作为经抗CD38灭毒的干扰素 α 2b构筑体的组分投与的干扰素的量在莫耳量方面等效。每天以25mg/kg通过腹膜内注射投与来那度胺持续21天。

[0026] 图2展示在用媒介对照物、针对与经灭毒的干扰素 α (145D) 融合的同型匹配抗体(与来自图1的抗CD38抗体相同的同型)的构筑体或经同型匹配抗体灭毒的干扰素 α 融合构筑体与来那度胺的组合治疗之后,具有多发性骨髓瘤异种移植的SCID小鼠中呈时间函数形式的肿瘤体积。尽管单独的来那度胺或与无关融合构筑体的组合可延缓快速肿瘤

生长的发生,但这些药剂或药剂组合皆不能阻止肿瘤生长。

[0027] 图3A-3J展示在用媒介剂对照物、持续21天每天以25mg/kg通过腹膜内注射投与的单独的来那度胺、以用于次最大肿瘤抑制的剂量或给药频率投与的与经灭毒的糖基化人类干扰素- α 2b (T106A) 融合的抗CD38抗体 (A10.21) 或以如表5中定义的用于次最大肿瘤抑制的剂量或给药频率投与的来那度胺与抗CD38抗体 (与经灭毒的糖基化干扰素融合) 的多种组合治疗之后,具有多发性骨髓瘤异种移植NCI-H929的个别SCID小鼠中呈时间函数形式的肿瘤体积。

[0028] 图4展示植入人类骨髓瘤细胞系NCI-H929的SCID小鼠中,与经灭毒的糖基化干扰素- α 2b融合的抗CD38抗体和来那度胺的次最佳剂量或给药间隔的组合对存活期的作用(卡普兰-迈耶图(Kaplan-Meier graph))。

[0029] 图5展示在用媒介剂对照物、包括与经灭毒的干扰素 α 2b (145D) 融合的抗CD38抗体的构筑体、单独的泊利度胺、干扰素- α 与泊利度胺的组合或经抗CD38灭毒的干扰素 α 融合构筑体与泊利度胺的组合治疗之后,具有多发性骨髓瘤异种移植的SCID小鼠中呈时间函数形式的中位肿瘤体积。用单独的经抗CD-38灭毒的IFN α 2b进行治疗引起肿瘤的的稳定收缩,其在研究期间是稳定的,但用单独的构筑体治疗的动物在治疗期间,在10只小鼠中的7只中显示一些肿瘤再生。泊利度胺与经抗CD38灭毒的IFN α 2b的组合也能够使肿瘤收缩,但实质上更少的小鼠(10只小鼠中的4只)在治疗期间显示肿瘤再生。

具体实施方式

[0030] 在整个本说明书和权利要求中使用许多与披露方面相关的术语。除非另有指示,否则这类术语具有其在所属领域中的一般含义。其它具体定义的术语将以符合本文中提供的定义的方式解释。

[0031] 术语个体与患者可互换使用并且包括任何哺乳动物,包括伴侣和农耕哺乳动物,以及啮齿动物,包括小鼠、兔和大鼠,以及其它啮齿动物。更优选是非人类灵长类动物,如食蟹猕猴(*Cynomolgus monkeys*),并且尤其优选是人类。

[0032] 如果分子(如抗体)由于人类介入而从其天然环境改变和/或移出,那么其为“经分离”的。

[0033] 如本文中所使用,除非以其它方式明确陈述,否则单数形式“一”和“所述”包括多个指示物。

[0034] 如本文中所使用,在本文中所描述的癌症、肿瘤、恶性疾病或前恶性疾病的任何方面中,术语“抗性”指癌症、肿瘤、恶性疾病或前恶性疾病对用来那度胺或泊利度胺进行治疗和/或对用经CD38灭毒的IFN α 2b构筑体进行治疗具有抗性,或未能完全对其起反应或由其消除。抗性可在治疗开始时发生或可在阳性反应时段之后的治疗期间持续。

[0035] “协同作用”或如本文关于来那度胺或泊利度胺与经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合的肿瘤治疗作用(例如协同肿瘤治疗)所使用,包含肿瘤生长抑制,包括肿瘤抑制、肿瘤生长或再生长延缓,和/或现有肿瘤的实质性消除,并且包括在停止治疗之后抑制肿瘤的重新产生,其与单独的来那度胺或泊利度胺或经抗CD38抗体灭毒的干扰素 α -2b构筑体的肿瘤治疗作用相比,或与分离的药剂的加成肿瘤治疗作用相比,在抑制量、程度、范围和/或速率方面显著更大,和/或在抑制重新产生的时间方面显著更长。因此,来那度胺

或泊利度胺的“协同有效量”或经抗CD38抗体灭毒的干扰素 α -2b构筑体的“协同有效量”是产生来那度胺或泊利度胺与经抗CD38抗体灭毒的干扰素 α -2b构筑体的“协同作用”的量,包括两种药机协同作用以实质上抑制、延缓或抑止肿瘤生长、实质上消除现有肿瘤和/或实质上抑制、延缓或抑止肿瘤重新产生的量。

[0036] 经抗CD38抗体灭毒的干扰素 α 2b构筑体包含特异性结合于CD38(其与经灭毒的干扰素(IFN) α -2b接合)的抗体。抗体可通过结合、交联或通过经由连接子或经由抗体与IFN分子之间的肽键融合而接合到IFN α -2b。

[0037] 已根据本发明观察到,经抗CD38抗体灭毒的干扰素 α -2b构筑体可与来那度胺或泊利度胺协同作用以抑制肿瘤生长并且在一些情况下,消除活体内的现有多发性骨髓瘤。这一协同作用优于纯粹的加成作用。举例来说,进一步观察到大部分用这一组合治疗的肿瘤在治疗期间或在治疗停止后并未复发,然而用单独的次最佳剂量的来那度胺或泊利度胺或次最佳剂量的经抗CD38抗体灭毒的IFN α 1pha2b构筑体治疗的肿瘤在治疗期间复发并且在治疗停止后体积继续增长。还观察到这一组合可克服肿瘤对来那度胺的预先存在或诱导的抗性。因此,本发明提供用于癌症治疗并且优选用于多发性骨髓瘤治疗的组合疗法。本发明提供组合疗法系统,其包含经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺,包含经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺的组合物、用于通过向癌症患者投与经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺来癌症的方法以及包含经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺以及使用构筑体和来那度胺或泊利度胺作为用于治疗癌症的方法中的组合疗法的说明的套组。本发明还提供用于通过组合来那度胺治疗与用经抗CD38抗体灭毒的干扰素 α -2b构筑体进行的治疗来增强来那度胺或泊利度胺治疗的抗肿瘤活性的方法。或者或另外,本发明提供通过组合来那度胺或泊利度胺治疗来增强用经抗CD38抗体灭毒的干扰素 α -2b构筑体进行的治疗的方法。本文中所描述之方法可活体外、离体、活体内或现场进行。

[0038] 在一个方面中,本发明提供组合疗法,其包含经抗CD38抗体灭毒的IFN- α 2b构筑体和来那度胺或泊利度胺。经抗CD38抗体灭毒的IFN- α 2b构筑体和来那度胺或泊利度胺优选是有效治疗肿瘤的量。在一些方面中,经抗CD38抗体灭毒的IFN- α 2b构筑体和来那度胺或泊利度胺是用于治疗肿瘤的协同有效量。肿瘤可以是B细胞淋巴瘤、多发性骨髓瘤、早期多发性骨髓瘤、前多发性骨髓瘤、瓦尔登斯特伦氏巨球蛋白血症、非霍奇金淋巴瘤、慢性骨髓性白血病、慢性淋巴细胞性白血病或急性淋巴细胞白血病。在一些方面中,组合疗法包含有包含经抗CD38抗体灭毒的IFN- α 2b构筑体和医药学上可接受的载剂的组合物以及包含来那度胺或泊利度胺和医药学上可接受的载剂的组合物。

[0039] 作为构筑体的一部分,抗CD38抗体可以是单克隆抗体,并且更优选是包含可变区重链和可变区轻链的全长单克隆抗体。在一些方面中,抗CD38抗体可包含抗体的衍生物或片段或部分,其保留CD38结合特异性并且还优选保留亲本抗体分子的大部分或所有所述(例如对于CD38)。举例来说,衍生物可包含至少一个可变区(重链或轻链可变区)。适合的抗体衍生物和片段的其它实例包括(但不限于)具有多表位特异性的抗体、双特异性抗体、多特异性抗体、双功能抗体、单链分子以及FAb、F(Ab')₂、Fd、Fab_c和Fv分子、单链(Sc)抗体、单链Fv抗体(scFv)、个别抗体轻链、个别抗体重链、抗体链与其他分子之间的融合物、重链单体或二聚体、轻链单体或二聚体、由一个重链和一个轻链组成的二聚体和其它多聚体。单链

Fv抗体可以是多价的。所有抗体同型可用于产生抗体衍生物、片段和部分。抗体衍生物、片段和/或部分可以以重组方式产生并且由任何细胞类型、原核或真核表达。

[0040] 对于用于治疗人类,非人类衍生的抗体可在投与人类患者时在结构上变得抗原性较弱,包括通过去免疫、嵌合或人类化或超人类化。在一些方面中,抗体是人类化抗体。人类化抗体是其中氨基酸直接参与抗原结合的抗体,例如互补决定区(CDR),并且在一些情况下,重链和/或轻链的构架区(FR)或其部分不是人类来源,而抗体中氨基酸的其余部分是人类或人类来源,例如人类抗体骨架。人类化抗体还包括其中人类蛋白质的一个或多个残基由一个或多个氨基酸取代修饰和/或人类蛋白质的一个或多个FR残基由相应的非人类残基置换的抗体。人类化抗体还可以包含既不在人类抗体中也不在非人类抗体中发现的残基。人类化抗体可以是超人类化抗体,例如美国专利第7,732,578号中所描述。抗体可以是人类化嵌合抗体。人类化抗体还包括具有恒定区序列(例如可变区构架序列)的抗体,所述序列是基于多个人类抗体的人造共同序列。

[0041] 在尤其优选方面中,抗CD38抗体是完全人类。完全人类抗体是其中完全分子是人类或人类来源的抗体,或包括与人类抗体序列一致或实质上一致的氨基酸序列。完全人类抗体包括从人类V基因文库获得的抗体,举例来说,其中编码抗体的可变区的人类基因以重组方式表达。完全人类抗体可用编码人类抗体的基因转型的其它生物体(例如小鼠和转基因鼠(xenomouse))或来自其它生物体的细胞中表达。然而,完全人类抗体可包括不由人类序列编码的氨基酸残基,例如通过随机或定点突变引入的突变。

[0042] 抗CD38抗体可以是任何类别的全长抗体,例如IgG1、IgG2或IgG4。在特定实施例中,抗CD38抗体是全长IgG4抗体。这类抗体的恒定结构域优选是人类。这类抗体的可变区可以是非人类来源,或优选是来源人类或经人类化。还可以使用抗体片段代替全长抗体。

[0043] 在一些方面中,抗CD38抗体可包含非免疫球蛋白衍生的蛋白质构架。举例来说,可参考(库(Ku)和舒茨(Schutz),美国国家科学院院刊(Proc.Natl.Acad.Sci.USA) 92:6552-6556,1995),其描述具有两个经随机化以产生CDR的环的四螺旋束蛋白质细胞色素b562,其已选择用于抗原结合。

[0044] 天然序列变异可在重链和轻链和编码其的基因中存在,并且因此,具有所属领域中一般技术的人员将期望在本文中描述和例示的抗体的氨基酸序列或编码其的基因内寻找一些程度的变化。术语抗体涵盖序列变异体,其保持CD38结合特异性并且其优选实质上保持亲本抗体的亲和力。这类期望部分归因于基因码的简并,以及已知的保守性氨基酸序列变异的进化成功,其不显著地改变所编码的蛋白质的性质。因此,这类变异体和同系物视为彼此实质上相同并且包括在本发明范畴内。因此,抗体包括单个或多个氨基酸取代、缺失、加成或置换的变异体,其保留亲本抗体的生物特性(例如结合特异性和结合亲和力)。变异体优选是保守性,但可以是非保守性。

[0045] 称为互补决定区(CDR)和构架区(FR)的氨基酸位置可根据感兴趣的免疫学蛋白质的Kabat序列(Kabat Sequences of Proteins of Immunological Interest),美国国家卫生研究院,马里兰州贝塞斯达(Bethesda,Md.),1987和1991(在本文中也称为Kabat编号系统)来定义。另外,称为CDR和FR的氨基酸位置可根据增强型Chothia编号方案(Enhanced Chothia Numbering Scheme)(<http://www.bioinfo.org.uk/mdex.html>)定义。抗体的重链恒定区可由EU编号系统(埃德尔曼,GM(Edelman,GM)等人(1969).,美国国家科学院院刊,

63,78-85) 定义。

[0046] 根据Kabat的编号系统,VH FR和CDR可安置如下:残基1-30 (FR1)、31-35 (CDR1)、36-49 (FR2)、50-65 (CDR2)、66-94 (FR3)、95-102 (CDR3) 和103-113 (FR4),并且VL FR和CDR安置如下:残基1-23 (FR1)、24-34 (CDR1)、35-49 (FR2)、50-56 (CDR2)、57-88 (FR3)、89-97 (CDR3) 和98-107 (FR4)。在一些情况下,可变区的长度可增加,并且根据Kabat编号系统,一些氨基酸可由数字后接一个字母指定。本说明书不限于如由Kabat编号系统定义的FWR和CDR,但包括所有编号系统,包括科西亚 (Chothia) 等人 (1987) 分子生物学杂志 (J.Mol.Biol.) 196:901-17;科西亚等人 (1989) 自然 (Nature) 342:877-83;和/或Al-拉兹卡尼 (Al-Lazikani) 等人 (1997) 分子生物学杂志 273:927-48 的标准编号系统;霍尼赫 (Honnegher) 等人 (2001) 分子生物学杂志,309:657-70 的编号系统;或朱迪切利 (Giudicelli) 等人, (1997) 核酸研究 (Nucleic Acids Res.) 25:206-11 中论述的IMGT系统。在一些方面中,CDR是根据Kabat编号系统定义。

[0047] 在一些特定方面中,对于本文中所描述的重链CDR2子域中的任一个,根据Kabat编号系统,五个C端氨基酸可不直接参与抗原结合,并且因此,应理解,这五个C端氨基酸中的任一个或多个可由另一种天然存在的氨基酸取代而不实质上不利地影响抗原结合。在一些方面中,对于本文中所描述的轻链CDR1子域中的任一个,根据Kabat编号系统,四个N端氨基酸可不直接参与抗原结合,并且因此,应理解,这四个氨基酸中的任一个或多个可由另一种天然存在的氨基酸取代而不实质上不利地影响抗原结合。举例来说,如由派得兰 (Padlan) 等人 (1995) 美国实验生物学学会联合会杂志 (FASEB J.) 9:133-139 所描述,重链CDR2 的五个C端氨基酸和/或轻链CDR1的四个N端氨基酸可不参与抗原结合。在一些方面中,重链CDR2 和轻链CDR1皆不直接参与抗原结合。

[0048] 在一些方面中,本文中描述和/或例示的抗体中可使用氨基酸的化学类似物。氨基酸的化学类似物的使用适用于例如使分子稳定(如若需要投与个体)。本文中涵盖的氨基酸的类似物包括(但不限于)侧链的修饰、非天然氨基酸和/或其衍生物在肽、多肽或蛋白质合成期间的并入以及使用交联剂和其它对蛋白质分子或其类似物施加构形限制的方法。

[0049] 抗CD38抗体可包含转译后修饰或部分,其可影响抗体活性或稳定性。这些修饰或部分包括(但不限于)甲基化、乙酰化、糖基化、硫酸化、磷酸化、羧化和酰胺化部分以及此项技术中众所周知的其它部分。部分包括在自然界中的免疫球蛋白分子上通常发现或以其它方式通过重组型表达系统(包括原核和真核表达系统)添加到抗体中的任何化学基团或基团组合。

[0050] 由本发明涵盖的侧链修饰的实例包括氨基的修饰,如通过与醛反应,接着用 NaBH_4 还原而进行的还原性烷基化;用甲基乙酰胺进行的酰胺化;用乙酸酐进行的酰化;用氰酸酯进行的氨基的甲氨酰化;用2,4,6-三硝基苯磺酸(TNBS)进行的氨基的三硝基苯甲基化;用丁二酸酐和四氢邻苯二甲酸酐进行的氨基的酰化;以及用吡哆醛-5-磷酸盐进行的赖氨酸的吡哆醛化,接着用 NaBH_4 进行的还原。

[0051] 精氨酸残基的胍基团可通过与如2,3-丁二酮、苯基乙二醛和乙二醛等试剂形成杂环缩合产物来修饰。羧基可通过经由O-酰基异脲形成进行碳化二亚胺活化,接着进行后续衍生作用,例如成为相应的酰胺来修饰。巯基可通过如以下等方法修饰:用碘乙酸或碘乙酰胺羧基甲基化;过甲酸氧化成半胱氨酸;与其它硫醇化合物形成混合二硫化物;与马来酰

亚胺、顺丁烯二酸酐或其它经取代的马来酰亚胺反应;使用4-氯汞基苯甲酸盐、4-氯汞基苯磺酸、氯化苯基汞、2-氯汞基-4-硝基苯酚和其它汞剂形成汞剂衍生物;用氰酸酯在碱性pH值下氨基甲酰化。色氨酸残基可通过例如用N-溴代丁二酰亚胺氧化或用2-羟基-5-硝基苯甲基溴或次磺酰基卤化物进行吲哚环的烷基化来修饰。另一方面,酪氨酸残基可通过用四硝基甲烷硝化以形成3-硝基酪氨酸衍生物来改变。组氨酸残基的咪唑环的修饰可通过用碘乙酸衍生物烷基化或用焦碳酸二乙酯进行N-乙酰基化来实现。

[0052] 可使用交联剂例如使抗CD38抗体和经抗CD38抗体灭毒的干扰素 α -2b构筑体的3D构型稳定,使用同质双官能交联剂,如具有 $(CH_2)_n$ 间隔基的双功能酰亚胺基酯(其中 $n=1$ 到 $n=6$)、戊二醛、N-羟基丁二酰亚胺酯和异质双官能试剂,其通常含有氨基反应性部分,如N-羟基丁二酰亚胺,和另一基团特异性-反应性部分,如马来酰亚胺基或二巯基部分(SH)或碳化二亚胺(COOH)。在一些方面中,抗体可通过已知保护/封端基团衍生以防止蛋白水解分裂或增强活性或稳定性。

[0053] 抗CD38抗体可以是亲和力成熟的,或可包含氨基酸变化,其降低免疫原性,例如通过移除预期的MHC II类结合主结构。本文中所描述的抗体的治疗性效用可通过调节其功能特性(如抗体依赖性细胞介导的细胞毒性(ADCC)、补体依赖性细胞毒性(CDC)、血清半衰期、生物分布和与Fc受体的结合或任何这些特性的组合)而进一步增强。这种调节可通过蛋白质工程改造、糖基工程改造或化学方法实现。视所需治疗应用而定,增加或降低这些活性中的任一种可以是有利的。使用Potelligent®方法的糖基工程改造的实例描述于新川(Shinkawa T.)等人(2003)生物化学杂志(J.Biol.Chem.) 278:3466-73中。

[0054] 抗CD38抗体可包括可调节其血清半衰期和生物分布的修饰,包括可调节抗体与新生Fc受体(FcRn)的相互作用的修饰,所述新生Fc受体是在保护IgG避免代谢和保持高血清抗体浓度中具有重要作用的受体。血清半衰期调节修饰可存在于IgG1或IgG4的Fc区中,包括M252Y/S254T/T256E的三重取代(根据EU编号系统埃德尔曼(Edelman,G.M.)等人(1969)美国国家科学院院刊63,78-85)编号)(例如SEQ ID NO:13、SEQ ID NO:14、SEQ ID NO:15、SEQ ID NO:16),如美国专利第7,083,784号中所描述。其它取代可在位置250和428处发生,参见例如美国专利第7,217,797号,以及在位置307、380和434处发生,参见例如WO 00/42072。调节与Fc受体的结合以及由这些受体介导的后续功能(包括FcRn结合和血清半衰期)的恒定域氨基酸取代的实例描述于美国公开案第2009/0142340号、第2009/0068175号和第2009/0092599号中。裸抗体可具有重链C端赖氨酸,其被忽略或移除以降低非均质性。人类IgG4中S228P(EU编号)的取代可使活体内抗体制造臂交换稳定(拉布林(Labrin)等人(2009)自然生物技术(Nature Biotechnology) 27:8;767-773)。

[0055] 已知连接到抗体分子的聚糖可影响抗体与Fc受体和聚糖受体的相互作用并且由此影响抗体活性,包括血清半衰期。因此,某些调节所需抗体活性的糖型可赋予治疗优势。用于产生经工程改造的糖型的方法包括(但不限于)美国专利第6,602,684号、第7,326,681号和第7,388,081号以及PCT公开案第WO 08/006554号中描述的方法。或者,抗体序列可经修饰以移除相关糖型连接位点。

[0056] 抗CD38抗体优选对CD38上的抗原决定基(其包括小于约 1×10^{-4} M的解离常数(Kd))具有结合亲和力。在一些实施例中,Kd小于约 1×10^{-5} M。在其它实施例中,Kd小于约 1×10^{-6} M。在其它实施例中,Kd小于约 1×10^{-7} M。在其它实施例中,Kd小于约 1×10^{-8} M。在其它实施例

中, K_d 小于约 $1 \times 10^{-9} M$ 。在其它实施例中, K_d 小于约 $1 \times 10^{-10} M$ 。在其它实施例中, K_d 小于约 $1 \times 10^{-11} M$ 。在一些实施例中, K_d 小于约 $1 \times 10^{-12} M$ 。在其它实施例中, K_d 小于约 $1 \times 10^{-13} M$ 。在其它实施例中, K_d 小于约 $1 \times 10^{-14} M$ 。在其它实施例中, K_d 小于约 $1 \times 10^{-15} M$ 。亲和力和值是指通过标准方法获得的值, 包括表面等离子共振, 如 Biacore™ 分析, 或使用 Octet® Red 96 (复地生物公司 (Forte Bio)) 浸渍读取系统 (Dip-and-Read system) 进行的分析。

[0057] 抗CD38抗体优选能够结合于CD38阳性细胞。抗体可以小于约100nM的 EC_{50} 值结合于CD38阳性细胞。抗体可以小于约75nM的 EC_{50} 值结合于CD38阳性细胞。抗体可以小于约50nM的 EC_{50} 值结合于CD38阳性细胞。抗体可以小于约30nM的 EC_{50} 值结合于CD38阳性细胞。抗体可以小于约25nM的 EC_{50} 值结合于CD38阳性细胞。抗体可以小于约20nM的 EC_{50} 值结合于CD38阳性细胞。抗体可以小于约18nM的 EC_{50} 值结合于CD38阳性细胞。抗体可以小于约15nM的 EC_{50} 值结合于CD38阳性细胞。抗体可以小于约13nM的 EC_{50} 值结合于CD38阳性细胞。抗体可以小于约10nM的 EC_{50} 值结合于CD38阳性细胞。

[0058] 抗CD38抗体可包含有包含SEQ ID NO:17、SEQ ID NO:18、SEQ ID NO:19或SEQ ID NO:20的氨基酸序列的重链。抗体可包含有包含SEQ ID NO:21、SEQ ID NO:22或SEQ ID NO:23的氨基酸序列的轻链。在一些方面中, SEQ ID NO:17的重链氨基酸序列不包括SEQ ID NO:24的氨基酸序列。在一些方面中, SEQ ID NO:21的轻链氨基酸序列不包括SEQ ID NO:25的氨基酸序列。这类抗CD38抗体的变异体可经工程改造和表达使得抗体在循环中具有降低的免疫原性、增强的稳定性和增强的半衰期, 而不显著损失抗体对CD38抗原的特异性或亲和力。这些变异型抗体可与经灭毒的干扰素融合。

[0059] 在一些方面中, 抗CD38抗体包含特定的重链和轻链配对。具有SEQ ID NO:17的氨基酸序列的重链中的任一个可与任何具有SEQ ID NO:21的氨基酸序列的轻链配对。具有SEQ ID NO:18的氨基酸序列的重链中的任一个可与任何具有SEQ ID NO:22的氨基酸序列的轻链配对。具有SEQ ID NO:19的氨基酸序列的重链中的任一个可与任何具有SEQ ID NO:21的氨基酸序列的轻链配对。具有SEQ ID NO:20的氨基酸序列的重链中的任一个可与任何具有SEQ ID NO:23的氨基酸序列的轻链配对。

[0060] 在一些优选方面中, 抗CD38抗体包含表1、表2或表3的重链和轻链配对。在更优选方面中, 抗CD38抗体包含表4的重链和轻链配对。在更优选方面中, 抗CD38抗体包含有包含SEQ ID NO:27的氨基酸序列的重链可变区和包含SEQ ID NO:29的氨基酸序列的轻链可变区。

[0061] 表1. 重链和轻链可变区配对

[0062]	抗体名称	可变重链 SEQ ID NO:(氨基酸)	可变轻链 SEQ ID NO: (氨基酸)
	A02.10	208	25

[0063]

A02.11	209	25
A02.112	43	77
A02.12	43	77
A02.13	44	77
A02.16	43	104
A02.17	43	105
A02.18	43	85
A02.19	43	86
A02.2	24	77
A02.20	43	87
A02.21	43	88
A02.22	43	89
A02.23	43	90
A02.24	43	91
A02.25	43	92
A02.26	43	93
A02.27	43	94
A02.28	43	95
A02.29	43	96
A02.3	206	77
A02.30	43	97
A02.31	43	98
A02.32	43	99
A02.33	43	100
A02.43	43	101
A02.35	43	102
A02.36	43	103
A02.37	43	78
A02.38	43	123
A02.39	43	122
A02.4	207	77
A02.40	131	77
A02.41	130	77
A02.43	130	123
A02.44	131	122
A02.46	43	79
A02.47	43	80
A02.48	43	81
A02.49	43	82
A02.5	208	77
A02.50	43	83
A02.51	43	84
A02.52	43	106
A02.53	43	107
A02.54	43	108
A02.55	43	109
A02.56	43	110
A02.57	43	111
A02.58	43	112
A02.59	43	113

[0064]

A02.6	209	77
A02.60	43	114
A02.61	43	115
A02.62	43	116
A02.63	43	117
A02.64	43	118
A02.65	43	119
A02.66	43	102
A02.67	43	121
A02.8	206	25
A02.9	207	25
X02.10	208	25
X02.100	24	70
X02.101	24	71
X02.102	24	72
X02.103	24	73
X02.104	24	74
X02.105	24	75
X02.106	24	76
X02.107	24	77
X02.108	41	25
X02.11	209	25
X02.110	42	25
X02.114	33	124
X02.115	33	125
X02.116	33	126
X02.117	33	127
X02.118	43	128
X02.119	43	129
X02.120	45	128
X02.121	46	128
X02.122	47	128
X02.123	48	128
X02.124	45	129
X02.125	46	129
X02.126	47	129
X02.127	48	129
X02.68	210	25
X02.69	31	25
X02.70	32	25
X02.71	33	25
X02.72	34	25
X02.73	35	25
X02.74	36	25
X02.75	37	25
X02.76	38	25
X02.77	39	25
X02.78	40	25
X02.8	206	25
X02.80	24	50

[0065]

X02.81	24	51
X02.82	24	52
X02.83	24	53
X02.84	24	54
X02.85	24	55
X02.86	24	56
X02.87	24	57
X02.88	24	58
X02.89	24	59
X02.9	207	25
X02.90	24	60
X02.91	24	61
X02.92	24	62
X02.93	24	63
X02.94	24	64
X02.95	24	65
X02.96	24	66
X02.97	24	67
X02.98	24	68
X02.99	24	69

[0066] 表2. 重链和轻链可变区配对

[0067]

抗体名称	可变重链 SEQ ID NO:(氨基酸)	可变轻链 SEQ ID NO: (氨基酸)
A10.1	139	167
A10.10	147	167
A10.11	148	167
A10.12	149	167
A10.13	150	167
A10.14	151	167
A10.15	152	167
A10.16	153	167
A10.17	27	171
A10.18	27	172
A10.19	27	173
A10.2	140	167
A10.20	27	174
A10.21	27	29
A10.22	27	175
A10.23	27	176
A10.24	27	177
A10.25	27	178
A10.26	27	179
A10.27	27	180
A10.28	27	181
A10.29	27	182
A10.3	28	167
A10.30	27	183
A10.31	27	184
A10.32	27	185

[0068]

A10.35	154	167
A10.36	27	186
A10.38	26	167
A10.39	26	171
A10.4	141	167
A10.40	26	172
A10.41	26	173
A10.42	26	174
A10.43	26	29
A10.44	26	175
A10.45	26	176
A10.46	26	177
A10.47	26	178
A10.48	26	179
A10.49	26	180
A10.5	142	167
A10.50	26	181
A10.51	26	182
A10.52	26	183
A10.53	26	184
A10.54	26	185
A10.57	26	186
A10.59	27	167
A10.6	143	167
A10.7	144	167
A10.8	145	167
A10.9	146	167
A10A2.0 (嵌合)	132	163
A10A2.1	133	164
A10A2.10	134	166
A10A2.11	134	167
A10A2.12	134	168
A10A2.13	134	169
A10A2.14	134	170
A10A2.15	135	164
A10A2.16	135	165
A10A2.17	135	166
A10A2.18	135	167
A10A2.19	135	168
A10A2.2	133	165
A10A2.20	135	169
A10A2.21	135	170
A10A2.22	26	164
A10A2.23	26	165
A10A2.24	26	166
A10A2.25	26	167
A10A2.26	26	168
A10A2.27	26	169
A10A2.28	26	170
A10A2.29	136	164

[0069]

A10A2.3	133	166
A10A2.30	136	165
A10A2.31	136	166
A10A2.32	136	167
A10A2.33	136	168
A10A2.34	136	169
A10A2.35	136	170
A10A2.36	137	164
A10A2.37	137	165
A10A2.38	137	166
A10A2.39	137	167
A10A2.4	133	167
A10A2.40	154	168
A10A2.41	137	169
A10A2.42	137	170
A10A2.43	137	164
A10A2.44	138	165
A10A2.45	138	166
A10A2.46	138	167
A10A2.47	138	168
A10A2.48	138	169
A10A2.49	138	170
A10A2.5	133	168
A10A2.50	27	164
A10A2.51	27	165
A10A2.52	27	166
A10A2.53	27	167
A10A2.54	27	168
A10A2.55	27	169
A10A2.56	27	170
A10A2.6	133	169
A10A2.7	133	170
A10A2.8	134	164
A10A2.9	134	165
X10.100	155	30
X10.101	156	30
X10.102	157	30
X10.103	158	30
X10.104	159	30
X10.105	160	30
X10.106	161	30
X10.107	162	30
X10.108	155	189
X10.109	156	189
X10.110	157	189
X10.111	158	189
X10.112	159	189
X10.113	160	189
X10.114	161	189
X10.115	162	189

[0070]

X10.116	155	190
X10.117	156	190
X10.118	157	190
X10.119	158	190
X10.120	159	190
X10.121	160	190
X10.122	161	190
X10.123	162	190
X10.124	155	191
X10.125	156	191
X10.126	157	191
X10.127	158	191
X10.128	159	191
X10.129	160	191
X10.130	161	191
X10.131	162	191
X10.132	155	192
X10.133	156	192
X10.134	157	192
X10.135	158	192
X10.136	159	192
X10.137	160	192
X10.138	161	192
X10.139	162	192
X10.140	155	193
X10.141	156	193
X10.142	157	193
X10.143	158	193
X10.144	159	193
X10.145	160	193
X10.146	161	193
X10.147	162	193
X10.60	27	187
X10.61	27	188
X10.62	27	30
X10.63	27	189
X10.64	27	190
X10.65	27	191
X10.66	27	192
X10.67	27	193
X10.68	155	167
X10.69	156	167
X10.70	157	167
X10.71	158	167
X10.72	159	167
X10.73	160	167
X10.74	161	167
X10.75	162	167
X10.76	26	187
X10.77	26	188

[0071]	X10.78	26	30
	X10.79	26	189
	X10.80	26	190
	X10.81	26	191
	X10.82	26	192
	X10.83	26	193
	X10.84	155	187
	X10.85	156	187
	X10.86	157	187
	X10.87	158	187
	X10.88	159	187
	X10.89	160	187
	X10.90	161	187
	X10.91	162	187
	X10.92	155	188
	X10.93	156	188
	X10.94	157	188
	X10.95	158	188
	X10.96	159	188
	X10.97	160	188
	X10.98	161	188
	X10.99	162	188

[0072] 表3. 重链和轻链可变区配对

[0073]	抗体名称	可变重链 SEQ ID NO: (氨基酸)	可变轻链 SEQ ID NO: (氨基酸)
	X910/12-HC-L0- IFN- α (A145D) IgG4	130	122
	X913/15-HC-L0- IFN- α (A145D) IgG4	131	123

[0074] 表4. 重链和轻链可变区配对

[0075]	抗体名称	可变重链 SEQ ID NO: (氨基酸)	可变轻链 SEQ ID NO: (氨基酸)
	X10.78	26	30
	A10.21	27	29
	A10.43	26	29
	A10.62	27	30
	A10.152	28	30

[0076] 抗CD38抗体可以是所属领域中描述的抗CD38抗体。可如本文中所描述使用的抗CD38抗体的实例包括美国专利第5,545,405号、第7,829,673号、第8,088,896号或第8,153,765号中描述或美国公开案第2002/0164788号、第2003/0211553号、第2009/0076249号、第2009/0123950号或第2010/0285004号中描述的抗体。

[0077] 作为构筑体的的一部分,抗CD38抗体优选接合到IFN α -2b的经灭毒形式。IFN α -2b灭毒与通过结合于细胞表面上的干扰素受体获得的干扰素的生物活性有关。灭毒可通过将某些氨基酸变化引入干扰素蛋白质序列来实现。

[0078] 经灭毒的干扰素分子接合到抗CD38抗体,使得抗体可充当经灭毒的干扰素的传递媒介,将其传递到CD38阳性细胞,引起由经灭毒的干扰素分子引起的脱靶干扰素活性的减弱。经抗CD38抗体灭毒的干扰素 α -2b构筑体包括(但不限于)本文中描述或例示的任何特异

性结合于CD38 (其接合到经灭毒的IFN α -2b蛋白质)的抗体,包括SEQ ID NO:3、SEQ ID NO:4、SEQ ID NO:5、SEQ ID NO:6、SEQ ID NO:7、SEQ ID NO:211、SEQ ID NO:212或SEQ ID NO:213的IFN α -2b。

[0079] 人类CD38包含SEQ ID NO:1的氨基酸序列,并且食蟹猕猴CD38包含SEQ ID NO:2的氨基酸序列。

[0080] 使用抗CD38抗体作为经灭毒的干扰素 α -2b的传递媒介。不希望受限于任何特定理论或机制或作用,相信抗体(作为传递媒介)补偿干扰素分子的减弱的结合于其受体(其灭毒)的能力。在此意义上,经灭毒的干扰素具有降低的与其在健康细胞上的受体,且具体来说不表达CD38的细胞相互作用的能力。相信同时使经灭毒的干扰素靠近其在CD38阳性细胞上的受体,抗体可增强经灭毒的干扰素结合于其相关受体和诱导治疗作用的能力,同时呈现减弱的诱导不表达CD38的健康细胞上的不合需要的作用的能力。使经灭毒的干扰素接合到抗CD38抗体并不显著影响抗体特异性结合于表达CD38的细胞(包括活体内细胞)上的CD38的能力。

[0081] 抗体可与经灭毒的配位体融合例如以形成经抗体灭毒的配位体构筑体,其与由经灭毒的配位体对细胞表面受体的作用引起的活化信号传导路径相比展示升高的抗原特异性指数(ASI)。这些构筑体是基于以下观察结果:在抗体-配位体构筑体的情形下,配位体部分可以使得抗原阴性细胞上的配位体活性显著灭毒的方式突变,而抗原阳性细胞上的配位体活性仅适当地(如果全部)灭毒。这类构筑体与自由配位体相比,显示比抗原阴性细胞大一个、两个、三个、四个或五个数量级的更大的对抗原阳性细胞的效能。在一些方面中,经抗体灭毒的配位体构筑体保持未经灭毒的自由(即未连接到抗体)配位体对抗原阳性细胞的效能的至少1%、至少10%、至少20%、至少30%、至少40%或至少50%。在一些方面中,经抗体灭毒的配位体构筑体保持未经灭毒的自由(即未连接到抗体)配位体的最大活性的至少30%、至少50%、至少75%或至少90%。最大活性包括在剂量反应曲线的高、平线区部分的信号传导活性(或其下游作用)的量,其中试剂的进一步增加并不会进一步增加反应量。

[0082] 在一些方面中,与干扰素配位体中的减毒突变的抗体融合和包含所述减毒突变与不具有融合物的抗体相比可使抗原特异性指数(ASI)增加超过10倍,优选超过50倍,优选超过100倍,优选超过1000倍或优选超过10,000倍。ASI与目标抗原阳性细胞上自由的未经突变的多肽配位体相比包含抗体-IFN配位体构筑体的信号传导活性的成倍效能增加,与目标抗原阴性细胞上自由的未经突变的多肽配位体相比,信号传导活性的效能的成倍降低。效能可由EC₅₀值定量表示,其为剂量反应曲线的数学中点,其中剂量指分析法中配位体或抗体-配位体构筑体的浓度,并且反应指在特定剂量下细胞对配位体的信号传导活性的定量反应。因此,举例来说,当展示第一化合物具有比相同细胞上第二化合物的EC₅₀低10倍的EC₅₀(例如以摩尔单位表示)时,通常在通过相同方法测量时,第一化合物称为具有10倍更高的效能。相反,当展示第一化合物具有比相同细胞上第二化合物的EC₅₀高10倍的EC₅₀时,通常在通过相同方法测量时,第一化合物称为具有10倍更低的效能。

[0083] 接合到抗CD38抗体的干扰素 α -2b配位体优选包含其氨基酸序列中的变化,包括使干扰素刺激其在不具有抗体结合的CD38的抗原细胞表面表达的细胞上的各别受体的活性降低的点突变和/或缺失。干扰素 α 的优选变异体包含SEQ ID NO:8的干扰素 α 2b氨基酸序列的位置168处的氨基酸变化。举例来说,位置168处的氨基酸(其在亲本IFN- α 2b分子(SEQ ID

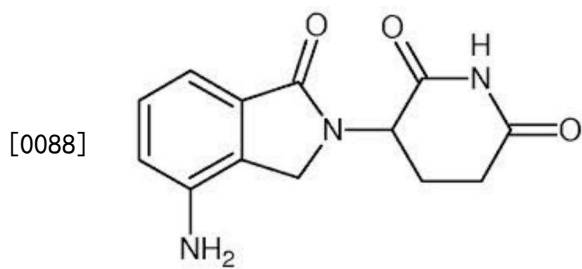
NO:8) 中是丙氨酸) 优选变成甘氨酸 (Gly/G) (SEQ ID NO:6) 或天冬氨酸 (Asp/D) (SEQ ID NO:3)。在一些优选方面中, IFN- α 2b 在 IFN- α 2b 与 IgG 重链恒定域 (如人类 IgG1 或人类 IgG4 重链恒定域) 融合时在其 N 端截短。截短的 IFN- α 2b 不具有 SEQ ID NO:8 的二十三个 N 端氨基酸 (Met 1 到 Gly 23 缺失), 并且截短的 IFN- α 2b 包含 SEQ ID NO:4 的氨基酸序列。截短的 IFN- α 2b 还可以包含先前是位置 168 处的氨基酸变化, 但其在截短的蛋白质中变成位置 145 (例如丙氨酸 168 变成丙氨酸 145)。在截短的 IFN- α 2b 中, 丙氨酸优选变成甘氨酸 (Gly/G) (SEQ ID NO:7) 或天冬氨酸 (Asp/D) (SEQ ID NO:5)。具有 A145D 变化 (SEQ ID NO:3 或 SEQ ID NO:5) 的干扰素尤其优选, 因为经灭毒的干扰素接合到本发明的抗体。IFN- α 的这些点突变、灭毒版本中的任一个可接合到本文中所描述的任何抗体, 例如以经抗体灭毒的干扰素构筑体形式。在一些方面中, 使经突变的 IFN α -2b 蛋白质 (如 SEQ ID NO:8) 接合到抗 CD38 抗体可减弱干扰素分子的生物活性。在本发明中, 经灭毒的干扰素、经灭毒的 IFN α -2b、IFN α -2b A145D 和 IFN α -2b A145G 可互换地使用。

[0084] 在尤其优选方面中, 抗 CD38 抗体与包含 SEQ ID NO:211、SEQ ID NO:212 或 SEQ ID NO:213 的氨基酸序列的经灭毒的干扰素 α 2b 融合。在这些经灭毒的干扰素 α 2b 分子中, 亲本干扰素 α 2b 分子的 N 端 23 个氨基酸缺失, 产生具有 165 个氨基酸的截短变异体, 使得亲本干扰素 α 2b 分子的第 24 号氨基酸变成截短变异体的第 1 号氨基酸。在这些截短变异体中, 某些其他氨基酸可以经取代。举例来说, 位置 106 处的苏氨酸可变成丙氨酸 (T106A) 以移除糖基化位点 (糖基化干扰素 α 2b) (例如 SEQ ID NO:211)。另外, 截短变异体的位置 145 处的丙氨酸可变成天冬氨酸 (SEQ ID NO:212) 或可变成甘氨酸 (SEQ ID NO:213)。

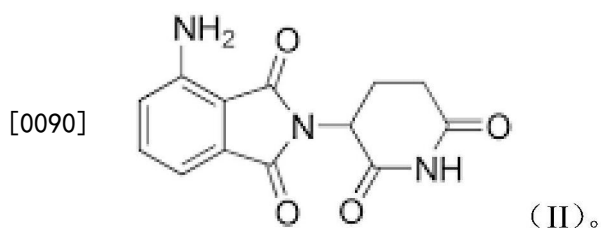
[0085] 在一些方面中, 抗体与干扰素之间的键包含融合物, 举例来说, 干扰素的 N 端或 C 端与抗体的重链或轻链的 N 端或 C 端之间的肽键。在一个优选方面中, 抗体与干扰素之间不存在连接子 (除融合蛋白的第一分量的最后一个 C 端氨基酸与融合蛋白的第二分量的 N 端氨基酸之间的以核糖体方式合成的肽键以外), 并且抗体和干扰素由此直接融合。相信直接融合 (无介入连接肽) 提供干扰素蛋白质的至少可测量的程度的灭毒, 并且还相信这一灭毒与源于引入到干扰素蛋白质中的突变 (包括本文中描述或例示的突变) 的干扰素蛋白质的灭毒加成。举例来说, 在一些方面中, 经抗 CD38 抗体灭毒的干扰素 α -2b 构筑体包含 SEQ ID NO:216 的氨基酸序列 (重链和干扰素) 以及 SEQ ID NO:217 的氨基酸序列 (轻链)。

[0086] 在一些方面中, 构筑体包括构筑体的第一蛋白质的最后一个 C 端氨基酸与构筑体的第二蛋白质的 N 端氨基酸之间的氨基酸的介入延伸子。这类肽连接子中的氨基酸数目长度可以是 1 到 50 个, 优选长度是 1-20 个。这类连接子的序列可包括主要由甘氨酸和丝氨酸组成的序列, 例如序列 $(G_4S)_n$, 其中 n 可以是 1 到约 10 的任何数字, 并且优选是 1 到约 4。

[0087] 作为治疗模态, 并且作为疗法或治疗方案的一部分, 经抗 CD38 抗体灭毒的干扰素 α -2b 构筑体与来那度胺配对。来那度胺, 也称为 (RS)-3-(4-氨基-1-侧氧基-1,3-二氢-2H-异吡啶-2-基) 哌啶-2,6-二酮, 具有化学式, 式 I:



[0089] 作为一种替代性治疗模态,并且作为疗法或治疗方案的一部分,经抗CD38抗体灭毒的干扰素 α -2b构筑体可与泊利度胺配对。因此,泊利度胺可在本文中描述或例示的任何系统、试剂盒、方法、组合物或用途中取代来那度胺。泊利度胺,也称为(RS)-4-氨基-2-(2,6-二侧氧基哌啶-3-基)异吲哚-1,3-二酮,具有化学式,式II:



[0091] 在一些方面中,经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺各自包含在组合物中。组合物可根据组合疗法使用。组合疗法可包含经抗CD38抗体灭毒的干扰素 α -2b构筑体与来那度胺或泊利度胺的单独组合物的组合物,或可包含两种药剂共同的组合物。组合物可包含至少一种任何适合的助剂,如(但不限于)一种或多种稀释剂、结合剂、稳定剂、缓冲剂、盐、亲油性溶剂、防腐剂、佐剂或其它适合的载剂和/或赋形剂。优选是医药学上可接受的助剂。经抗CD38抗体灭毒的干扰素 α -2b构筑体和/或来那度胺或泊利度胺可与可接受的载剂(如医药学上可接受的载剂)一起调配。适合的载剂包括任何不干扰抗体和/或干扰素的生物活性并且优选对接受其投药的主体无毒性的介质。载剂可以是水性溶液,如水、生理食盐水或乙醇,或生理学上相容的缓冲液,如汉克氏溶液(Hanks's solution)、林格氏溶液(Ringer's solution)或生理盐水缓冲液。载剂可含有调配剂,如悬浮剂、稳定剂和/或分散剂。

[0092] 适用于组合物中的药物赋形剂和添加剂包括包括(但不限于)蛋白质、肽、氨基酸、脂质和碳水化合物(例如糖,包括单糖、二糖、三糖、四糖和寡糖;衍生糖,如糖醇、醛糖酸、酯化糖和其它已知糖;以及多糖或糖聚合物),其可以单独或以组合形式存在,并且单独或以组合形式占任何适合的重重量或体积。例示性蛋白质赋形剂包括血清白蛋白,如人类血清白蛋白(HSA)、重组型人类白蛋白(rHA)、明胶、酪蛋白和其它已知蛋白质。还可以发挥缓冲能力的代表性氨基酸包括丙氨酸、甘氨酸、精氨酸、甜菜碱、组氨酸、谷氨酸、天冬氨酸、半胱氨酸、赖氨酸、白氨酸、异白氨酸、缬氨酸、甲硫氨酸、苯丙氨酸和阿斯巴甜糖(aspartame)。一种优选氨基酸是组氨酸。第二优选氨基酸是精氨酸。

[0093] 适用于组合物中的碳水化合物赋形剂包括例如单糖,如果糖、麦芽糖、半乳糖、葡萄糖、D-甘露糖和山梨糖;双糖,如乳糖、蔗糖、海藻糖和纤维二糖;多糖,如棉籽糖、松三糖、麦芽糊精、聚葡萄糖和淀粉;以及糖醇,如甘露醇、木糖醇、麦芽糖醇、乳糖醇、木糖醇、山梨糖醇(葡糖醇)和肌醇。用于本发明中的优选碳水化合物赋形剂是甘露醇、海藻糖和棉籽糖。

[0094] 组合物可包括缓冲液或pH值调节剂;通常,缓冲液是由有机酸或碱制成的盐。代表

性缓冲剂包括有机酸盐,如柠檬酸、抗坏血酸、葡糖酸、碳酸、酒石酸、丁二酸、乙酸或邻苯二甲酸的盐;Tris、缓血酸胺盐酸盐或磷酸盐缓冲剂。用于组合物中的优选缓冲剂是有机酸盐,如柠檬酸盐。

[0095] 组合物可包括聚合赋形剂/添加剂,如聚乙烯基吡咯烷酮、聚蔗糖(聚合糖)、葡萄糖结合剂(例如环糊精,如2-羟丙基- β -环糊精)、聚乙二醇、抗菌剂、抗氧化剂、抗静电剂、表面活性剂(例如聚山梨醇酯,如“TWEEN® 20”和“TWEEN® 80”)、脂质(例如磷脂、脂肪酸)、类固醇(例如胆固醇)和螯合剂(例如EDTA)。

[0096] 组合物可在持续释放媒剂或储存制剂中调配。举例来说,组合物可用适合的聚合或疏水性材料(例如以可接受油中的乳液形式)离子交换树脂调配,或以微溶性衍生物形式,例如以微溶性盐形式。脂质体和乳液是适用作疏水性药物的载剂的传递媒剂的众所周知的实例。

[0097] 组合物可经调配以用于以任何适合的剂型向个体投药。组合物可经调配以用于经口、颊内、经鼻、经皮、非经肠、可注射、静脉内、皮下、肌肉内、经直肠或经阴道投药。组合物可在适合的控制释放媒剂中与佐剂一起或以储槽式调配物形式调配。来那度胺优选是固体剂型,如丸剂或片剂。构筑体优选是用于非经肠投药的液体剂型。

[0098] 用于非经肠投药的制剂包括易于注射的无菌溶液、准备在临用前与溶剂组合的无菌干燥可溶性产物(包括皮下片剂)、易于注射的无菌悬浮液、准备在临用前与媒剂组合的无菌干燥不可溶产物和无菌乳液。

[0099] 包含与来那度胺或泊利度胺配对的经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合疗法可用于例如抑制、减少、降低、阻断或阻止在表面上表达CD38的细胞的增殖。包含与来那度胺或泊利度胺配对的经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合疗法可用于例如诱导、促进或增强在表面上表达CD38的细胞的细胞凋亡。表达CD38的细胞可以是淋巴细胞、自体免疫性淋巴细胞或肿瘤细胞,如白血病细胞、多发性骨髓瘤细胞或淋巴瘤细胞。优选的是,表达CD38的细胞是肿瘤细胞,并且肿瘤细胞可对来那度胺或泊利度胺具有抗性,包括在初始积极反应治疗期之后产生的抗性,使得肿瘤对组合疗法积极响应。

[0100] 包含与来那度胺或泊利度胺配对的经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合疗法系统可用于治疗具有肿瘤的患者,所述肿瘤包含在表面上表达CD38的细胞和/或至少部分由所述细胞介导。在一些方面中,用于治疗肿瘤的方法通常包含向需要治疗肿瘤的患者投与经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺。构筑体和来那度胺或泊利度胺中的每一个以可有效治疗患者中的肿瘤的量投与。组合物中可包含构筑体和来那度胺或泊利度胺中的每一个,其中每种试剂包含在单独的组合物中或包含在相同组合物中。组合疗法产生构筑体与来那度胺或泊利度胺的协同作用,使得与用经抗CD38抗体灭毒的干扰素 α -2b构筑体或来那度胺或泊利度胺但并非其两者治疗的相同类型的肿瘤细胞相比,存在肿瘤中细胞的抑制增强或增殖降低、增强的肿瘤中细胞的细胞凋亡的诱导和/或增强的肿瘤中CD38阳性细胞的杀死中的一种或多种。在一些方面中,肿瘤细胞可对来那度胺或泊利度胺具有抗性,包括在初始积极反应治疗期之后产生的抗性,使得肿瘤对组合疗法积极响应。因此,举例来说,组合疗法杀死停止对用单独的来那度胺或泊利度胺进行的治疗积极响应的肿瘤细胞。

[0101] 根据肿瘤治疗,与来那度胺或泊利度胺配对的经抗CD38抗体灭毒的干扰素 α -2b构

筑体的组合疗法可抑制或阻止肿瘤的再生和重新产生。可在某一时间段上测量这类再生和重新产生的抑制,例如至少约一年的时段、至少约2年的时段、至少约3年的时段、至少约5年的时段或超过5年的时段。

[0102] 作为组合疗法,可通过向血液中投与经抗CD38抗体灭毒的干扰素 α -2b构筑体或其组合物和来那度胺或其组合物(例如经由皮下或静脉内投药)来向肿瘤投与经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺,或包含经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合物和包含来那度胺或泊利度胺的组合物。可投与经抗CD38灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺使得每种药剂经由血流扩散到和/或扩散入肿瘤细胞。通过向肿瘤投与经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺,治疗接受经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺投药的患者。

[0103] 因此,组合疗法包含向具有肿瘤和需要治疗的患者投与一定量的经抗CD38抗体灭毒的干扰素 α -2b构筑体和一定量的来那度胺或泊利度胺,所述量(例如协同有效量)可有效治疗患者中的肿瘤。肿瘤可以是来那度胺抗性肿瘤,或可包含对来那度胺或泊利度胺具有抗性的细胞,包括在初始积极反应治疗期之后产生的抗性,使得肿瘤对组合疗法积极响应。经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺可实质上同时投与,举例来说,借助于共同包含这些药剂的组合物共同投与,或通过同时投与每种药剂的单独的组合物来进行。经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺可依序投与,其中经抗CD38抗体灭毒的干扰素 α -2b构筑体在来那度胺之前投与,或反之亦然。

[0104] 可用经抗CD38抗体灭毒的干扰素 α -2b构筑体与来那度胺或泊利度胺的组合疗法治疗的肿瘤包括(但不限于)以下肿瘤的来那度胺抗性形式:AIDS相关癌症、听神经瘤、急性淋巴细胞白血病、急性骨髓性白血病、腺样囊性癌、肾上腺皮质癌、原因不明性骨髓细胞化生、秃发、齿槽软部肉瘤、肛门癌、血管肉瘤、再生障碍性贫血、星形细胞瘤、共济失调-毛细血管扩张症、基底细胞癌(皮肤)、膀胱癌、骨癌、肠癌、脑干神经胶质瘤、大脑和CNS肿瘤、乳腺癌、CNS肿瘤、类癌、子宫颈癌、儿童脑部肿瘤、儿童癌症、儿童白血病、儿童软组织肉瘤、软骨肉瘤、绒膜癌、慢性淋巴细胞性白血病、慢性骨髓白血病、结肠直肠癌、皮肤T细胞淋巴瘤、皮肤纤维肉瘤、促结缔组织增生小型圆形细胞肿瘤、乳腺管癌瘤、内分泌癌症、子宫内膜癌、室管膜瘤、食道癌、尤文氏肉瘤(Ewing's sarcoma)、肝外胆管癌症、眼癌、眼:黑素瘤、成视网膜细胞瘤、输卵管癌症、范康尼氏贫血(fanconi anemia)、纤维肉瘤、胆囊癌、胃癌、胃肠道癌症、胃肠道类癌瘤、泌尿生殖癌症、生殖细胞肿瘤、妊娠期滋养细胞疾病、神经胶质瘤、妇科癌症、血液学恶性病、毛状细胞白血病、头颈癌、肝细胞癌症、遗传性乳癌、组织细胞增多病、霍奇金氏疾病(Hodgkin's disease)、人类乳突状瘤病毒、葡萄胎、高钙血症、喉咽癌、眼内黑素瘤、胰岛细胞癌症、卡堡氏肉瘤(Kaposi's sarcoma)、肾脏癌、兰格汉氏细胞组织细胞增多病(Langerhan's-cell-histiocytosis)、喉部癌症、平滑肌肉瘤、白血病、劳梅尼综合征(Li-Fraumeni syndrome)、唇癌、脂肪肉瘤、肝癌、肺癌、淋巴水肿、淋巴瘤、霍奇金氏淋巴瘤、非霍奇金淋巴瘤、雄性乳癌、肾脏的恶性横纹肌样肿瘤、神经管胚细胞瘤、黑素瘤、默克尔细胞癌(merkel cell cancer)、间皮瘤、转移性癌症、口腔癌、多重内分泌赘瘤形成、蕈样真菌病、骨髓发育不良综合症、多发性骨髓瘤、骨髓增生病症、鼻癌、鼻咽癌、肾胚细胞瘤、神经母细胞瘤、神经纤维瘤、奈梅亨断裂综合症(nijmegen breakage syndrome)、非黑素瘤皮肤癌、非小细胞肺癌(NSCLC)、眼癌、食道癌、口腔癌、口咽癌、骨肉瘤、造口术卵巢癌、胰腺

癌、副鼻癌、副甲状腺癌、腮腺癌、阴茎癌、周边神经外胚层肿瘤、垂体癌、真性红细胞增多症、前列腺癌、罕见癌症和相关病症、肾细胞癌、成视网膜细胞瘤、横纹肌肉瘤、罗特蒙德-汤姆森综合征 (Rothmund-Thomson syndrome)、唾液腺癌、肉瘤、神经鞘瘤、塞扎里综合征 (Sezary syndrome)、皮肤癌、小细胞肺癌 (SCLC)、小肠癌、软组织肉瘤、脊髓肿瘤、鳞状细胞癌瘤(皮肤)、胃癌、滑膜肉瘤、睾丸癌、胸腺癌、甲状腺癌、移行细胞癌(膀胱)、移行细胞癌(肾-骨盆-/-输尿管)、滋养细胞癌、尿道癌、泌尿系统癌、尿空斑蛋白、子宫肉瘤、子宫癌、阴道癌、外阴癌、瓦尔登斯特伦氏巨球蛋白血症和韦尔姆斯氏瘤 (Wilms' tumor)。在一个实施例中,肿瘤选自多发性骨髓瘤或非霍奇金淋巴瘤的群组。

[0105] 在优选方面中,经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺的组合疗法用于治疗具有多发性骨髓瘤、早期多发性骨髓瘤、前多发性骨髓瘤、瓦尔登斯特伦氏巨球蛋白血症、非霍奇金淋巴瘤、慢性骨髓性白血病、慢性淋巴细胞性白血病或急性淋巴细胞白血病的患者中的多发性骨髓瘤、早期多发性骨髓瘤、前多发性骨髓瘤、瓦尔登斯特伦氏巨球蛋白血症、非霍奇金淋巴瘤、慢性骨髓性白血病、慢性淋巴细胞性白血病或急性淋巴细胞白血病,包括多发性骨髓瘤、非霍奇金淋巴瘤、慢性骨髓性白血病、慢性淋巴细胞性白血病或急性淋巴细胞白血病的来那度胺抗性形式。在一些尤其优选方面中,经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺的组合疗法用于治疗具有多发性骨髓瘤、白血病或淋巴瘤的患者中的多发性骨髓瘤、白血病或淋巴瘤,包括多发性骨髓瘤、白血病或淋巴瘤的来那度胺抗性形式。在一些尤其优选方面中,经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺的组合疗法用于治疗具有多发性骨髓瘤的患者中的多发性骨髓瘤,包括多发性骨髓瘤的来那度胺抗性形式。来那度胺抗性包括在对来那度胺的初始反应治疗期之后产生的抗性,使得肿瘤对组合疗法积极响应。

[0106] 提供使用经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺或经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合物和来那度胺或泊利度胺的组合物作为肿瘤治疗中的组合疗法。本发明还提供经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺或经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合物和来那度胺或泊利度胺的组合物,其用作B细胞淋巴瘤治疗中的组合疗法。本发明还提供经抗CD38抗体灭毒的 α -2b构筑体和来那度胺或泊利度胺或经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合物和来那度胺或泊利度胺的组合物,其用作多发性骨髓瘤治疗中的组合疗法。本发明还提供经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺或经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合物和来那度胺或泊利度胺的组合物,其用作非霍奇金淋巴瘤治疗中的组合疗法。本发明还提供经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺或经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合物和来那度胺或泊利度胺的组合物,其用作慢性骨髓性白血病治疗中的组合疗法。本发明还提供经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺或经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合物和来那度胺或泊利度胺的组合物,其用作慢性淋巴细胞性白血病治疗中的组合疗法。本发明还提供经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺或经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合物和来那度胺或泊利度胺的组合物,其用作急性淋巴细胞白血病治疗中的组合疗法。本发明还提供经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺或经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合物和来那

度胺或泊利度胺的组合物的用途,其用作早期多发性骨髓瘤治疗中的组合疗法。本发明还提供经抗CD38抗体灭毒的 α -2b构筑体和来那度胺或泊利度胺或经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合物和来那度胺或泊利度胺的组合物的用途,其用作前多发性骨髓瘤治疗中的组合疗法。本发明还提供经抗CD38抗体灭毒的 α -2b构筑体和来那度胺或泊利度胺或经抗CD38抗体灭毒的干扰素 α -2b构筑体的组合物和来那度胺或泊利度胺的组合物的用途,其用作急性淋巴细胞白血病瓦尔登斯特伦氏巨球蛋白血症治疗中的组合疗法。

[0107] 在一个方面中,本发明提供试剂盒。试剂盒包含经抗CD38抗体灭毒的干扰素 α -2b构筑体、来那度胺或泊利度胺,和在组合疗法中使用构筑体和来那度胺治疗癌症(包括来那度胺抗性癌症)的说明。经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺可各自呈单独剂型形式、可各自呈如本文中所描述的组合物形式或可共同呈如本文中所描述的组合物形式,或可分离但意图在投与具有癌症的患者之前在适合的载剂中组合或混合。在一些方面中,试剂盒包含医药学上可接受的载剂和用于混合经抗CD38抗体灭毒的干扰素 α -2b构筑体与载剂的说明,以及用于混合来那度胺或泊利度胺与载剂的说明。经抗CD38抗体灭毒的干扰素 α -2b构筑体的医药学上可接受的载剂可与来那度胺或泊利度胺的医药学上可接受的载剂相同或不同。经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺优选以可有效治疗具有癌症的患者中的癌症的量存在中试剂盒中,例如协同有效量,包括可有效协同治疗来那度胺抗性癌症的量,或试剂盒可包括用于产生和/或投与用于治疗癌症的协同有效量的说明。对于非经肠投药,试剂盒可包含用于将经抗CD38抗体灭毒的干扰素 α -2b构筑体和/或来那度胺或泊利度胺或其组合物注入个体的装置,包括(但不限于)注射器和针,或导管。来那度胺抗性包括在初始积极反应治疗期之后产生的抗性,使得肿瘤对组合疗法积极响应。

[0108] 在本文中描述或例示的系统、组合物、试剂盒、方法和用法中的任一种中,经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺中的任一个或两个的协同有效量可与配对中包含另一种药剂有关。举例来说,来那度胺或泊利度胺的协同有效量可以是经抗CD38抗体灭毒的干扰素 α -2b构筑体的协同有效量的函数,或经抗CD38抗体灭毒的干扰素 α -2b构筑体的协同有效量可以是来那度胺或泊利度胺的协同有效量的函数。经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺与单独的每种药剂的肿瘤杀死作用相比,发挥协同作用以产生增强的肿瘤杀死作用。协同有效量可例如根据患者的年龄、性别、总体健康状况、患者的生理特征、肿瘤类型、肿瘤状态以及预期将以组合疗法形式向患者投与经抗CD38抗体灭毒的干扰素 α -2b构筑体和来那度胺或泊利度胺的医师已知的其它因素而变化。

[0109] 提供以下实例以更详细地描述本发明。其意图说明而非限制本发明。

[0110] 实例1

[0111] 经抗CD38抗体灭毒的IFN α -2b构筑体+来那度胺组合疗法的细胞系模型

[0112] 在这些实验中,将含有一千万个NCI-H929多发性骨髓瘤细胞的0.2ml 50% MATRIGEL®基质在侧腹皮下植入8-12周龄的雌性CB.17重症联合免疫缺陷(SCID)小鼠中。当肿瘤达到200-300mm³的平均尺寸时,将小鼠成对匹配到不同组中并且接着用媒剂(PBS)、0.5mg/kg的自由的未经灭毒的干扰素 α (IFN- α)、次最佳剂量的抗CD38抗体-IFN α -2b-145D构筑体(2.5mg/kg与0.5mg/kg IFN等效的摩尔当量;腹膜内,两周一次,其通过前述

活体内功效研究测定)、同型匹配抗体-IFN α -2b-145D构筑体(与抗CD38抗体同型匹配,不具有抗CD38特异性)、单独的来那度胺(2.5mg/kg)、自由的未经灭毒的干扰素 α 与来那度胺的组合、来那度胺与次最佳剂量的抗CD38抗体-IFN α -2b-145D构筑体的组合或同型对照抗体-IFN α -2b-145D构筑体与来那度胺的联合治疗。所投与的经抗CD38抗体灭毒的IFN α -2b构筑体的量真对向动物投与的0.5mg/kg的自由干扰素的IFN- α 摩尔当量标准化。这些实验的结果展示于图1和图2中。如果在研究完成之前,肿瘤生长到超过2000mm³的体积,那么处死动物。

[0113] 图2展示未经灭毒的干扰素 α (自由干扰素,不是构筑体的一部分)与来那度胺的较小协同作用。干扰素与来那度胺的组合与单独的干扰素或来那度胺相比延缓肿瘤生长,但最终肿瘤生长开始,并且肿瘤体积在开始治疗约一个月内快速增加。

[0114] 相比之下,图1展示经抗CD38抗体灭毒的IFN α -2b构筑体与来那度胺的联合的协同作用。尽管在单独使用时,构筑体、来那度胺和干扰素 α 中的每一个与媒介对照物相比延缓肿瘤生长,但最终开始肿瘤生长并且在两周到约一个月内加速。相比之下,构筑体与来那度胺的联合在整个实验持续时间内显示肿瘤生长抑制。所述作用较显著且与干扰素和来那度胺的加成作用明显不同,使得即使在使用同型对照抗体构筑体时,经抗CD38抗体灭毒的干扰素 α -2b构筑体的存在仍可解决所观察到的肿瘤生长开始的问题。

[0115] 实例2

[0116] 经抗CD38抗体灭毒的糖基化IFN α -2b构筑体+来那度胺联合疗法的细胞系模型

[0117] 在这一实验中,将含 1×10^7 个H929多发性骨髓瘤细胞的50% Matrigel®在侧腹皮下植入8-12周龄的雌性CB.17重症联合免疫缺陷(SCID)小鼠中。两周一次通过卡尺测量肿瘤体积。当肿瘤达到170-350mm³的平均尺寸时,随机分配小鼠并且开始治疗。如果在第60天完成研究之前,肿瘤生长到超过2000mm³的体积,那么处死动物。

[0118] 在这一实例中,研究抗CD38抗体与经灭毒的糖基化干扰素- α 2b(A10.21(T106A))的融合物与来那度胺的联合的投药的剂量水准和给药间隔。A10.21(T106A)是与糖基化经灭毒的IFN α 2b融合的抗CD38IgG4抗体x10.21,其具有取代A145D和T106A。治疗方案和结果概述于表5中并且个别动物的资料展示于图3A到3J中。十只动物分配到组1到10中的每个组中。治疗可引起动物中肿瘤的“部分消退”(PR)或完全消退(CR)。在PR反应中,在研究过程期间的三个连续测量结果中,肿瘤体积是其第1天体积的50%或更小,并且在这三个测量结果中的一个或多个中等于或大于13.5mm³。在CR反应中,在研究期间的三个连续测量结果中,肿瘤体积小于13.5mm³。任何在研究结束时具有CR反应的动物额外分类为无肿瘤存活者(TFS)。

[0119] 表5. 组合疗法治疗方案和结果概述。

组号	治疗 1				治疗 2				第 60 天	PR	CR	TFS
	药剂	mg/kg	途径	疗程	药剂	mg/kg	途径	疗程	MTV (n)			
1#	媒剂	-	腹膜内	两周一次持续 29 天	-	-	-	-	-	0	0	0
2	来那度胺	25	腹膜内	qd×21	-	-	-	-	726 (1)	1	0	0
3	A10.21 (T106A)	0.3	腹膜内	两周一次持续 29 天	-	-	-	-	425 (4)	0	0	0
4	A10.21 (T106A)	0.3	腹膜内	两周一次持续 29 天	来那度胺	25	腹膜内	qd×21	405 (8)	8	0	0
[0120] 5	A10.21 (T106A)	1	腹膜内	两周一次持续 29 天	-	-	-	-	70 (10)	2	4	4
6	A10.21 (T106A)	1	腹膜内	两周一次持续 29 天	来那度胺	25	腹膜内	qd×21	0 (10)	1	9	9
7	A10.21 (T106A)	1	腹膜内	四周一次持续 29 天	-	-	-	-	1008 (5)	0	0	0
8	A10.21 (T106A)	1	腹膜内	四周一次持续 29 天	来那度胺	25	腹膜内	qd×21	304 (10)	5	3	1
9	A10.21 (T106A)	3	腹膜内	四周一次持续 29 天	-	-	-	-	2 (8)	1	6	5
10	A10.21 (T106A)	3	腹膜内	四周一次持续 29 天	来那度胺	25	腹膜内	qd×21	0 (10)	0	10	10

- 对照组（媒剂）

研究终点 - 最开始 60 天或肿瘤体积超过 2000 mm³

[0121] MTV (n) - 研究结束时的中位肿瘤体积（用于计算的存活动物的数目）

PR - 部分消退的数目

CR - 完全消退的数目

TFS - 无肿瘤存活者的数目

[0122] 表5和图3展示次最佳剂量的与经灭毒的糖基化干扰素-α2b (T106A) 融合的抗CD38 抗体与来那度胺的组合的协同作用。来那度胺和与经灭毒的糖基化干扰素-α2b融合的抗 CD38抗体的组合实现有效抑制肿瘤生长所需的构筑体的剂量降低和给药间隔增加。当在单独使用时, 构筑体或来那度胺与媒剂对照物相比延缓肿瘤生长, 但肿瘤最终开始生长。相比之下, 经A10.21抗体灭毒的糖基化IFNα2b (T106A) 构筑体与来那度胺的组合在长时段内显示肿瘤生长抑制。此外, 在 (i) 所有每4周一次用3mg/kg的A10.21 (T106A) 与来那度胺的组合治疗持续29天的动物或 (ii) 所有每两周一次用1mg/kg的A10.21 (T106A) 与来那度胺的组合治疗持续29天的动物中实现第60天的无肿瘤存活。这些化合物的组合与单独的来那度胺相比, 卡普兰-迈耶存活曲线 (图4) 展示改良的第60天 (最长研究区间) 时的存活。因此, 这些化合物的组合有助于来那度胺和经抗CD-38灭毒的IFNα2b中的任一个或两个的不太频繁的给药和较低剂量水准投药。

[0123] 实例3

[0124] 泊利度胺研究

[0125] 进行这些实验以测定在雌性CB17SCID小鼠中的H929人类多发性骨髓瘤异种移植

模型中,与经灭毒的干扰素- α 2b融合的抗CD38抗体的非治愈性剂量疗法与泊利度胺的非治愈性剂量疗法的组合的功效。泊利度胺,与来那度胺类似,是撒利多胺的衍生物和类似物,其具有增加的针对多发性骨髓瘤的效能和降低的毒性。

[0126] 简单来说,将 1×10^7 个H929肿瘤细胞皮下注射到六十只雌性CB.17SCID小鼠的右侧腹中。在肿瘤达到 150mm^3 的平均体积时开始用泊利度胺和与经灭毒的干扰素至 α 2b融合的抗CD38抗体进行的治疗。研究在肿瘤体积达到 2000mm^3 时终止。群组划分如下,如表6中概述:第1组,媒剂(PBS);第2组,单独的泊利度胺(2.5mg/kg);第3组,经抗CD38灭毒的IFN α - (40微克/给药);第4组,抗同型-IFN α -灭毒(40微克/给药),第5组,泊利度胺(2.5mg/kg)加经抗CD38灭毒的IFN α (40微克/给药);以及第6组,泊利度胺(2.5mg/kg)加经抗同型灭毒的IFN α - (40微克/给药),泊利度胺投药在第1天开始并且在第21天终止;抗体-干扰素融合构筑体投药在第1天开始并且在第28天终止。

[0127] 表6. 分组、药物和治疗。

组号	N	方案 1				方案 2			
		药剂	mg/kg	途径	疗程	药剂	mg/kg	途径	疗程
[0128] [0129]	1	10	媒剂 (PBS)	40*	腹膜内	每两周一次 $\times 4$	-	-	-
	2	10	泊利度胺	2.5	腹膜内	qd $\times 21$	-	-	-
	3	10	经抗 CD38 灭毒的 IFN α 2b (h10A2-IFN-145D)	40*	腹膜内	每两周一次 $\times 4$	-	-	-
	4	10	同型对照物 (KLH-IFN-145D)	40*	腹膜内	每两周一次 $\times 4$	-	-	-
	5	10	泊利度胺	2.5	腹膜内	qd $\times 21$	经抗 CD38 灭毒的 IFN α 2b (h10A2-IFN-145D)	40*	腹膜内 每两周一次 $\times 4$
	6	10	泊利度胺	2.5	腹膜内	qd $\times 21$	同型对照物 (KLH-IFN-145D)	40*	腹膜内 每两周一次 $\times 4$

[0130] 40*=40微克剂量/小鼠,其为约2mg/kg

[0131] 单独的泊利度胺治疗在所使用的剂量下并未实质上减缓肿瘤生长。单独的经抗CD38灭毒的IFN α 2b治疗在研究持续时间内引起稳定的肿瘤收缩。10只小鼠中的七只展示最小肿瘤再生(图5)。相比之下,在用泊利度胺与经抗CD38灭毒的IFN α 2b的组合治疗的小鼠中,10只小鼠中仅4只展示最小再生,其中10只小鼠中的6只明显具有其肿瘤治愈。用泊利度胺与经无关同型对照抗体灭毒的IFN α 2b治疗的小鼠的肿瘤在约10天时段内稳定,但接着肿瘤开始生长,尽管速度略慢于媒剂对照物。

[0132] 本发明不限于上文描述和例示的实施例,而是能够在随附权利要求书的范畴内进行改变和变化。

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[0004]	<130> 185704-3020															
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[0032]	Val	Val	His	Val	Met	Leu	Asn	Gly	Ser	Arg	Ser	Lys	Ile	Phe	Asp	Lys
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[0110]	Cys Ala Trp Glu Val Val Arg Asp Glu Ile Met Arg Ser Phe Ser Leu
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[0135]	Tyr	Leu	Lys	Glu	Lys	Lys	Tyr	Ser	Pro	Cys	Ala	Trp	Glu	Val	Val	Arg
[0136]					130					135				140		
[0137]	Ala	Glu	Ile	Met	Arg	Ser	Phe	Ser	Leu	Ser	Thr	Asn	Leu	Gln	Glu	Ser
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[0218]	Leu Asp Lys Phe Tyr Thr Glu Leu Tyr Gln Gln Leu Asn Asp Leu Glu
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[0220]	Ala Cys Val Ile Gln Gly Val Gly Val Thr Glu Thr Pro Leu Met Lys
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[0257]	Ser Thr Asn Leu Gln Glu Ser Leu Arg Ser Lys Glu
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[0270]	Phe Pro Glu Pro Val Thr Val Ser Trp Asn Ser Gly Ala Leu Thr Ser
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[0278]	Arg Val Glu Ser Lys Tyr Gly Pro Pro Cys Pro Ser Cys Pro Ala Pro		
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[0290]	Trp Leu Asn Gly Lys Glu Tyr Lys Cys Lys Val Ser Asn Lys Gly Leu		
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[0292]	Pro Ser Ser Ile Glu Lys Thr Ile Ser Lys Ala Lys Gly Gln Pro Arg		
[0293]	210	215	220
[0294]	Glu Pro Gln Val Tyr Thr Leu Pro Pro Ser Gln Glu Glu Met Thr Lys		
[0295]	225	230	235
[0296]	Asn Gln Val Ser Leu Thr Cys Leu Val Lys Gly Phe Tyr Pro Ser Asp		
[0297]	245	250	255
[0298]	Ile Ala Val Glu Trp Glu Ser Asn Gly Gln Pro Glu Asn Asn Tyr Lys		
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[0300]	Thr Thr Pro Pro Val Leu Asp Ser Asp Gly Ser Phe Phe Leu Tyr Ser		
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[0302]	Arg Leu Thr Val Asp Lys Ser Arg Trp Gln Glu Gly Asn Val Phe Ser		
[0303]	290	295	300
[0304]	Cys Ser Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Lys Ser		
[0305]	305	310	315
[0306]	Leu Ser Leu Ser Leu Gly Lys Cys Asp Leu Pro Gln Thr His Ser Leu		
[0307]	325	330	335
[0308]	Gly Ser Arg Arg Thr Leu Met Leu Leu Ala Gln Met Arg Arg Ile Ser		
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[0314]	Glu Met Ile Gln Gln Ile Phe Asn Leu Phe Ser Thr Lys Asp Ser Ser
[0315]	385 390 395 400
[0316]	Ala Ala Trp Asp Glu Thr Leu Leu Asp Lys Phe Tyr Thr Glu Leu Tyr
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[0320]	Thr Glu Thr Pro Leu Met Lys Glu Asp Ser Ile Leu Ala Val Arg Lys
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[0324]	Cys Ala Trp Glu Val Val Arg Asp Glu Ile Met Arg Ser Phe Ser Leu
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[0342]	50 55 60
[0343]	Leu Ser Ser Val Val Thr Val Pro Ser Ser Ser Leu Gly Thr Lys Thr
[0344]	65 70 75 80
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[0347]	Arg Val Glu Ser Lys Tyr Gly Pro Pro Cys Pro Ser Cys Pro Ala Pro
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[0349]	Glu Phe Leu Gly Gly Pro Ser Val Phe Leu Phe Pro Pro Lys Pro Lys
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[0354]	145 150 155 160
[0355]	Gly Val Glu Val His Asn Ala Lys Thr Lys Pro Arg Glu Glu Gln Phe
[0356]	165 170 175
[0357]	Asn Ser Thr Tyr Arg Val Val Ser Val Leu Thr Val Leu His Gln Asp
[0358]	180 185 190
[0359]	Trp Leu Asn Gly Lys Glu Tyr Lys Cys Lys Val Ser Asn Lys Gly Leu
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[0361]	Pro Ser Ser Ile Glu Lys Thr Ile Ser Lys Ala Lys Gly Gln Pro Arg
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[0363]	Glu Pro Gln Val Tyr Thr Leu Pro Pro Ser Gln Glu Glu Met Thr Lys
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[0365]	Asn Gln Val Ser Leu Thr Cys Leu Val Lys Gly Phe Tyr Pro Ser Asp
[0366]	245 250 255
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[0370]	275 280 285
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[0373]	Cys Ser Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Lys Ser
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[0377]	Gly Ser Arg Arg Thr Leu Met Leu Leu Ala Gln Met Arg Arg Ile Ser
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[0382]	370 375 380
[0383]	Glu Met Ile Gln Gln Ile Phe Asn Leu Phe Ser Thr Lys Asp Ser Ser
[0384]	385 390 395 400
[0385]	Ala Ala Trp Asp Glu Thr Leu Leu Asp Lys Phe Tyr Thr Glu Leu Tyr
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[0406]	Ser Thr Ser Glu Ser Thr Ala Ala Leu Gly Cys Leu Val Lys Asp Tyr		
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[0415]	85 90 95		
[0416]	Arg Val Glu Ser Lys Tyr Gly Pro Pro Cys Pro Pro Cys Pro Ala Pro		
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[0420]	Asp Thr Leu Met Ile Ser Arg Thr Pro Glu Val Thr Cys Val Val Val		
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[0422]	Asp Val Ser Gln Glu Asp Pro Glu Val Gln Phe Asn Trp Tyr Val Asp		
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[0424]	Gly Val Glu Val His Asn Ala Lys Thr Lys Pro Arg Glu Glu Gln Phe		
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[0426]	Asn Ser Thr Tyr Arg Val Val Ser Val Leu Thr Val Leu His Gln Asp		
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[0434]	Asn Gln Val Ser Leu Thr Cys Leu Val Lys Gly Phe Tyr Pro Ser Asp		
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[0436]	Ile Ala Val Glu Trp Glu Ser Asn Gly Gln Pro Glu Asn Asn Tyr Lys		
[0437]	260	265	270
[0438]	Thr Thr Pro Pro Val Leu Asp Ser Asp Gly Ser Phe Phe Leu Tyr Ser		
[0439]	275	280	285
[0440]	Arg Leu Thr Val Asp Lys Ser Arg Trp Gln Glu Gly Asn Val Phe Ser		
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[0442]	Cys Ser Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Lys Ser		
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[0444]	Leu Ser Leu Ser Leu Gly Lys Cys Asp Leu Pro Gln Thr His Ser Leu		
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[0446]	Gly Ser Arg Arg Thr Leu Met Leu Leu Ala Gln Met Arg Arg Ile Ser		
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[0448]	Leu Phe Ser Cys Leu Lys Asp Arg His Asp Phe Gly Phe Pro Gln Glu		
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[0450]	Glu Phe Gly Asn Gln Phe Gln Lys Ala Glu Thr Ile Pro Val Leu His		
[0451]	370	375	380
[0452]	Glu Met Ile Gln Gln Ile Phe Asn Leu Phe Ser Thr Lys Asp Ser Ser		
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[0454]	Ala Ala Trp Asp Glu Thr Leu Leu Asp Lys Phe Tyr Thr Glu Leu Tyr		
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[0456]	Gln Gln Leu Asn Asp Leu Glu Ala Cys Val Ile Gln Gly Val Gly Val		
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[0458]	Thr Glu Thr Pro Leu Met Lys Glu Asp Ser Ile Leu Ala Val Arg Lys		
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[0460]	Tyr Phe Gln Arg Ile Thr Leu Tyr Leu Lys Glu Lys Lys Tyr Ser Pro		
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[0462]	Cys Ala Trp Glu Val Val Arg Gly Glu Ile Met Arg Ser Phe Ser Leu		
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[0479]	Gly	Val	His	Thr	Phe	Pro	Ala	Val	Leu	Gln	Ser	Ser	Gly	Leu	Tyr	Ser
[0480]		50					55					60				
[0481]	Leu	Ser	Ser	Val	Val	Thr	Val	Pro	Ser	Ser	Ser	Leu	Gly	Thr	Gln	Thr
[0482]	65					70				75					80	
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[0485]	Lys	Val	Glu	Pro	Lys	Ser	Cys	Asp	Lys	Thr	His	Thr	Cys	Pro	Pro	Cys
[0486]				100					105				110			
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[0488]				115				120					125			
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[0511]	Val Phe Ser Cys Ser Val Met His Glu Ala Leu His Asn His Tyr Thr		
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[0513]	Gln Lys Ser Leu Ser Leu Ser Pro Gly Lys Cys Asp Leu Pro Gln Thr		
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[0515]	His Ser Leu Gly Ser Arg Arg Thr Leu Met Leu Leu Ala Gln Met Arg		
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[0517]	Arg Ile Ser Leu Phe Ser Cys Leu Lys Asp Arg His Asp Phe Gly Phe		
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[0519]	Pro Gln Glu Glu Phe Gly Asn Gln Phe Gln Lys Ala Glu Thr Ile Pro		
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[0523]	Asp Ser Ser Ala Ala Trp Asp Glu Thr Leu Leu Asp Lys Phe Tyr Thr		
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[0525]	Glu Leu Tyr Gln Gln Leu Asn Asp Leu Glu Ala Cys Val Ile Gln Gly		
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[0527]	Val Gly Val Thr Glu Thr Pro Leu Met Lys Glu Asp Ser Ile Leu Ala		
[0528]	435	440	445
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[0544]	Ser Thr Ser Gly Gly Thr Ala Ala Leu Gly Cys Leu Val Lys Asp Tyr		
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[0633]	Glu Gln Tyr Asn Ser Thr Tyr Arg Val Val Ser Val Leu Thr Val Leu		
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[0635]	His Gln Asp Trp Leu Asn Gly Lys Glu Tyr Lys Cys Lys Val Ser Asn		
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[0643]	Pro Ser Asp Ile Ala Val Glu Trp Glu Ser Asn Gly Gln Pro Glu Asn		
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[0645]	Asn Tyr Lys Thr Thr Pro Pro Val Leu Asp Ser Asp Gly Ser Phe Phe		
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[0647]	Leu Tyr Ser Lys Leu Thr Val Asp Lys Ser Arg Trp Gln Gln Gly Asn		
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[0649]	Val Phe Ser Cys Ser Val Met His Glu Ala Leu His Asn His Tyr Thr		
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[0651]	Gln Lys Ser Leu Ser Leu Ser Pro Gly Lys Cys Asp Leu Pro Gln Thr		
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[0653]	His Ser Leu Gly Ser Arg Arg Thr Leu Met Leu Leu Ala Gln Met Arg		
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[0657]	Pro Gln Glu Glu Phe Gly Asn Gln Phe Gln Lys Ala Glu Thr Ile Pro		
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[0659]	Val Leu His Glu Met Ile Gln Gln Ile Phe Asn Leu Phe Ser Thr Lys		
[0660]	385	390	395
[0661]	Asp Ser Ser Ala Ala Trp Asp Glu Thr Leu Leu Asp Lys Phe Tyr Thr		
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[0663]	Glu Leu Tyr Gln Gln Leu Asn Asp Leu Glu Ala Cys Val Ile Gln Gly
[0664]	420 425 430
[0665]	Val Gly Val Thr Glu Thr Pro Leu Met Lys Glu Asp Ser Ile Leu Ala
[0666]	435 440 445
[0667]	Val Arg Lys Tyr Phe Gln Arg Ile Thr Leu Tyr Leu Lys Glu Lys Lys
[0668]	450 455 460
[0669]	Tyr Ser Pro Cys Ala Trp Glu Val Val Arg Gly Glu Ile Met Arg Ser
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[0690]	Tyr Thr Cys Asn Val Asp His Lys Pro Ser Asn Thr Lys Val Asp Lys
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[0698]	Asp Val Ser Gln Glu Asp Pro Glu Val Gln Phe Asn Trp Tyr Val Asp
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[0702]	Asn Ser Thr Tyr Arg Val Val Ser Val Leu Thr Val Leu His Gln Asp
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[0711]	245 250 255
[0712]	Ile Ala Val Glu Trp Glu Ser Asn Gly Gln Pro Glu Asn Asn Tyr Lys
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[0729]	385 390 395 400
[0730]	Ala Ala Trp Asp Glu Thr Leu Leu Asp Lys Phe Tyr Thr Glu Leu Tyr
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[0759]	Tyr Thr Cys Asn Val Asp His Lys Pro Ser Asn Thr Lys Val Asp Lys			
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[0854]	Cys	Ala	Arg	Val	Gly	Gly	Ala	Gly	Gly	Trp	Pro	Xaa	Asp	Val	Trp	Gly		
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[0874]	<222> (20) .. (20)
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[0876]	<220>
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[0878]	<222> (29) .. (29)
[0879]	<223> X = Phe 或 Leu
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[0882]	<222> (38) .. (38)
[0883]	<223> X = Gln 或 Arg
[0884]	<220>
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[0886]	<222> (40) .. (40)
[0887]	<223> X = Glu 或 Gly 或 His 或 Ala
[0888]	<220>
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[0890]	<222> (62) .. (62)
[0891]	<223> X = Glu 或 Gln
[0892]	<220>
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[0894]	<222> (70) .. (70)
[0895]	<223> X = Ile 或 Met
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[0899]	<223>	X = Lys 或 Gly 或 Thr
[0900]	<220>	
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[0909]	Val Met Asn Trp Val Xaa Gln Xaa Pro Gly Lys Gly Leu Glu Trp Met	
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[0911]	Gly Trp Ile Asp Pro Glu Tyr Gly Arg Thr Asp Val Ala Xaa Lys Phe	
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[0913]	Gln Gly Arg Val Thr Xaa Thr Ala Asp Thr Ser Thr Asp Thr Ala Tyr	
[0914]	65	70 75 80
[0915]	Met Glu Leu Ser Ser Leu Arg Ser Glu Asp Thr Ala Val Tyr Tyr Cys	
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[0917]	Ala Arg Thr Xaa Tyr Xaa Ser Gly Tyr Gly Phe Pro Tyr Trp Gly Gln	
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[0919]	Gly Thr Thr Val Thr Val Ser Ser	
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[0930]	<223>	X = Ala 或 Pro
[0931]	<220>	
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[0942]	<223>	X = Met 或 Leu
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[0949]		35 40 45
[0950]		Gly Trp Ile Asn Pro Asn Asn Gly Gly Xaa Thr Phe Ala Gln Lys Phe
[0951]		50 55 60
[0952]		Gln Gly Arg Val Thr Met Thr Arg Asp Thr Ser Ile Ser Thr Ala Tyr
[0953]		65 70 75 80
[0954]		Met Asp Leu Ser Ser Leu Arg Ser Asp Asp Thr Ala Val Tyr Xaa Cys
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[0970]	<220>	
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[0993]	Xaa Val Lys Xaa Ser Cys Lys Val Ser Gly Tyr Thr Xaa Thr Asp Ser																
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[0995]	Val Met Asn Trp Val Xaa Gln Xaa Pro Gly Lys Gly Leu Glu Trp Met																
[0996]	35 40 45																
[0997]	Gly Trp Ile Asp Pro Glu Tyr Gly Arg Thr Asp Val Ala Glu Lys Phe																
[0998]	50 55 60																
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[1001]	Met Glu Leu Ser Ser Leu Arg Ser Glu Asp Thr Ala Val Tyr Tyr Cys																
[1002]	85 90 95																
[1003]	Ala Arg Thr Lys Tyr Asn Ser Gly Tyr Gly Phe Pro Tyr Trp Gly Gln																
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[1015]	<222> (31) .. (31)
[1016]	<223> X = Arg 或 Gly
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[1019]	<222> (32) .. (32)
[1020]	<223> X = Tyr 或 Ser
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[1023]	<222> (54) .. (54)
[1024]	<223> X = Ser 或 Glu 或 Gln
[1025]	<220>
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[1028]	<223> X = Asn 或 Thr
[1029]	<220>
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[1031]	<222> (97) .. (97)
[1032]	<223> X = Met 或 Leu
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[1035]	<222> (102) .. (102)
[1036]	<223> X = Asn 或 Gln 或 Glu
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[1040]	Ser Ala Arg Leu Thr Cys Thr Leu Pro Ser Asp Ile Asn Val Xaa Xaa
[1041]	20 25 30
[1042]	Tyr Asn Ile Tyr Trp Tyr Gln Gln Lys Pro Gly Ser Pro Pro Arg Tyr
[1043]	35 40 45
[1044]	Leu Leu Tyr Tyr Tyr Xaa Asp Ser His Lys Gly Gln Gly Ser Gly Val
[1045]	50 55 60
[1046]	Pro Ser Arg Phe Ser Gly Ser Lys Asp Val Ser Thr Xaa Ser Gly Ile
[1047]	65 70 75 80
[1048]	Leu Leu Ile Ser Gly Leu Gln Ser Glu Asp Ile Ala Thr Tyr Tyr Cys
[1049]	85 90 95
[1050]	Xaa Thr Trp Ser Ser Xaa Gly Ser Gly Val Phe Gly Gly Gly Thr Gln
[1051]	100 105 110
[1052]	Leu Thr Val Leu Gly

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[1063]	<223> X = Lys 或 Gly 或 Gln
[1064]	<220>
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[1066]	<222> (53) .. (53)
[1067]	<223> X = Asn 或 Gln
[1068]	<220>
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[1070]	<222> (54) .. (54)
[1071]	<223> X = Arg 或 Asp
[1072]	<220>
[1073]	<221> 变异体
[1074]	<222> (89) .. (89)
[1075]	<223> X = Met 或 Ala
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[1078]	1 5 10 15
[1079]	Asp Arg Val Thr Ile Thr Cys Xaa Ala Ser Gln Asn Val Asp Ser Asp
[1080]	20 25 30
[1081]	Val Asp Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile
[1082]	35 40 45
[1083]	Tyr Lys Ala Ser Xaa Xaa Tyr Thr Gly Val Pro Ser Arg Phe Ser Gly
[1084]	50 55 60
[1085]	Ser Gly Ser Gly Thr Asp Phe Thr Phe Thr Ile Ser Ser Leu Gln Pro
[1086]	65 70 75 80
[1087]	Glu Asp Ile Ala Thr Tyr Tyr Cys Xaa Gln Ser Asn Thr His Pro Arg
[1088]	85 90 95
[1089]	Thr Phe Gly Gly Gly Thr Lys Val Glu Ile Lys Arg
[1090]	100 105
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[1099]	<222>	(24) .. (24)
[1100]	<223>	X1 = K 或 Q
[1101]	<220>	
[1102]	<221>	变异体
[1103]	<222>	(89) .. (89)
[1104]	<223>	X2 = A 或 M
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[1107]	1 5 10 15	
[1108]	Asp Arg Val Thr Ile Thr Cys Xaa Ala Ser Gln Asn Val Asp Ser Asp	
[1109]	20 25 30	
[1110]	Val Asp Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile	
[1111]	35 40 45	
[1112]	Tyr Lys Ala Ser Asn Asp Tyr Thr Gly Val Pro Ser Arg Phe Ser Gly	
[1113]	50 55 60	
[1114]	Ser Gly Ser Gly Thr Asp Phe Thr Phe Thr Ile Ser Ser Leu Gln Pro	
[1115]	65 70 75 80	
[1116]	Glu Asp Ile Ala Thr Tyr Tyr Cys Xaa Gln Ser Asn Thr His Pro Arg	
[1117]	85 90 95	
[1118]	Thr Phe Gly Gly Gly Thr Lys Val Glu Ile Lys Arg	
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[1124]	<400>	24
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[1126]	1 5 10 15	
[1127]	Thr Leu Ser Leu Thr Cys Thr Val Ser Gly Gly Ser Ile Ser Ser Ser	
[1128]	20 25 30	
[1129]	Ser Tyr Tyr Trp Ser Trp Ile Arg Gln His Pro Gly Lys Gly Leu Glu	
[1130]	35 40 45	

[1131] Trp Ile Gly Tyr Ile Tyr Tyr Ser Gly Ser Thr Asn Tyr Asn Pro Ser
 [1132] 50 55 60
 [1133] Leu Lys Ser Arg Val Thr Ile Ser Val Asp Thr Leu Lys Asn Gln Ile
 [1134] 65 70 75 80
 [1135] Ser Leu Arg Leu Thr Ser Val Thr Ala Ala Asp Thr Ala Val Tyr Tyr
 [1136] 85 90 95
 [1137] Cys Ala Arg Val Gly Gly Ala Gly Gly Trp Pro Met Asp Val Trp Gly
 [1138] 100 105 110
 [1139] Gln Gly Thr Thr Val Thr Val Ser Ser
 [1140] 115 120
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 [1145] <400> 25
 [1146] Gln Ala Val Leu Thr Gln Pro Ala Ser Leu Ser Ala Ser Pro Gly Glu
 [1147] 1 5 10 15
 [1148] Ser Ala Arg Leu Thr Cys Thr Leu Pro Ser Asp Ile Asn Val Arg Tyr
 [1149] 20 25 30
 [1150] Tyr Asn Ile Tyr Trp Tyr Gln Gln Lys Pro Gly Ser Pro Pro Arg Tyr
 [1151] 35 40 45
 [1152] Leu Leu Tyr Tyr Tyr Ser Asp Ser His Lys Gly Gln Gly Ser Gly Val
 [1153] 50 55 60
 [1154] Pro Ser Arg Phe Ser Gly Ser Lys Asp Val Ser Thr Asn Ser Gly Ile
 [1155] 65 70 75 80
 [1156] Leu Leu Ile Ser Gly Leu Gln Ser Glu Asp Glu Ala Asp Tyr Tyr Cys
 [1157] 85 90 95
 [1158] Met Thr Trp Ser Ser Asn Gly Ser Gly Val Phe Gly Gly Gly Thr Gln
 [1159] 100 105 110
 [1160] Leu Thr Val Leu Gly
 [1161] 115
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 [1168] <400> 26
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[1171]	Ser Val Lys Val Ser Cys Lys Val Ser Gly Tyr Thr Leu Thr Asp Ser			
[1172]	20	25	30	
[1173]	Val Met Asn Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Met			
[1174]	35	40	45	
[1175]	Gly Trp Ile Asp Pro Glu Tyr Gly Arg Thr Asp Val Ala Gln Lys Phe			
[1176]	50	55	60	
[1177]	Gln Gly Arg Val Thr Met Thr Ala Asp Thr Ser Thr Asp Thr Ala Tyr			
[1178]	65	70	75	80
[1179]	Met Glu Leu Ser Ser Leu Arg Ser Glu Asp Thr Ala Val Tyr Tyr Cys			
[1180]	85	90	95	
[1181]	Ala Arg Thr Lys Tyr Asn Ser Gly Tyr Gly Phe Pro Tyr Trp Gly Gln			
[1182]	100	105	110	
[1183]	Gly Thr Thr Val Thr Val Ser Ser			
[1184]	115	120		
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[1194]	Thr Val Lys Ile Ser Cys Lys Val Ser Gly Tyr Thr Phe Thr Asp Ser			
[1195]	20	25	30	
[1196]	Val Met Asn Trp Val Gln Gln Ala Pro Gly Lys Gly Leu Glu Trp Met			
[1197]	35	40	45	
[1198]	Gly Trp Ile Asp Pro Glu Tyr Gly Arg Thr Asp Val Ala Glu Lys Phe			
[1199]	50	55	60	
[1200]	Gln Gly Arg Val Thr Ile Thr Ala Asp Thr Ser Thr Asp Thr Ala Tyr			
[1201]	65	70	75	80
[1202]	Met Glu Leu Ser Ser Leu Arg Ser Glu Asp Thr Ala Val Tyr Tyr Cys			
[1203]	85	90	95	
[1204]	Ala Arg Thr Lys Tyr Asn Ser Gly Tyr Gly Phe Pro Tyr Trp Gly Gln			
[1205]	100	105	110	
[1206]	Gly Thr Thr Val Thr Val Ser Ser			
[1207]	115	120		
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[1217]	Thr Val Lys Ile Ser Cys Lys Val Ser Gly Tyr Thr Phe Thr Asp Ser	
[1218]	20	25 30
[1219]	Val Met Asn Trp Val Gln Gln His Pro Gly Lys Gly Leu Glu Trp Met	
[1220]	35	40 45
[1221]	Gly Trp Ile Asp Pro Glu Tyr Gly Arg Thr Asp Val Ala Glu Lys Phe	
[1222]	50	55 60
[1223]	Gln Gly Arg Val Thr Ile Thr Ala Asp Thr Ser Thr Asp Thr Ala Tyr	
[1224]	65	70 75 80
[1225]	Met Glu Leu Ser Ser Leu Arg Ser Glu Asp Thr Ala Val Tyr Tyr Cys	
[1226]	85	90 95
[1227]	Ala Arg Thr Lys Tyr Asn Ser Gly Tyr Gly Phe Pro Tyr Trp Gly Gln	
[1228]	100	105 110
[1229]	Gly Thr Thr Val Thr Val Ser Ser	
[1230]	115	120
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[1238]	Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly	
[1239]	1	5 10 15
[1240]	Asp Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Asn Val Asp Ser Asp	
[1241]	20	25 30
[1242]	Val Asp Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile	
[1243]	35	40 45
[1244]	Tyr Lys Ala Ser Asn Asp Tyr Thr Gly Val Pro Ser Arg Phe Ser Gly	
[1245]	50	55 60
[1246]	Ser Gly Ser Gly Thr Asp Phe Thr Phe Thr Ile Ser Ser Leu Gln Pro	
[1247]	65	70 75 80

[1248]	Glu Asp Ile Ala Thr Tyr Tyr Cys Met Gln Ser Asn Thr His Pro Arg
[1249]	85 90 95
[1250]	Thr Phe Gly Gly Gly Thr Lys Val Glu Ile Lys Arg
[1251]	100 105
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[1259]	Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly
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[1261]	Asp Arg Val Thr Ile Thr Cys Gln Ala Ser Gln Asn Val Asp Ser Asp
[1262]	20 25 30
[1263]	Val Asp Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile
[1264]	35 40 45
[1265]	Tyr Lys Ala Ser Asn Asp Tyr Thr Gly Val Pro Ser Arg Phe Ser Gly
[1266]	50 55 60
[1267]	Ser Gly Ser Gly Thr Asp Phe Thr Phe Thr Ile Ser Ser Leu Gln Pro
[1268]	65 70 75 80
[1269]	Glu Asp Ile Ala Thr Tyr Tyr Cys Ala Gln Ser Asn Thr His Pro Arg
[1270]	85 90 95
[1271]	Thr Phe Gly Gly Gly Thr Lys Val Glu Ile Lys Arg
[1272]	100 105
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[1279]	<400> 31
[1280]	Gln Val Gln Leu Gln Glu Ser Gly Pro Gly Leu Val Lys Pro Ser Glu
[1281]	1 5 10 15
[1282]	Thr Leu Ser Leu Thr Cys Thr Val Ser Gly Gly Ser Ile Ser Ser Ser
[1283]	20 25 30
[1284]	Ser Tyr Tyr Trp Ser Trp Ile Arg Gln His Pro Gly Lys Gly Leu Glu
[1285]	35 40 45
[1286]	Trp Ile Gly Tyr Ile Tyr Tyr Ser Gly Ser Thr Asn Tyr Asn Pro Ser

[1287]	50	55	60
[1288]	Leu Lys Ser Arg Val Thr Ile Ser Val Asp Thr Leu Lys Asn Gln Ile		
[1289]	65	70	75
[1290]	Ser Leu Arg Leu Thr Ser Val Thr Ala Ala Asp Thr Ala Val Tyr Tyr		
[1291]	85	90	95
[1292]	Cys Ala Arg Val Gly Gly Ala Gly Gly Trp Pro Met Asp Val Trp Gly		
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[1294]	Gln Gly Thr Thr Val Thr Val Ser Ser		
[1295]	115	120	
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[1305]	Thr Leu Ser Leu Thr Cys Thr Val Ser Gly Gly Ser Ile Ser Ser Ser		
[1306]	20	25	30
[1307]	Ser Tyr Tyr Trp Ser Trp Ile Arg Gln His Pro Gly Lys Gly Leu Glu		
[1308]	35	40	45
[1309]	Trp Ile Gly Tyr Ile Tyr Tyr Ser Gly Ser Thr Asn Tyr Asn Pro Ser		
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[1311]	Leu Lys Ser Arg Val Thr Ile Ser Arg Asp Thr Leu Lys Asn Gln Ile		
[1312]	65	70	75
[1313]	Ser Leu Arg Leu Thr Ser Val Thr Ala Ala Asp Thr Ala Val Tyr Tyr		
[1314]	85	90	95
[1315]	Cys Ala Arg Val Gly Gly Ala Gly Gly Trp Pro Met Asp Val Trp Gly		
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[1317]	Gln Gly Thr Thr Val Thr Val Ser Ser		
[1318]	115	120	
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[1329]				20						25					30	
[1330]	Ser	Tyr	Tyr	Trp	Ser	Trp	Ile	Arg	Gln	His	Pro	Gly	Lys	Gly	Leu	Glu
[1331]			35					40					45			
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[1333]		50					55					60				
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[1335]	65					70					75					80
[1336]	Ser	Leu	Arg	Leu	Thr	Ser	Val	Thr	Ala	Ala	Asp	Thr	Ala	Val	Tyr	Tyr
[1337]					85					90					95	
[1338]	Cys	Ala	Arg	Val	Gly	Gly	Ala	Gly	Gly	Trp	Pro	Met	Asp	Val	Trp	Gly
[1339]				100					105					110		
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[1341]			115					120								
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[1347]	<223> 完全合成.															
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[1352]				20						25					30	
[1353]	Ser	Tyr	Tyr	Trp	Ser	Trp	Ile	Arg	Gln	His	Pro	Gly	Lys	Gly	Leu	Glu
[1354]			35					40					45			
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[1356]		50					55					60				
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[1358]	65					70					75					80
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[1360]					85					90					95	
[1361]	Cys	Ala	Arg	Val	Gly	Gly	Ala	Gly	Gly	Trp	Pro	Met	Asp	Val	Trp	Gly
[1362]				100					105					110		
[1363]	Gln	Gly	Thr	Thr	Val	Thr	Val	Ser	Ser							
[1364]			115					120								

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[1374]	Thr Leu Lys Leu Thr Cys Thr Val Ser Gly Gly Ser Ile Ser Ser Ser	
[1375]	20	25 30
[1376]	Ser Tyr Tyr Trp Ser Trp Ile Arg Gln His Pro Gly Lys Gly Leu Glu	
[1377]	35	40 45
[1378]	Trp Ile Gly Tyr Ile Tyr Tyr Ser Gly Ser Thr Asn Tyr Asn Pro Ser	
[1379]	50	55 60
[1380]	Leu Lys Ser Arg Val Thr Ile Ser Val Asp Thr Leu Lys Asn Gln Ile	
[1381]	65	70 75 80
[1382]	Ser Leu Arg Leu Thr Ser Val Thr Ala Ala Asp Thr Ala Val Tyr Tyr	
[1383]	85	90 95
[1384]	Cys Ala Arg Val Gly Gly Ala Gly Gly Trp Pro Met Asp Val Trp Gly	
[1385]	100	105 110
[1386]	Gln Gly Thr Thr Val Thr Val Ser Ser	
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[1394]	<400>	36
[1395]	Gln Leu Gln Leu Gln Glu Ser Gly Pro Gly Leu Val Lys Pro Ser Glu	
[1396]	1	5 10 15
[1397]	Thr Leu Ser Leu Thr Cys Thr Val Ser Gly Gly Ser Ile Ser Ser Ser	
[1398]	20	25 30
[1399]	Ser Tyr Tyr Trp Ser Trp Ile Arg Gln His Pro Gly Lys Gly Leu Glu	
[1400]	35	40 45
[1401]	Trp Ile Gly Tyr Ile Tyr Tyr Ser Gly Ser Thr Asn Tyr Asn Pro Ser	
[1402]	50	55 60
[1403]	Leu Lys Ser Arg Val Thr Ile Ser Val Asp Thr Leu Lys Asn Gln Ile	

[1404]	65	70	75	80
[1405]	Ser Leu Arg Leu Thr Ser Val Thr Ala Ala Asp Thr Ala Val Tyr Tyr			
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[1607]	35 40 45
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[1836]	Leu Leu Tyr Tyr Tyr Ser Asp Ser His Lys Gly Gln Gly Ser Gly Val		
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[1840]	Ile Ser Gly Leu Gln Ser Glu Asp Glu Ala Asp Tyr Tyr Cys Met Thr		
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[1842]	Trp Ser Ser Asn Gly Ser Gly Val Phe Gly Gly Gly Thr Gln Leu Thr		
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[1857]	Tyr Asn Ile Tyr Trp Tyr Gln Gln Lys Pro Gly Ser Pro Pro Arg Tyr		
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[1863]	Leu Leu Ile Ser Gly Leu Gln Ser Glu Asp Glu Ala Asp Tyr Tyr Cys		
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[1865]	Met Thr Trp Ser Ser Asn Gly Ser Gly Val Phe Gly Gly Gly Thr Gln		
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[1924]	Ser Ala Arg Leu Thr Cys Thr Leu Pro Ser Asp Ile Asn Val Arg Tyr
[1925]	20 25 30
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[1927]	35 40 45
[1928]	Leu Leu Tyr Tyr Arg Ser Asp Ser His Lys Gly Gln Gly Ser Gly Val
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[1953]	Pro Ser Arg Phe Ser Gly Ser Lys Asp Val Ser Thr Asn Ser Gly Ile		
[1954]	65	70	75
[1955]	Leu Leu Ile Ser Gly Leu Gln Ser Glu Asp Glu Ala Asp Tyr Tyr Cys		
[1956]	85	90	95
[1957]	Met Thr Trp Ser Ser Thr Gly Ser Gly Val Phe Gly Gly Gly Thr Gln		
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[1972]	Tyr Asn Ile Tyr Trp Tyr Gln Gln Lys Pro Gly Ser Pro Pro Arg Tyr		
[1973]	35	40	45
[1974]	Leu Leu Tyr Tyr Tyr Ser Asp Ser His Lys Gly Gln Gly Ser Gly Val		
[1975]	50	55	60
[1976]	Pro Ser Arg Phe Ser Gly Ser Lys Asp Val Ser Thr Asn Ser Gly Ile		
[1977]	65	70	75
[1978]	Leu Leu Ile Ser Gly Leu Gln Ser Glu Asp Glu Ala Asp Tyr Tyr Cys		
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[1996]				35					40					45		
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[1999]	Pro	Ser	Arg	Phe	Ser	Gly	Ser	Lys	Asp	Val	Ser	Thr	Asn	Ser	Gly	Ile
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[2017]				20					25					30		
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[2023]	65					70					75				80	
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[2039]	Ser Ala Arg Leu Thr Cys Thr Leu Pro Ser Asp Ile Asn Val Arg Tyr
[2040]	20 25 30
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[2042]	35 40 45
[2043]	Leu Leu Tyr Tyr Tyr Ser Asp Ser His Lys Gly Gln Gly Ser Gly Val
[2044]	50 55 60
[2045]	Pro Ser Arg Phe Ser Gly Ser Lys Asp Val Ser Thr Asn Ser Gly Ile
[2046]	65 70 75 80
[2047]	Leu Leu Ile Ser Gly Leu Gln Ser Glu Asp Glu Ala Asp Tyr Tyr Cys
[2048]	85 90 95
[2049]	Leu Thr Trp Ser Ser Asn Gly Ser Gly Val Phe Gly Gly Gly Thr Gln
[2050]	100 105 110
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[2062]	Ser Ala Arg Leu Thr Cys Thr Leu Pro Ser Asp Ile Asn Val Arg Tyr
[2063]	20 25 30
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[2065]	35 40 45
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[2089]	Leu Leu Tyr Tyr Tyr Ser Asp Ser His Lys Gly Gln Gly Ser Gly Val		45
[2090]	50	55	60
[2091]	Pro Ser Arg Phe Ser Gly Ser Lys Asp Val Ser Thr Asn Ser Gly Ile		
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[2093]	Leu Leu Ile Ser Gly Leu Gln Ser Glu Asp Glu Ala Asp Tyr Tyr Cys		80
[2094]		85	90
[2095]	Met Thr Trp Ser Ser Asn Gly Ser Gly Val Phe Gly Gly Gly Thr Gln		95
[2096]		100	105
[2097]	Leu Thr Val Leu Gly		110
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[2114]	Pro	Ser	Arg	Phe	Ser	Gly	Ser	Lys	Asp	Val	Ser	Thr	Asn	Ser	Gly	Ile
[2115]	65					70					75				80	
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[2157]	35 40 45
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[2159]	50 55 60
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[2164]	Met Thr Trp Ser Ser Asn Gly Ser Gly Val Phe Gly Gly Gly Thr Gln
[2165]	100 105 110
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[2176]	1 5 10 15
[2177]	Ser Ala Arg Leu Thr Cys Thr Leu Pro Ser Asp Ile Asn Val Arg Tyr
[2178]	20 25 30
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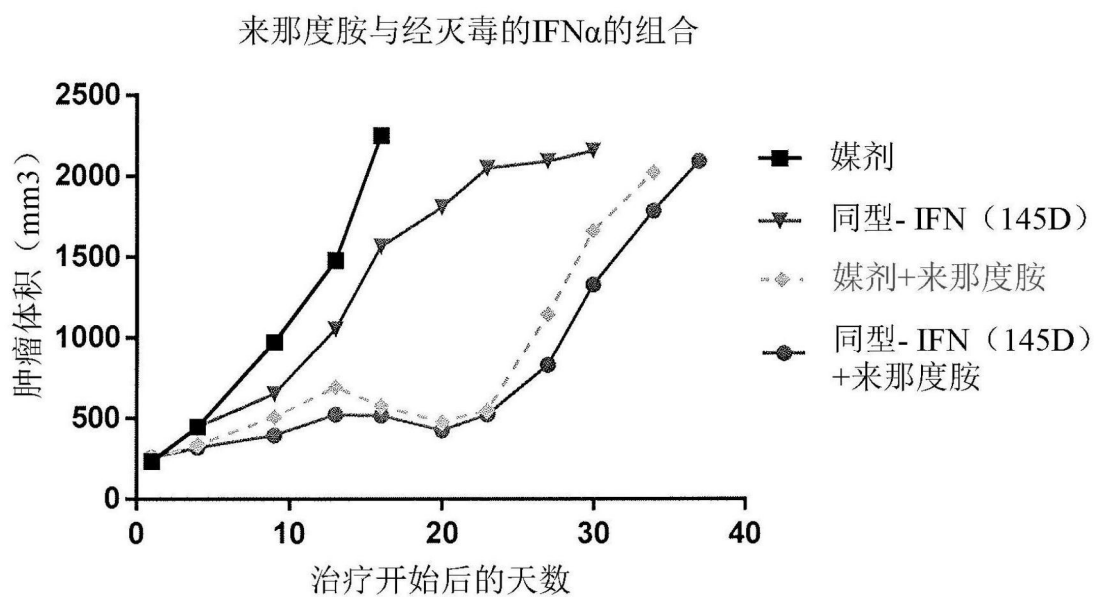
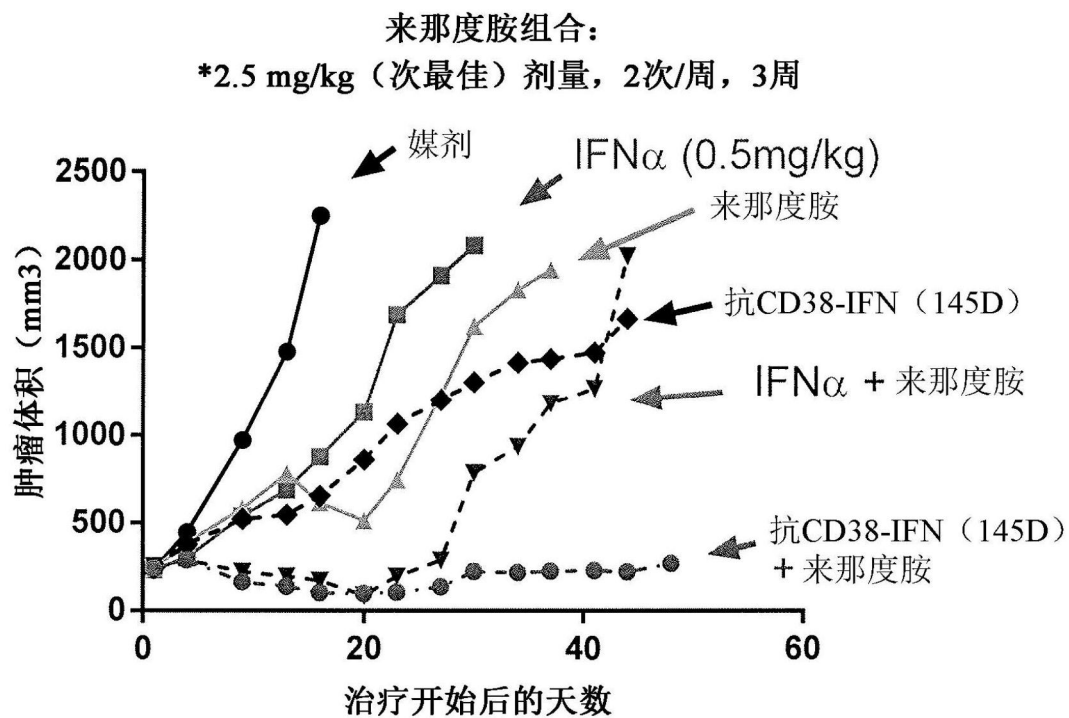
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[5610]	Tyr Gln Gln Leu Asn Asp Leu Glu Ala Cys Val Ile Gln Gly Val Gly		
[5611]	565	570	575
[5612]	Val Ala Glu Thr Pro Leu Met Lys Glu Asp Ser Ile Leu Ala Val Arg		
[5613]	580	585	590
[5614]	Lys Tyr Phe Gln Arg Ile Thr Leu Tyr Leu Lys Glu Lys Lys Tyr Ser		
[5615]	595	600	605

[5616]	Pro	Cys	Ala	Trp	Glu	Val	Val	Arg	Asp	Glu	Ile	Met	Arg	Ser	Phe	Ser
[5617]	610							615				620				
[5618]	Leu	Ser	Thr	Asn	Leu	Gln	Glu	Ser	Leu	Arg	Ser	Lys	Glu			
[5619]	625							630				635				
[5620]	<210>	217														
[5621]	<211>	214														
[5622]	<212>	PRT														
[5623]	<213>	智人														
[5624]	<400>	217														
[5625]	Asp	Ile	Gln	Met	Thr	Gln	Ser	Pro	Ser	Ser	Leu	Ser	Ala	Ser	Val	Gly
[5626]	1				5					10					15	
[5627]	Asp	Arg	Val	Thr	Ile	Thr	Cys	Lys	Ala	Ser	Gln	Asn	Val	Asp	Ser	Asp
[5628]				20					25					30		
[5629]	Val	Asp	Trp	Tyr	Gln	Gln	Lys	Pro	Gly	Lys	Ala	Pro	Lys	Leu	Leu	Ile
[5630]			35					40					45			
[5631]	Tyr	Lys	Ala	Ser	Asn	Asp	Tyr	Thr	Gly	Val	Pro	Ser	Arg	Phe	Ser	Gly
[5632]		50					55					60				
[5633]	Ser	Gly	Ser	Gly	Thr	Asp	Phe	Thr	Phe	Thr	Ile	Ser	Ser	Leu	Gln	Pro
[5634]	65					70					75				80	
[5635]	Glu	Asp	Ile	Ala	Thr	Tyr	Tyr	Cys	Met	Gln	Ser	Asn	Thr	His	Pro	Arg
[5636]					85					90				95		
[5637]	Thr	Phe	Gly	Gly	Gly	Thr	Lys	Val	Glu	Ile	Lys	Arg	Thr	Val	Ala	Ala
[5638]				100					105					110		
[5639]	Pro	Ser	Val	Phe	Ile	Phe	Pro	Pro	Ser	Asp	Glu	Gln	Leu	Lys	Ser	Gly
[5640]		115						120					125			
[5641]	Thr	Ala	Ser	Val	Val	Cys	Leu	Leu	Asn	Asn	Phe	Tyr	Pro	Arg	Glu	Ala
[5642]		130					135					140				
[5643]	Lys	Val	Gln	Trp	Lys	Val	Asp	Asn	Ala	Leu	Gln	Ser	Gly	Asn	Ser	Gln
[5644]	145					150					155				160	
[5645]	Glu	Ser	Val	Thr	Glu	Gln	Asp	Ser	Lys	Asp	Ser	Thr	Tyr	Ser	Leu	Ser
[5646]					165					170					175	
[5647]	Ser	Thr	Leu	Thr	Leu	Ser	Lys	Ala	Asp	Tyr	Glu	Lys	His	Lys	Val	Tyr
[5648]				180					185					190		
[5649]	Ala	Cys	Glu	Val	Thr	His	Gln	Gly	Leu	Ser	Ser	Pro	Val	Thr	Lys	Ser
[5650]			195					200					205			
[5651]	Phe	Asn	Arg	Gly	Glu	Cys										
[5652]		210														



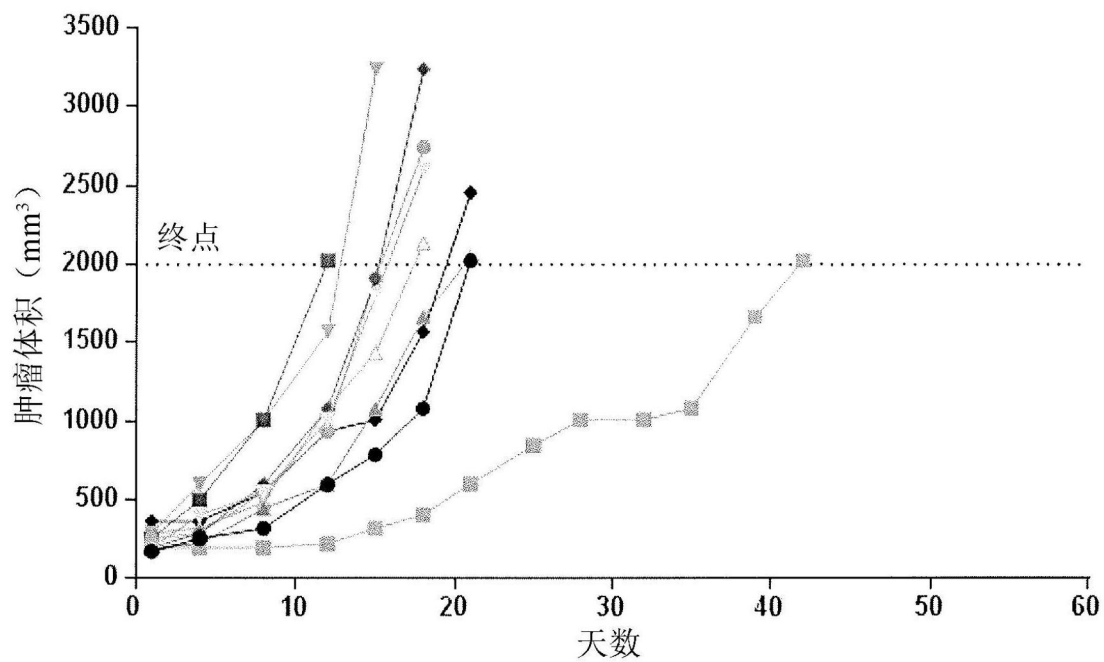


图3A

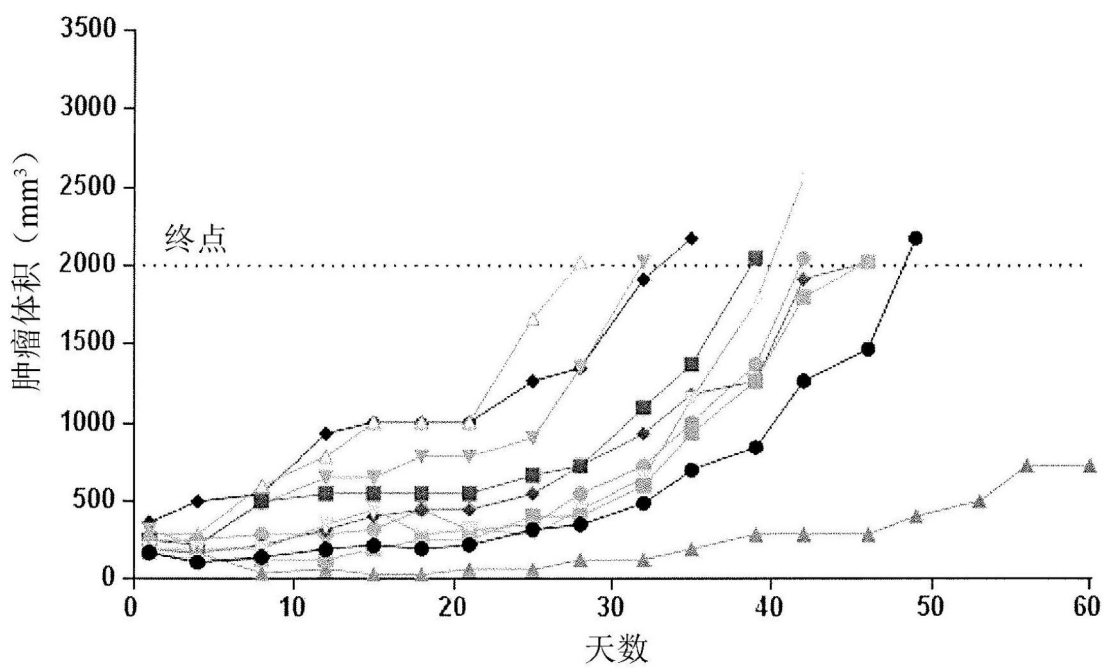


图3B

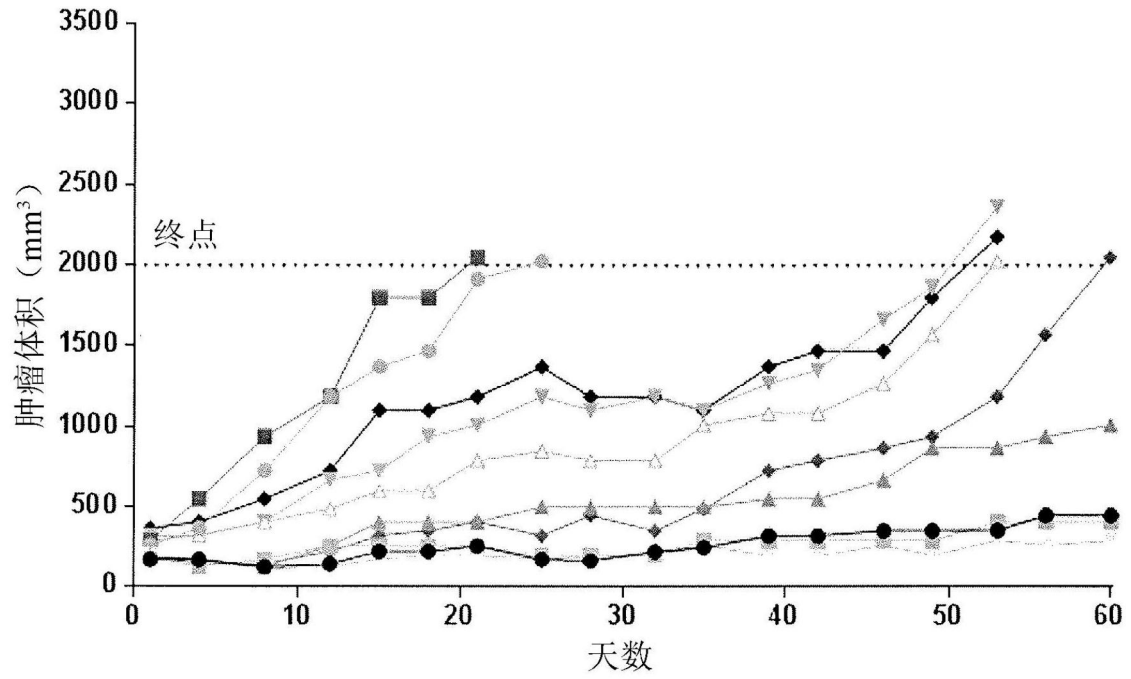


图3C

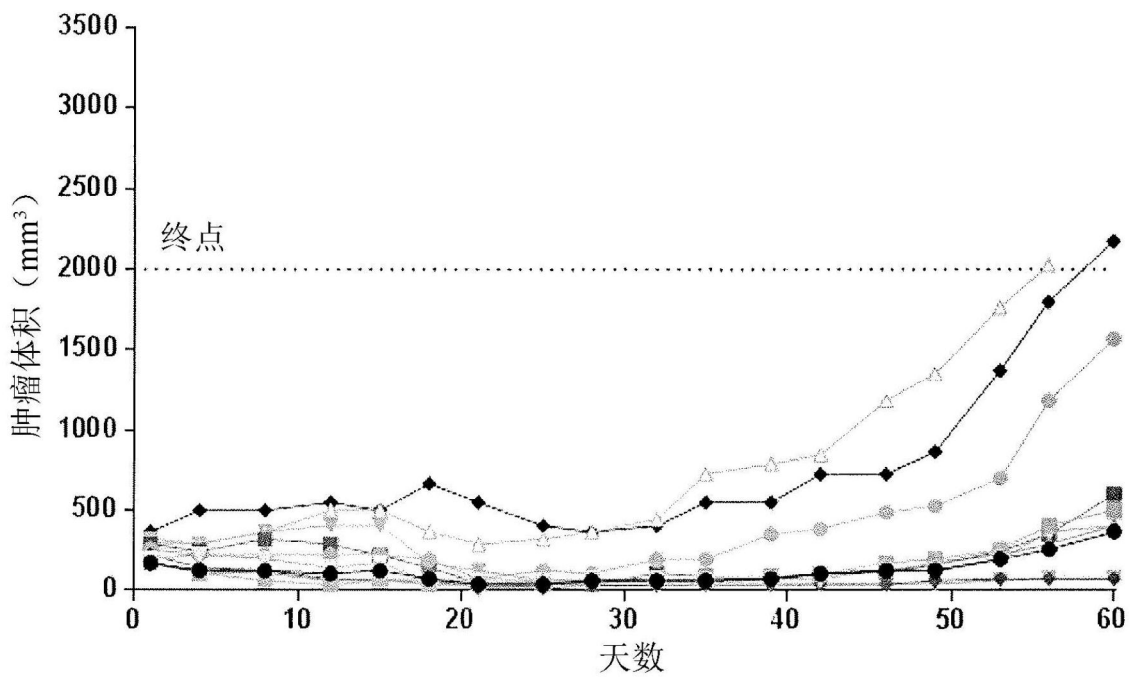


图3D

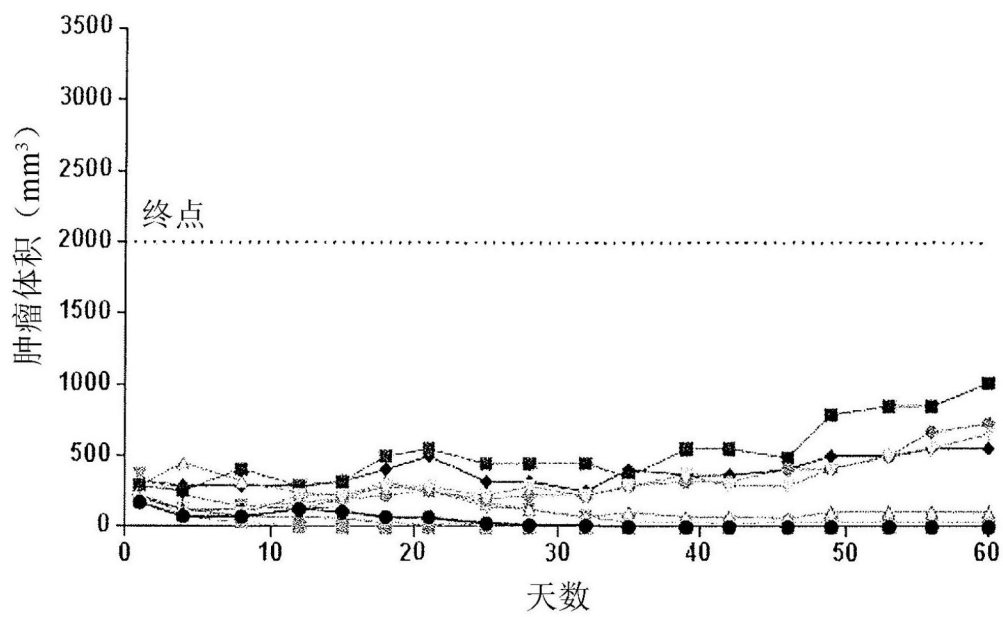


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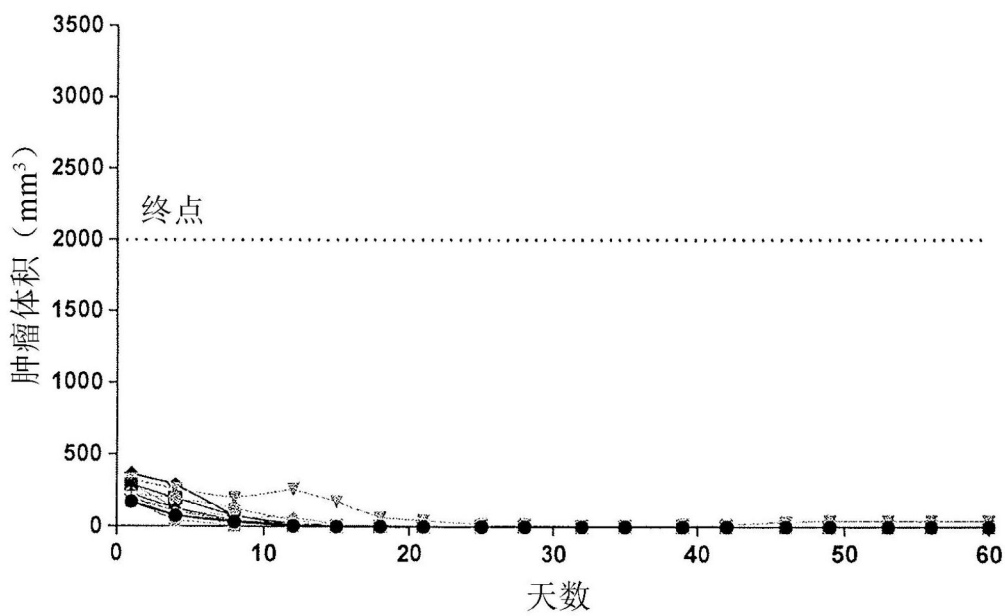


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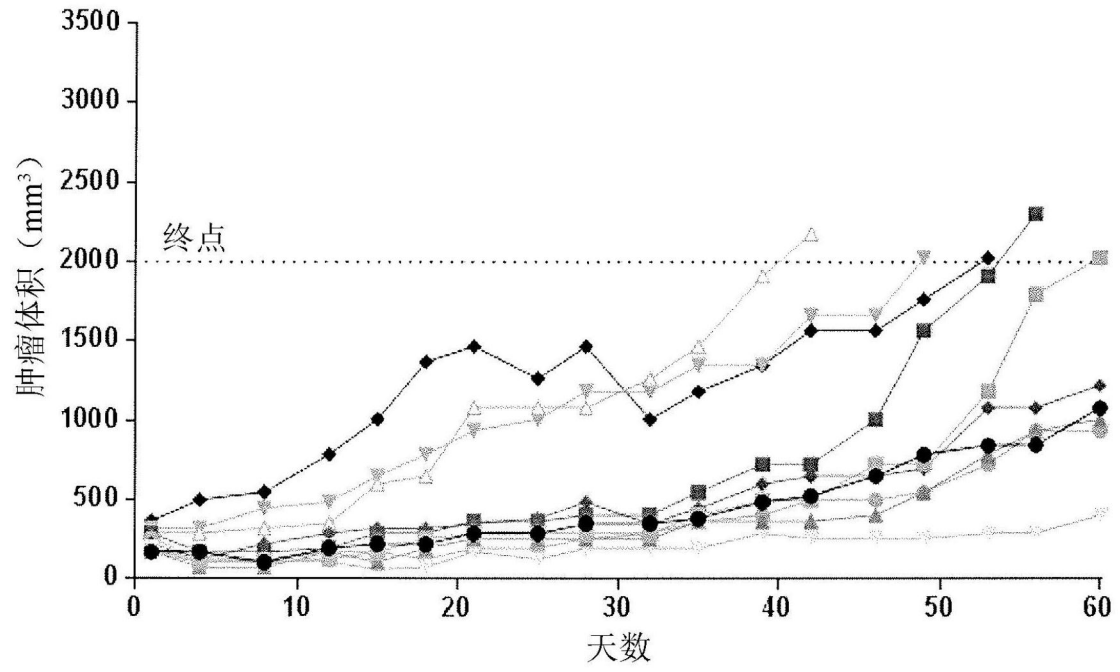


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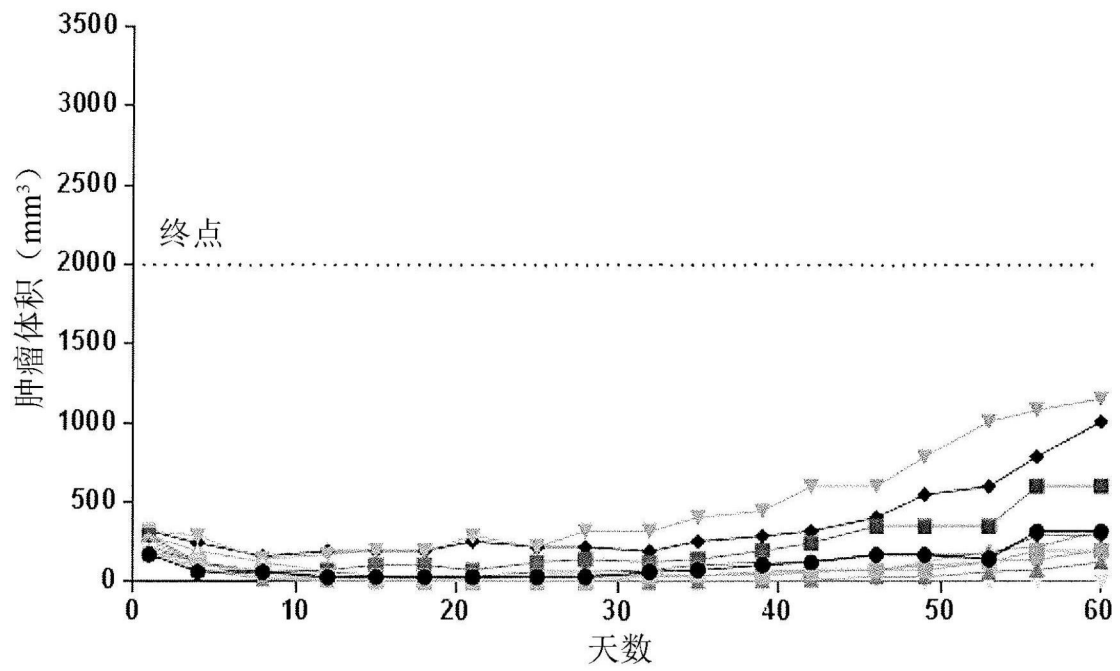


图3H

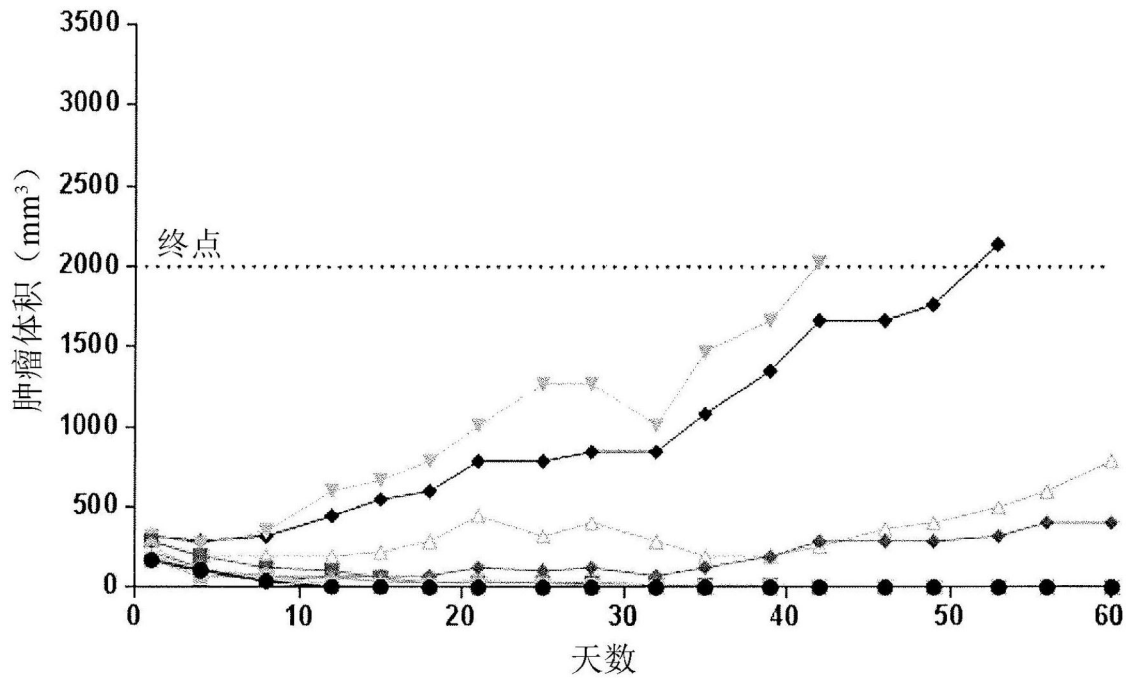


图3I

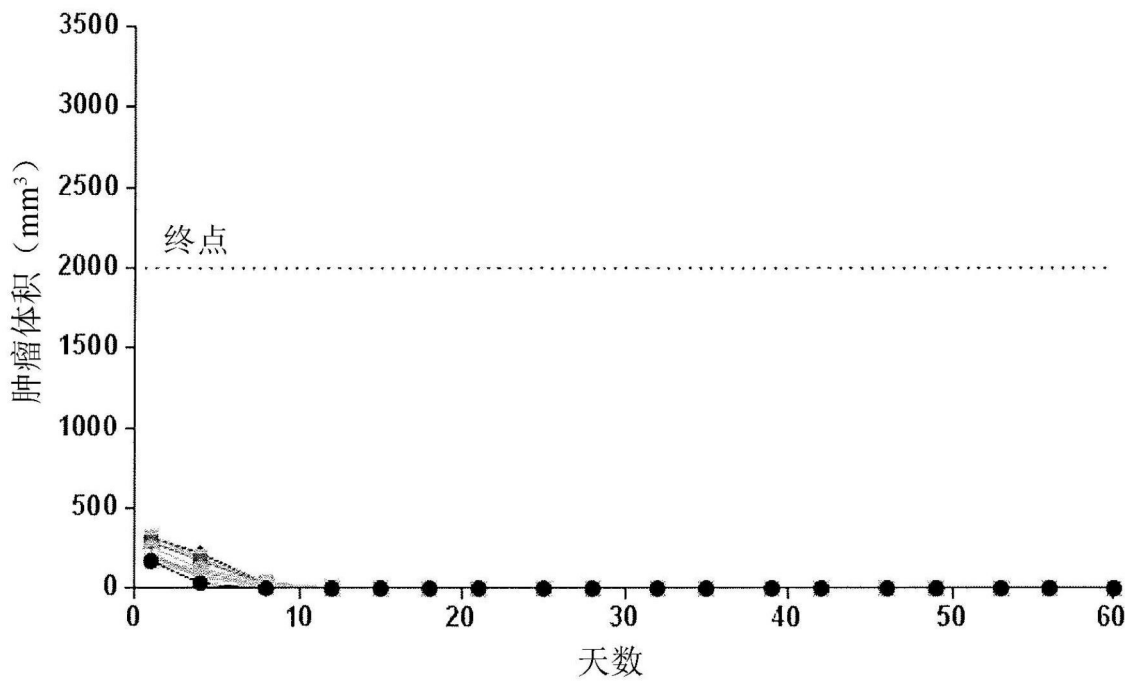
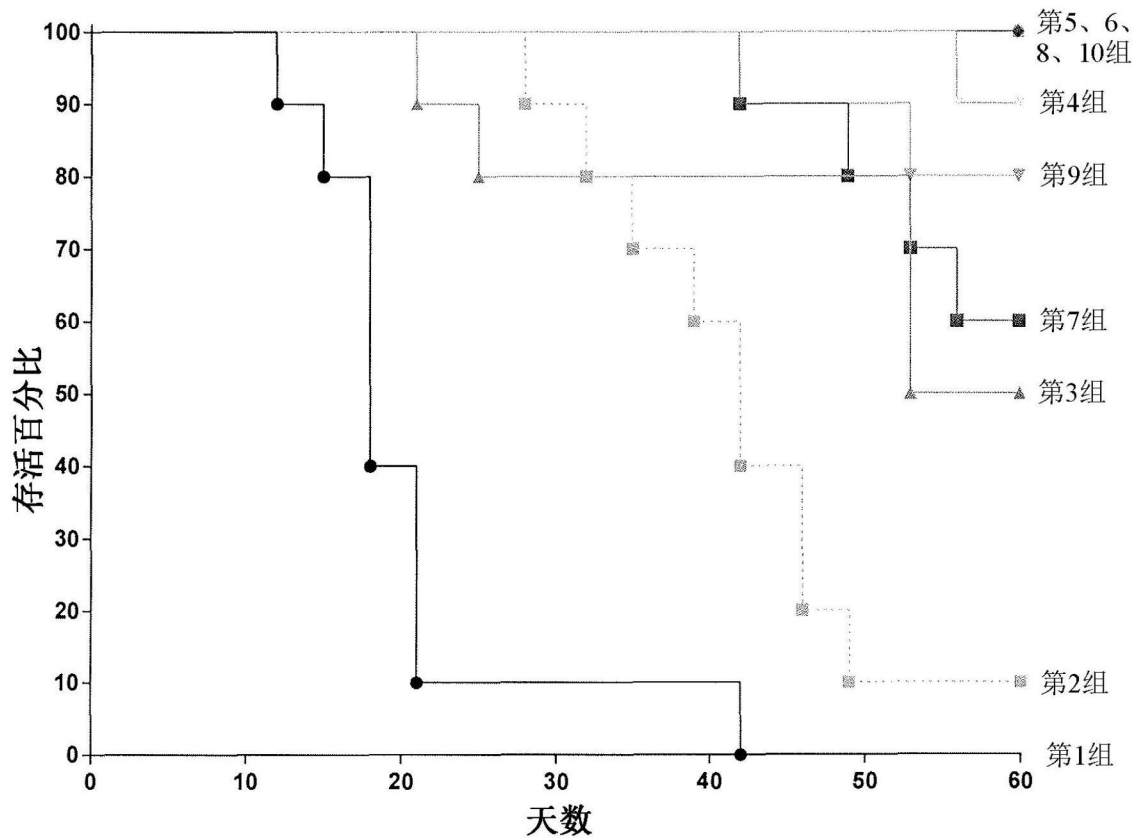


图3J



- 第1组：媒剂(腹膜内, 每两周一次持续29天)
- 第2组：来那度胺 (25 mg/kg, 腹膜内, qd x 21)
- ▲ 第3组：A10.21 (T106A) (0.3mg/kg, 腹膜内, 每两周一次持续29天)
- ◆ 第4组：A10.21 (T106A) (0.3mg/kg, 腹膜内, 每两周一次持续29天), 来那度胺(25 mg/kg, 腹膜内, qd x 21)
- ◆ 第5组：A10.21 (T106A) (1.0mg/kg, 腹膜内, 每两周一次持续29天)
- ◆ 第6组：A10.21 (T106A) (1.0mg/kg, 腹膜内, 每两周一次持续29天), 来那度胺(25 mg/kg, 腹膜内, qd x 21)
- 第7组：A10.21 (T106A) (1.0mg/kg, 腹膜内, 每四周一次持续29天)
- ▲ 第8组：A10.21 (T106A) (1.0mg/kg, 腹膜内, 每四周一次持续29天), 来那度胺(25 mg/kg, 腹膜内, qd x 21)
- ▼ 第9组：A10.21 (T106A) (3.0mg/kg, 腹膜内, 每四周一次持续29天)
- ◆ 第10组：A10.21 (T106A) (3.0mg/kg, 腹膜内, 每四周一次持续29天), 来那度胺(25 mg/kg, 腹膜内, qd x 21)

图4

泊利度胺组合研究编号1
H929
临时资料

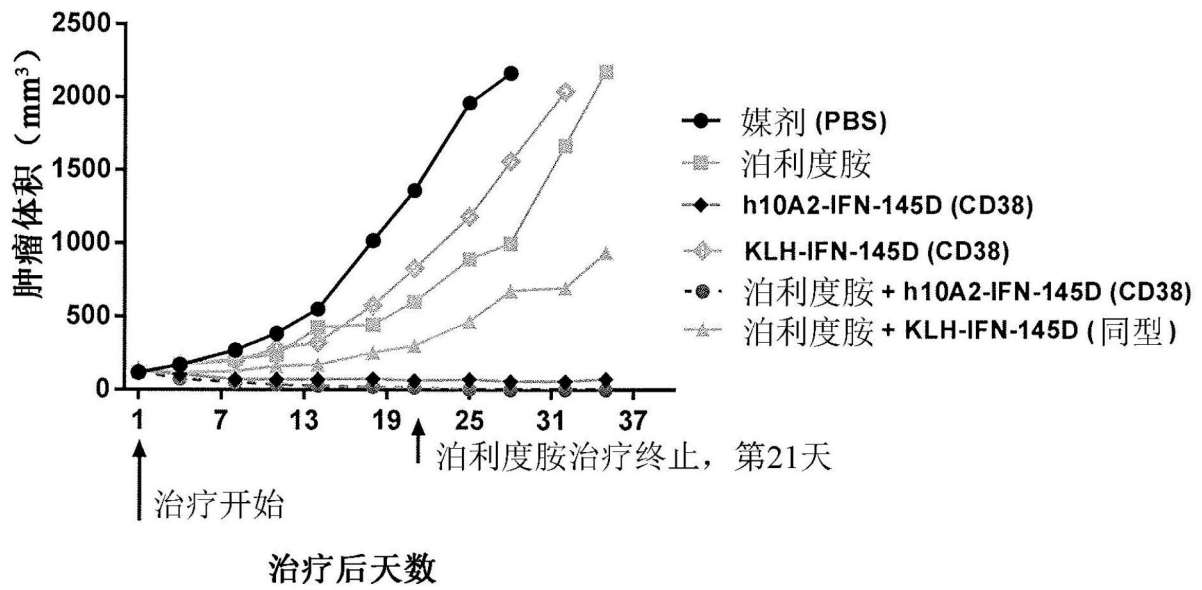


图5