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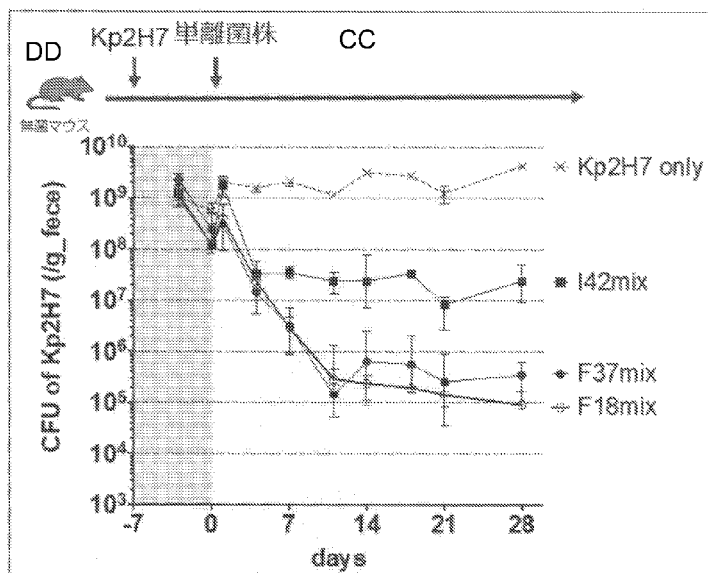
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(54) Title: ANTIBACTERIAL COMPOSITION AGAINST DRUG-RESISTANT BACTERIA OR PRO-INFLAMMATORY BACTERIA

(54) 発明の名称: 薬剤耐性細菌又は炎症惹起性細菌に対する抗菌組成物

[図11]



CC Kp2H7 isolated strain
DD Germ-free mouse

(57) Abstract: An antibacterial composition against drug-resistant bacteria or pro-inflammatory bacteria containing intestinal bacteria as an active ingredient or a pharmaceutical composition for treating, improving, or preventing infectious disease and inflammatory disease, etc.

(57) 要約: 腸内細菌を有効成分として含有する、薬剤耐性細菌若しくは炎症惹起性細菌に対する抗菌組成物、又は感染症及び炎症性疾患等を治療、改善若しくは予防するための医薬組成物。

[続葉有]

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- 明細書の別個の部分として表した配列リスト (規則5.2(a))

[DESCRIPTION]

[Title of Invention] ANTIBACTERIAL COMPOSITION AGAINST
DRUG-RESISTANT BACTERIA OR PRO-INFLAMMATORY BACTERIA

[Technical Field]

5 The present invention relates to an antibacterial
composition against drug-resistant bacteria or pro-
inflammatory bacteria. The present invention also relates
to a pharmaceutical composition or method for treating,
ameliorating, or preventing diseases caused by drug-
10 resistant bacteria or pro-inflammatory bacteria.

[Background Art]

Various commensal bacteria are resident on mucous
membranes such as the digestive tract and the oral cavity,
forming a flora as a whole. The resident flora plays a
15 very important role in the physiology and health
maintenance of the host. A compositional abnormality of
the resident flora is called dysbiosis, and has been
gradually revealed to be a cause of various diseases. The
advancement of the elucidation of the resident mucosal
20 flora may have a high possibility of developing new disease
countermeasures and treatments for various diseases, but
the detailed mechanism of such flora has not been
sufficiently clarified due to its complexity.

Humans produce and swallow about 1.5 liters of saliva
25 every day. Normally, bacteria contained in saliva (oral
bacteria) simply pass through the intestinal tract and do

not colonize. However, in some situations, oral bacteria may colonize in the intestinal tract. Particularly in Crohn's disease, liver cirrhosis, and colorectal cancer, it has been reported that colonization of oral bacteria in the intestinal tract was observed even at the early stages of disease onset. In addition, it is known that the colonized oral bacteria affect the pathological conditions of the diseases (NPLs 1 to 6).

Meanwhile, by allowing oral bacteria of patients with Crohn's disease and the like to colonize in the intestinal tract to induce Th1 cells, the present inventors succeeded in isolating, culturing, and identifying the bacteria involved in the onset of the diseases (PTL 1). More specifically, the present inventors found that as a result of oral administration of saliva derived from a patient with Crohn's disease to germ-free mice, interferon gamma (IFN- γ)-producing CD4-positive T-cells (Th1 cells) significantly increased in the large intestine. Then, the present inventors succeeded in isolating and culturing the Kp2H7 strain, which is considered to belong to *Klebsiella pneumoniae*, from the intestines of the mice in which this increase in Th1 cells was observed. Furthermore, it was also clarified that the bacteria derived from the saliva of patients with Crohn's disease were involved in the development of enteritis by colonizing in the intestinal tract and inducing the proliferation or activation of Th1

cells.

Moreover, the present inventors found that when Kp2H7 strains were orally administered to SPF (specific-pathogen-free) mice, intestinal colonization of these bacterial strains was not observed, unlike the case of the germ-free mice. Furthermore, it was also clarified that by administering an antibiotic to the SPF mice, these bacterial strains might be able to colonize in the intestinal tracts of the mice. Then, from these results, the present inventors considered that the intestinal tract contains intestinal bacteria that inhibit the intestinal colonization of Th1 cell-inducible bacteria such as the Kp2H7 strain, and administration of the antibiotic eliminates the intestinal bacteria from the intestinal tract, making the intestinal colonization of the bacteria possible.

Therefore, the present inventors tried to identify the bacteria that suppress the intestinal colonization of Th1 cell-inducible bacteria among human intestinal bacteria. As a result, the present inventors succeeded in isolating and culturing 68 strains, 37 strains, and 42 strains of intestinal bacterial strains from fecal samples derived from 3 healthy subjects (#K, #F, and #I), respectively, and in determining the sequence of 16S rDNA of each bacterial strain. Furthermore, it was clarified that administration of these bacterial strains suppressed

intestinal colonization of Th1 cell-inducible bacteria
(PTL 2).

[Citation List]

[Patent Literature]

5 [PTL 1] International Publication No. WO2018/084172

[PTL 2] International Publication No. WO2019/017389

[Non Patent Literature]

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[Summary of Invention]

20 [Technical Problem]

The present invention has an object to find an
intestinal bacterium having antibacterial activity against
drug-resistant bacteria or pro-inflammatory bacteria, and
to provide an antibacterial composition against drug-
25 resistant bacteria or pro-inflammatory bacteria containing
the intestinal bacterium as an active ingredient as well

as a pharmaceutical composition or method for treating, ameliorating, or preventing diseases caused by drug-resistant bacteria or pro-inflammatory bacteria.

[Solution to Problem]

5 The present inventors have made earnest studies to achieve the above object, and have clarified as a result that the above-mentioned bacteria that suppress the intestinal colonization of Th1 cell-inducible bacteria (68 strains of intestinal bacteria derived from healthy
10 subject #K, 37 strains of intestinal bacteria derived from healthy subject #F, and 42 strains of intestinal bacteria derived from healthy subject #I) can suppress the intestinal colonization of multidrug-resistant bacteria (carbapenem-resistant Enterobacteriaceae, vancomycin-
15 resistant enterococci, Clostridium difficile, Campylobacter jejuni) and pro-inflammatory bacteria (adherent-invasive Escherichia coli).

 Furthermore, regarding the ability to suppress bacterial colonization in the intestine, the present
20 inventors have succeeded in selecting 18 strains from the 37 strains of intestinal bacteria derived from healthy subject #F, which are capable of exerting the same level of suppression ability as the 37 strains. Thus, the present invention has been completed.

25 Specifically, the present invention provides the following.

[1] An antibacterial composition against drug-resistant bacteria or pro-inflammatory bacteria comprising: an intestinal bacterium as an active ingredient.

[2] The antibacterial composition according to [1],
5 wherein the intestinal bacterium is at least one bacterium which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69 to 105 or a base sequence having at least 90% identity to the base sequence.

[3] The antibacterial composition according to [1],
10 wherein the intestinal bacterium is at least one bacterium which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69, 80, 85 to 92, 94, 96, 98 to 101, 103, and 105 or a base sequence having at least 90% identity to the base sequence.

[4] The antibacterial composition according to [1],
15 wherein the intestinal bacterium is at least one bacterium specified by any of accession numbers NITE BP-03147 to 03164.

[5] The antibacterial composition according to [1],
20 wherein the intestinal bacterium is at least one bacterium which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 1 to 147 or a base sequence having at least 90% identity to the base sequence.

[6] The antibacterial composition according to [1],
25 wherein the intestinal bacterium is at least one bacterium which includes a DNA composed of a base sequence set forth

in any of SEQ ID NOs: 1 to 68 or a base sequence having at least 90% identity to the base sequence.

[7] The antibacterial composition according to [1], wherein the intestinal bacterium is at least one bacterium which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 106 to 147 or a base sequence having at least 90% identity to the base sequence.

[8] The antibacterial composition according to any one of [1] to [7], which is a pharmaceutical composition.

[9] The antibacterial composition according to any one of [1] to [7], which is a pharmaceutical composition for treating, ameliorating, or preventing an infectious disease or an inflammatory disease.

[Advantageous Effects of Invention]

According to the present invention, by suppressing the colonization and the like of drug-resistant bacteria or pro-inflammatory bacteria in the intestinal tract, it is possible to suppress the proliferation or activation of these bacteria, and it is possible to treat, ameliorate, or prevent diseases caused by these bacteria.

[Brief Description of Drawings]

[Fig. 1] Fig. 1 is a graph in CFU showing changes over time in the bacterial amount of fecal *Klebsiella* when *Klebsiella* 2H7 strains (Kp2H7) were administered to germ-free mice, and one week later, fecal samples of healthy subjects were administered to the mice (FMT). Five types

of feces were used, and Klebsiella was significantly reduced in all samples.

[Fig. 2] Fig. 2 is a bar graph showing the results of 16S meta-analysis of three types of feces derived from healthy donors F, I, and K. Each segment indicates one bacterial strain, and its size indicates the proportion of that bacterium to the total bacterial amount. The three types of feces were cultured in an anaerobic environment, and the strains that could be cultured and isolated there are shown in yellow in the adjacent graph (under color display). The total number of bacteria that could be isolated is shown below.

[Fig. 3] Fig. 3 is a graph in CFU showing changes over time in the bacterial amount of fecal Kp2H7 when Kp2H7 was colonized in germ-free mice and then the bacterial strains isolated from feces were mixed and administered. The 37 strains derived from the feces derived from healthy donor F (feces F) reduced Klebsiella as much as the fecal sample.

[Fig. 4] Fig. 4 is a graph in CFU showing changes over time in the bacterial amount of fecal Kp2H7 when Kp2H7 was colonized in germ-free mice and then the bacterial strains isolated from feces were mixed and administered. The 37 strains derived from feces F and 68 bacterial strains isolated from the feces derived from healthy donor K (feces K) reduced Klebsiella as much as the fecal sample.

[Fig. 5] Fig. 5 is a graph in CFU showing changes over

time in the bacterial amount of fecal Kp2H7 when Kp2H7 was administered to germ-free mice, then F37mix (37 bacterial strains derived from feces F) was administered one week later, and ampicillin was further administered by drinking water one month later. Klebsiella transiently increased with ampicillin administration, but then decreased again.

[Fig. 6A] Fig. 6A is a diagram showing changes over time in the abundance ratio of each bacterial amount (F31, F22, F20, F32) in the total bacterial amount in the experiment shown in Fig. 5. Below the figure, the number of each bacterium, r (Spearman's rank correlation coefficient with Klebsiella), and the bacterium's name are shown.

[Fig. 6B] Fig. 6B is a diagram showing changes over time in the abundance ratio of each bacterial amount (F26, F28, F21, F30) in the total bacterial amount in the experiment shown in Fig. 5. Below the figure, the number of each bacterium, r (Spearman's rank correlation coefficient with Klebsiella), and the bacterium's name are shown.

[Fig. 6C] Fig. 6C is a diagram showing changes over time in the abundance ratio of each bacterial amount (F24, F23/F25, F35/F36, F09) in the total bacterial amount in the experiment shown in Fig. 5. Below the figure, the number of each bacterium, r (Spearman's rank correlation coefficient with Klebsiella), and the bacterium's name are shown.

[Fig. 6D] Fig. 6D is a diagram showing changes over time

in the abundance ratio of each bacterial amount (F33, F12, F17/F19, F18) in the total bacterial amount in the experiment shown in Fig. 5. Below the figure, the number of each bacterium, r (Spearman's rank correlation coefficient with *Klebsiella*), and the bacterium's name are shown.

[Fig. 6E] Fig. 6E is a diagram showing changes over time in the abundance ratio of each bacterial amount (F34, F03/F08, F29, F13) in the total bacterial amount in the experiment shown in Fig. 5. Below the figure, the number of each bacterium, r (Spearman's rank correlation coefficient with *Klebsiella*), and the bacterium's name are shown.

[Fig. 6F] Fig. 6F is a diagram showing changes over time in the abundance ratio of each bacterial amount (F04/F08, F37, F01, F02) in the total bacterial amount in the experiment shown in Fig. 5. Below the figure, the number of each bacterium, r (Spearman's rank correlation coefficient with *Klebsiella*), and the bacterium's name are shown.

[Fig. 6G] Fig. 6G is a diagram showing changes over time in the abundance ratio of each bacterial amount (F05, F07, F14) in the total bacterial amount in the experiment shown in Fig. 5. Below the figure, the number of each bacterium, r (Spearman's rank correlation coefficient with *Klebsiella*), and the bacterium's name are shown.

[Fig. 6H] Fig. 6H is a diagram showing changes over time in the abundance ratio of each bacterial amount (F10/F15, F16, F11/F27) in the total bacterial amount in the experiment shown in Fig. 5. Below the figure, the number of each bacterium, r (Spearman's rank correlation coefficient with *Klebsiella*), and the bacterium's name are shown.

[Fig. 7] Fig. 7 is a diagram in which bacteria are arranged in the order of positive correlation in Spearman's rank correlation coefficient between Kp2H7 and each bacterium in terms of bacterial amount. Many of the *Bacteroides* are not related in movement to Kp2H7, and many of the negatively correlated ones are in the *Firmicutes* genus.

[Fig. 8] Fig. 8 is a graph in CFU showing changes over time in the bacterial amount of fecal Kp2H7 when Kp2H7 was administered to germ-free mice for colonization, and then the 37 strains shown in Fig. 7 (F37mix), 8 strains of the 37 strains belonging to the *Bacteroidetes* genus (F8mix), or the 29 other bacterial strains (F29mix) were mixed and administered. The 29 strains excluding *Bacteroides* also showed a decrease in *Klebsiella* that was almost the same as the 37 strains, and it is considered that *Bacteroides* is unnecessary for the elimination of *Klebsiella*.

[Fig. 9] Fig. 9 is a phylogenetic tree showing a breakdown of F37mix, F8mix, and F29mix used in the

experiment shown in Fig. 8. The phylogenetic tree was prepared by the Neighbor-joining method using MEGA X with the DNA base sequence of the 16S rDNA analysis result of the isolated bacteria by the Sanger method. The same
 5 applies to the preparation of the phylogenetic trees in Figs. 10 and 12.

[Fig. 10] Fig. 10 is a phylogenetic tree showing a breakdown of 18 bacterial strains derived from feces F (F18mix).

10 [Fig. 11] Fig. 11 is a graph in CFU showing changes over time in the bacterial amount of fecal Kp2H7 when Kp2H7 was colonized in germ-free mice, and then 37 bacterial strains derived from feces F (F37mix described above), 18 bacterial strains derived from feces F (F18mix shown in
 15 Fig. 10), or 42 bacterial strains derived from the feces derived from healthy subject I (feces I) were mixed and administered. F18mix also successfully eliminated *Klebsiella* in the same way as F37mix.

[Fig. 12] Fig. 12 is a phylogenetic tree that classifies
 20 18 bacterial strains derived from feces F (F18mix) into 4 groups and shows the breakdown thereof. These 4 groups were subtracted from the 18 bacterial strains (F18mix) to prepare bacterial strain groups of F15mix (F18mix-other phyla), F12mix (F18mix-Lachnoclostridium), F14mix (F18mix-
 25 Blautia), and F13mix (F18mix-other Firmicutes), which were subjected to the experiment shown in Fig. 11.

[Fig. 13] Fig. 13 is a graph in CFU showing changes over time in the bacterial amount of fecal Kp2H7 when Kp2H7 was colonized in germ-free mice, and then the group excluding the duplicate strains and F18mix from F37mix (F31-18mix), F15mix described above, F12mix described above, F14mix described above, F13mix described above, or F18mix described above was mixed and administered. Note that Fig. 13 shows data from the experiments performed twice. Excluding any group shown in Fig. 12 from F18mix decreased the ability to eliminate Klebsiella, and it has become clear that every group is important for the elimination of Klebsiella.

[Fig. 14] Fig. 14 is a graph showing the CFU of fecal Kp2H7 in each group at the time of day 28 in the experiment shown in Fig. 13. The F18mix-administered group has a statistically significantly smaller bacterial amount of Klebsiella than the other administration groups excluding F37mix.

[Fig. 15] Fig. 15 is a dot plot diagram showing the results of flow cytometric analysis of immune cells in the lamina propria of the large intestine of mice in the F37mix-administered group, the F18mix-administered group, or the Kp2H7 alone administration group. The numerical value in each gate (square) in the figure shows the proportion of CD4+IFN γ + cells. The induction of CD4+IFN γ + cells is suppressed in the F37mix-administered group and

the F18mix-administered group as compared with the Kp2H7 alone administration group.

[Fig. 16] Fig. 16 is a graph in CFU showing changes over time in the bacterial amount of fecal Kp2H7 when Kp2H7 was administered to germ-free mice, and one week later, each bacterial strain mix was administered. The "F15mix" in the figure shows the result of mice administered by removing three strains of *E. coli*, *Fusobacterium*, and *Bifidobacterium* from F18mix, and "F18mix-*E. coli*", "F18mix-*Fusobacterium*," and "F18mix-*Bifidobacterium*" each show the result of mice administered by removing one of the three strains from F18mix. The effects of all three strains diminished when removed from F18mix, indicating that each was involved in the elimination of *Klebsiella*.

[Fig. 17] Fig. 17 is a graph in CFU showing changes over time in the bacterial amount of fecal Kp2H7 when Kp2H7 was administered to a germ-free Rag2^{-/-}γc^{-/-} mouse, MyD88^{-/-} Triff^{-/-} mouse, or wild-type mouse (WT), and one week later, mixed F37mix was administered. *Klebsiella* was equally eliminated in all types of mice. This suggests that the major innate immunity and acquired immunity of the host are not involved in the elimination of *Klebsiella*.

[Fig. 18] Fig. 18 is a graph in CFU showing changes over time in the bacterial amount of fecal CRE when *Klebsiella* (Kp-CRE) was administered to germ-free mice, and one week later, the isolated bacteria mixes (F37mix, K68mix,

I42mix) were administered to the mice. F37mix and K68mix can also reduce CRE.

[Fig. 19] Fig. 19 is a photomicrograph showing the results of analyzing the large intestine of a mouse at the end of the experiment shown in Fig. 18 by HE staining. No inflammatory findings were observed in any of the isolated bacterium mix-administered mice.

[Fig. 20] Fig. 20 is a graph in CFU showing changes over time in the bacterial amount of fecal VRE when VRE (vancomycin-resistant enterococci) was administered to germ-free mice, and one week later, the isolated bacteria mixes (F37mix, K68mix, I42mix) were administered to the mice. For VRE, K68mix was able to reduce the bacterial amount more than F37mix.

[Fig. 21] Fig. 21 is a photomicrograph showing the results of analyzing the large intestine of a mouse at the end of the experiment shown in Fig. 18 by HE staining. No inflammatory findings were observed in any of the isolated bacterium mix-administered mice.

[Fig. 22] Fig. 22 is a graph in CFU showing changes over time in the bacterial amount of fecal AIEC when AIEC was administered to germ-free mice, and one week later, the isolated bacteria mixes (F37mix, K68mix, I42mix) were administered to the mice. For AIEC, F37mix was most effective in reducing the bacterial amount.

[Fig. 23] Fig. 23 is a graph in CFU showing changes over

time in the bacterial amount of fecal ESBL-producing Klebsiella when ESBL-producing Klebsiella was administered to germ-free mice, and one week later, the isolated bacterium mixes (F37mix, K68mix, I42mix) were administered to the mice. F37mix and K68mix were able to eliminate ESBL-producing Klebsiella as much as F-derived feces.

[Fig. 24] Fig. 24 is a graph in CFU showing changes over time in the bacterial amount of fecal Campylobacter when Campylobacter jejuni was administered to germ-free mice, and one week later, fecal samples derived from the isolated bacteria mixes (F37mix, K68mix, I42mix) or healthy subject F were administered to the mice. Elimination of Campylobacter jejuni was observed to the same extent in all the isolated bacteria mix-administered groups.

[Fig. 25] Fig. 25 is a graph showing changes over time in the bacterial amount of fecal Campylobacter as a relative value divided by the total bacterial amount when Campylobacter jejuni was administered to germ-free mice, and one week later, fecal samples derived from the isolated bacteria mixes (F37mix, K68mix, I42mix) or healthy subject F were administered to the mice. Elimination of Campylobacter jejuni was observed to the same extent in all the isolated bacteria mix-administered groups.

[Fig. 26] Fig. 26 is a graph showing the results of qPCR analysis of changes over time in the bacterial amount of fecal Clostridium difficile when Clostridium difficile was

administered to germ-free mice, and one week later, fecal samples derived from the isolated bacteria mixes (F37mix, K68mix, I42mix, K47mix) or healthy subject F were administered to the mice. K68mix and K47mix were able to eliminate *Clostridium difficile* as compared with F-derived feces, but the elimination effect of F37mix was lower.

[Description of Embodiments]

<Intestinal Bacteria>

In the present invention, the intestinal bacteria contained as an active ingredient of the antibacterial composition have an antibacterial action against drug-resistant bacteria or pro-inflammatory bacteria (hereinafter also referred to as "drug-resistant bacteria and the like") in the intestinal tract.

In the present invention, the "antibacterial activity" means an activity of suppressing the activity of bacteria, more specifically, an activity of suppressing the proliferation or colonization of bacteria or killing bacteria, and examples thereof include the activity of suppressing the colonization of bacteria in the intestines and the activity of eliminating bacteria from the intestines.

The "Intestinal bacteria" mean bacteria present in the intestinal tract of an animal. Examples of animals in which such bacteria are present include humans and non-human animals (such as mice, rats, monkeys, pigs, cattle,

horses, sheep, goats, chickens, ducks, ostriches, domestic ducks, dogs, cats, rabbits, and hamsters), but among these animals, humans are preferable.

In the present invention, the "intestinal bacteria" may be a single strain of bacteria or a mixture of bacterial strains composed of multiple strains of bacteria. Note that when composed of multiple strains of bacteria, it is desirable that at least one of the bacterial strains has antibacterial activity against drug-resistant bacteria and the like. Further, in such a case, the multiple strains of bacteria may even include a bacterial strain that does not have the antibacterial activity, such as a bacterial strain having an action of enhancing the activity of a bacterial strain having the antibacterial activity, a bacterial strain having an action of maintaining the proliferation or colonization of a bacterial strain having antibacterial activity, and a bacterial strain having an action of suppressing the inhibitory activity of a bacterium that inhibits the antibacterial activity.

In the present invention, examples of the "intestinal bacteria" include at least one bacterium having a DNA composed of a base sequence set forth in any of SEQ ID NOs: 1 to 147 or a base sequence having at least 70% identity to the base sequence, at least one bacterium having a DNA composed of a base sequence set forth in any

of SEQ ID NOs: 1 to 68 or a base sequence having at least 70% identity to the base sequence, at least one bacterium having a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69 to 105 or a base sequence having at least 70% identity to the base sequence (for example, at least one bacterium having a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69, 80, 85 to 92, 94, 96, 98 to 101, 103, and 105 or a base sequence having at least 70% identity to the base sequence), and at least one bacterium having a DNA composed of a base sequence set forth in any of SEQ ID NOs: 106 to 147 or a base sequence having at least 70% identity to the base sequence.

The sequence indicated by each SEQ ID NO is the sequence of 16S rDNA of each bacterium K68, F37, and I43 in the attached document. Tables 1 to 4 below show the correspondence between each bacterium, the SEQ ID NO indicating the sequence of each 16S rDNA, and each bacterial species estimated from the sequence. In addition, K, F, and I represent intestinal bacteria isolated from the feces of three healthy subjects (Japanese) (see PTL 2).

[Table 1]

No.	Species	SEQ ID NO:	subject id	NCBI TAX ID	% identity
K01	<i>Drancourtella massiliensis</i>	1	NR_144722.1	1632013	99.57
K02	<i>Bacteroides ovatus</i>	2	NR_112940.1	28116	99.86
K03	<i>Blautia coccoides</i>	3	NR_104700.1	1532	99.86
K04	<i>Blautia hominis</i>	4	NR_163638.1	2025493	99.43
K05	<i>Desulfovibrio vulgaris</i>	5	NR_074446.1	882	91.58
K06	<i>Alistipes onderdonkii</i>	6	NR_043318.1	328813	99.86
K07	<i>Eisenbergiella massiliensis</i>	7	NR_144731.1	1720294	99.50
K08	[<i>Clostridium</i>] <i>innocuum</i>	8	NR_029164.1	1522	97.93
K09	<i>Bacteroides fragilis</i>	9	NR_112936.1	817	99.71
K10	<i>Eggerthella lenta</i>	10	NR_074377.1	84112	100.00
K11	<i>Dielma fastidiosa</i>	11	NR_125593.1	1034346	99.71
K12	<i>Erysipelatoclostridium ramosum</i>	12	NR_113243.1	1547	100.00
K13	<i>Enterococcus faecalis</i>	13	NR_115765.1	1351	99.93
K14	<i>Bacteroides intestinalis</i>	14	NR_041307.1	329854	99.35
K15	[<i>Clostridium</i>] <i>symbiosum</i>	15	NR_118730.1	1512	98.35
K16	[<i>Clostridium</i>] <i>hylemonae</i>	16	NR_024719.1	89153	99.50
K17	<i>Hungatella effluvii</i>	17	NR_133762.1	1096246	98.49
K18	<i>Bacteroides dorei</i>	18	NR_041351.1	357276	99.93
K19	[<i>Clostridium</i>] <i>clostridioforme</i>	19	NR_044715.2	1531	98.99
K20	<i>Flavonifractor plautii</i>	20	NR_029356.1	292800	100.00
K21	<i>Bacteroides xylanisolvens</i>	21	NR_112947.1	657309	99.50
K22	<i>Bacteroides thetaiotaomicron</i>	22	NR_112944.1	818	99.93
K23	<i>Parabacteroides merdae</i>	23	NR_041343.1	46503	99.78
K24	<i>Bacteroides vulgatus</i>	24	NR_074515.1	435590	100.00
K25	[<i>Clostridium</i>] <i>aldenense</i>	25	NR_043680.1	358742	99.41
K26	<i>Bacteroides uniformis</i>	26	NR_112945.1	820	97.39
K27	<i>Gordonibacter urolithinifaciens</i>	27	NR_148261.1	1335613	99.56
K28	<i>Coprococcus comes</i>	28	NR_044048.1	470146	92.77
K29	<i>Anaerostipes caccae</i>	29	NR_028915.1	105841	98.78
K30	[<i>Ruminococcus</i>] <i>gnavus</i>	30	NR_036800.1	411470	99.78
K31	[<i>Ruminococcus</i>] <i>gnavus</i>	31	NR_036800.1	411470	98.71
K32	<i>Alistipes shahii</i>	32	NR_113153.1	328814	100.00
K33	<i>Bacteroides stercoris</i>	33	NR_112943.1	449673	98.77
K34	<i>Blautia hominis</i>	34	NR_163634.1	2025493	98.71

[Table 2]

No.	Species	SEQ ID NO:	subject id	NCBI TAX ID	% identity
K35	<i>Butyricoccus faecihominis</i>	35	NR_152060.1	1712515	97.89
K36	[<i>Clostridium</i>] <i>bolteae</i>	36	NR_025567.1	208479	99.50
K37	<i>Phoceia massiliensis</i>	37	NR_144748.1	1841867	99.93
K38	<i>Holdemania massiliensis</i>	38	NR_125628.1	1211819	99.71
K39	<i>Escherichia coli</i>	39	NR_114042.1	562	99.79
K40	<i>Agathobaculum desmolans</i>	40	NR_044644.2	39484	96.68
K41	[<i>Eubacterium</i>] <i>rectale</i>	41	NR_074634.1	515619	100.00
K42	<i>Lactonifactor longoviformis</i>	42	NR_043551.1	341220	100.00
K43	<i>Oscillibacter ruminantium</i>	43	NR_118156.1	1007096	95.90
K44	<i>Pseudoflavonifractor phocaeensis</i>	44	NR_147370.1	1870988	97.42
K45	<i>Streptococcus pasteurianus</i>	45	NR_043660.1	197614	100.00
K46	<i>Sutterella wadsworthensis</i>	46	NR_117778.1	40545	99.93
K47	<i>Bifidobacterium faecale</i>	47	NR_133982.1	1454229	99.35
K48	<i>Eisenbergiella massiliensis</i>	48	NR_144731.1	1720294	93.50
K49	<i>Fusicatenibacter saccharivorans</i>	49	NR_114326.1	1150298	99.78
K50	<i>Robinsoniella peoriensis</i>	50	NR_041882.1	180332	93.40
K51	<i>Massilimicrobiota timonensis</i>	51	NR_144738.1	1776392	99.93
K52	<i>Blautia faecis</i>	52	NR_109014.1	871665	99.92
K53	<i>Blautia wexlerae</i>	53	NR_044054.1	1121115	98.63
K54	<i>Phascolarctobacterium faecium</i>	54	NR_026111.1	33025	99.72
K55	<i>Odoribacter splanchnicus</i>	55	NR_113075.1	28118	99.71
K56	<i>Faecalibacterium prausnitzii</i>	56	NR_028961.1	853	97.29
K57	<i>Colidextribacter massiliensis</i>	57	NR_147375.1	1870986	94.70
K58	<i>Emergencia timonensis</i>	58	NR_144737.1	1776384	91.61
K59	<i>Alistipes finegoldii</i>	59	NR_043064.1	214856	99.42
K60	<i>Ruthenibacterium lactatiformans</i>	60	NR_151900.1	1550024	99.93
K61	<i>Christensenella minuta</i>	61	NR_112900.1	626937	98.71
K62	[<i>Clostridium</i>] <i>scindens</i>	62	NR_028785.1	29347	98.49
K63	<i>Enterococcus faecalis</i>	63	NR_115765.1	1351	100.00
K64	<i>Blautia coccoides</i>	64	NR_104700.1	1532	99.93
K65	<i>Alistipes ihumii</i>	65	NR_144706.1	1211813	99.28
K66	<i>Intestinimonas butyriciproducens</i>	66	NR_118554.1	1297617	99.86
K67	<i>Bacteroides uniformis</i>	67	NR_112945.1	820	99.93
K68	<i>Akkermansia muciniphila</i>	68	NR_074436.1	239935	98.91

[Table 3]

No.	Species	SEQ ID NO:	subject id	NCBI TAX ID	% identity
F01	<i>Bifidobacterium longum</i>	69	NR_145535.1	1931217	99.60
F02	<i>Bacteroides xylanisolvens</i>	70	NR_112947.1	657309	99.59
F03	<i>Bacteroides fragilis</i>	71	NR_074784.2	817	98.95
F04	<i>Bacteroides uniformis</i>	72	NR_112945.1	820	99.93
F05	<i>Bacteroides thetaiotaomicron</i>	73	NR_074277.1	818	99.80
F06	<i>Bacteroides uniformis</i>	74	NR_112945.1	820	99.93
F07	<i>Bacteroides acidifaciens</i>	75	NR_112931.1	85831	97.18
F08	<i>Bacteroides fragilis</i>	76	NR_074784.2	817	98.95
F09	<i>Parabacteroides goldsteinii</i>	77	NR_113076.1	927665	98.98
F10	[<i>Ruminococcus</i>] <i>gnavus</i>	78	NR_036800.1	411470	99.80
F11	<i>Blautia luti</i>	79	NR_114315.1	649762	97.57
F12	<i>Faecalimonas umbilicata</i>	80	NR_156907.1	1912855	99.87
F13	[<i>Clostridium</i>] <i>saccharolyticum</i>	81	NR_102852.1	84030	94.34
F14	<i>Tyzzellerella nexilis</i>	82	NR_029248.1	500632	98.49
F15	[<i>Ruminococcus</i>] <i>gnavus</i>	83	NR_036800.1	411470	99.80
F16	<i>Anaerostipes hadrus</i>	84	NR_117138.2	649756	99.74
F17	<i>Blautia glucerasea</i>	85	NR_113231.1	536633	96.91
F18	[<i>Clostridium</i>] <i>bolteae</i>	86	NR_025567.1	208479	99.60
F19	<i>Blautia caecimuris</i>	87	NR_144607.1	1796615	99.13
F20	[<i>Clostridium</i>] <i>innocuum</i>	88	NR_029164.1	1522	98.71
F21	<i>Blautia marasmi</i>	89	NR_147395.1	1917868	98.20
F22	[<i>Clostridium</i>] <i>lavalense</i>	90	NR_044289.1	460384	98.97
F23	[<i>Clostridium</i>] <i>glycyrrhizinilyticum</i>	91	NR_112553.1	342942	99.13
F24	<i>Eisenbergiella massiliensis</i>	92	NR_144731.1	1720294	97.73
F25	[<i>Clostridium</i>] <i>glycyrrhizinilyticum</i>	93	NR_112553.1	342942	99.13
F26	<i>Flavonifractor plautii</i>	94	NR_029356.1	292800	100.00
F27	<i>Blautia luti</i>	95	NR_114315.1	649762	97.57
F28	<i>Intestinibacter bartlettii</i>	96	NR_027573.1	261299	99.93
F29	[<i>Ruminococcus</i>] <i>gnavus</i>	97	NR_036800.1	411470	98.85
F30	<i>Massilimicrobiota timonensis</i>	98	NR_144738.1	1776392	100.00
F31	<i>Anaerostipes caccae</i>	99	NR_028915.1	105841	97.32
F32	<i>Blautia coccoides</i>	100	NR_104700.1	1532	99.93
F33	<i>Erysipelatoclostridium ramosum</i>	101	NR_029247.1	1547	99.41
F34	<i>Veillonella parvula</i>	102	NR_074980.1	479436	99.16
F35	<i>Fusobacterium varium</i>	103	NR_113384.1	856	99.73
F36	<i>Fusobacterium varium</i>	104	NR_113384.1	856	99.73
F37	<i>Escherichia fergusonii</i>	105	NR_074902.1	585054	99.87

[Table 4]

No.	Species	SEQ ID NO:	subject id	NCBI TAX ID	% identity
I01	<i>Bifidobacterium faecale</i>	106	NR_133982.1	1454229	98.06
I02	<i>Bifidobacterium pseudocatenulatum</i>	107	NR_037117.1	28026	99.63
I03	<i>Bifidobacterium bifidum</i>	108	NR_044771.1	1681	100.00
I04	<i>Bifidobacterium longum</i>	109	NR_145535.1	1931217	99.70
I05	<i>Collinsella aerofaciens</i>	110	NR_113316.1	74426	99.92
I06	<i>Collinsella aerofaciens</i>	111	NR_113316.1	74426	99.72
I07	<i>Bifidobacterium longum</i>	112	NR_145535.1	1931217	99.70
I08	<i>Bacteroides stercoris</i>	113	NR_112943.1	449673	99.18
I09	<i>Bacteroides massiliensis</i>	114	NR_042745.1	204516	99.70
I10	<i>Bacteroides vulgatus</i>	115	NR_074515.1	435590	100.00
I11	<i>Bacteroides dorei</i>	116	NR_041351.1	357276	100.00
I12	<i>Parabacteroides merdae</i>	117	NR_041343.1	46503	99.85
I13	<i>Parabacteroides distasonis</i>	118	NR_041342.1	823	99.03
I14	<i>Alistipes putredinis</i>	119	NR_113152.1	28117	100.00
I15	<i>Bacteroides uniformis</i>	120	NR_040866.1	820	97.75
I16	<i>Bacteroides koreensis</i>	121	NR_159117.1	1912896	99.26
I17	<i>Alistipes shahii</i>	122	NR_113153.1	328814	99.63
I18	<i>Odoribacter splanchnicus</i>	123	NR_113075.1	28118	99.77
I19	<i>Faecalibacterium prausnitzii</i>	124	NR_028961.1	853	97.24
I20	<i>Faecalibacterium prausnitzii</i>	125	NR_028961.1	853	97.24
I21	<i>Blautia wexlerae</i>	126	NR_044054.1	1121115	98.38
I22	<i>Ruminococcus lactaris</i>	127	NR_027579.1	471875	97.24
I23	<i>Ruminococcus albus</i>	128	NR_113032.1	1264	94.68
I24	<i>Faecalibacterium prausnitzii</i>	129	NR_028961.1	853	96.86
I25	<i>Dorea longicatena</i>	130	NR_028883.1	88431	99.47
I26	<i>Dorea formicigenerans</i>	131	NR_044645.2	39486	97.94
I27	<i>Anaerostipes hadrus</i>	132	NR_117138.2	649756	99.85
I28	<i>Intestinibacter bartlettii</i>	133	NR_027573.1	261299	99.55
I29	<i>Flavonifractor plautii</i>	134	NR_029356.1	292800	99.71
I30	<i>Pseudoflavonifractor phocaeensis</i>	135	NR_147370.1	1870988	97.86
I31	[<i>Clostridium</i>] <i>spiroforme</i>	136	NR_114393.1	29348	93.09
I32	<i>Megasphaera elsdenii</i>	137	NR_102980.1	1064535	99.35
I33	<i>Dialister succinatiphilus</i>	138	NR_041666.1	742743	97.02
I34	<i>Acidaminococcus intestini</i>	139	NR_041894.1	187327	99.93
I35	<i>Allisonella histaminiformans</i>	140	NR_028862.1	209880	99.50
I36	<i>Megasphaera massiliensis</i>	141	NR_133027.1	1232428	99.00
I37	<i>Sutterella wadsworthensis</i>	142	NR_104851.1	40545	99.78
I38	<i>Clostridium baratii</i>	143	NR_029229.1	1561	99.85
I39	<i>Anaeromassilibacillus senegalensis</i>	144	NR_144727.1	1673717	97.27
I40	<i>Colidextribacter massiliensis</i>	145	NR_147375.1	1870986	95.00
I41	<i>Flavonifractor plautii</i>	146	NR_029356.1	292800	97.96
I42	[<i>Clostridium</i>] <i>leptum</i>	147	NR_114789.1	1535	94.55

Tables 1 to 4 show the top-hit species names and RefSeq accessions as a result of a BLAST search for the sequences set forth in the SEQ ID NOs on the RefSeq 16s DNA sequence database (as of January 8, 2020). Note that it is generally said %identity>97% can identify species, and >94% can identify genus. Therefore, it should be understood that bacterial strains with a %identity of >94% are bacteria that can be identified at the genus level.

The "at least 70% identity" in the intestinal bacteria of the present invention means that the identity to each base sequence is preferably 80% or more, more preferably 85% or more, further preferably 90% or more (for example, 91% or more, 92% or more, 93% or more, and 94% or more), more preferably 94% or more (for example, 95% or more, 96% or more, 97% or more, and 98% or more), and particularly preferably 99% or more.

In addition, the homology or identity of a sequence (amino acid sequence or nucleotide (base) sequence) can be determined by using the BLAST (Basic Local Alignment Search) program (Altschul et al. J. Mol. Biol., 215: 403-410, 1990). The program is based on the algorithm BLAST by Karlin and Altschul (Proc. Natl. Acad. Sci. USA, 87: 2264-2268, 1990, Proc. Natl. Acad. Sci. USA, 90: 5873-5877, 1993). When analyzing the homology or identity between sequences by BLAST, it can be determined by using, for example, the BLAST of the National Center for

Biotechnology Information (NCBI) (for example, by using the default, that is, initially set parameters).

In the present invention, the "intestinal bacterium" which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 1 to 147 or a base sequence having at least 70% identity to the base sequence is preferably at least 15 bacteria in the intestinal bacterial group, more preferably at least 30 bacteria in the intestinal bacterial group, further preferably at least 75 bacteria in the intestinal bacterial group, more preferably at least 120 bacteria in the intestinal bacterial group, further preferably at least 135 bacteria in the intestinal bacterial group, more preferably at least 140 bacteria in the intestinal bacterial group, further preferably 147 intestinal bacteria each of which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 1 to 147 or a base sequence having at least 70% identity to the base sequence, and particularly preferably 147 bacteria each of which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 1 to 147.

In the present invention, the "intestinal bacterium" which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 1 to 68 or a base sequence having at least 70% identity to the base sequence is preferably at least 7 bacteria in the intestinal bacterial group, more preferably at least 15 bacteria in the intestinal

bacterial group, further preferably at least 35 bacteria in the intestinal bacterial group, more preferably at least 60 bacteria in the intestinal bacterial group, further preferably at least 65 bacteria in the intestinal bacterial group, more preferably 68 intestinal bacteria each of which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 1 to 68 or a base sequence having at least 70% identity to the base sequence, and particularly preferably 68 bacteria each of which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 1 to 68. In addition, the "intestinal bacterium" which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 1 to 68 or a base sequence having at least 70% identity to the base sequence desirably has resistance to ampicillin. Also, the present invention uses 46 bacteria each of which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 1 to 46 or a base sequence having at least 70% identity to the base sequence.

In the present invention, the "intestinal bacterium" which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69 to 105 or a base sequence having at least 70% identity to the base sequence is preferably at least 4 bacteria in the intestinal bacterial group, more preferably at least 8 bacteria in the intestinal bacterial group, further preferably at least 18 bacteria

in the intestinal bacterial group, more preferably at least 29 bacteria in the intestinal bacterial group, further preferably at least 33 bacteria in the intestinal bacterial group, more preferably at least 35 bacteria in the intestinal bacterial group, further preferably 37 intestinal bacteria each of which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69 to 105 or a base sequence having at least 70% identity to the base sequence, and particularly preferably 37 bacteria each of which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69 to 105. In addition, the "intestinal bacterium" which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69 to 105 or a base sequence having at least 70% identity to the base sequence desirably has sensitivity to ampicillin.

In the present invention, the "intestinal bacterium" which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69, 80, 85 to 92, 94, 96, 98 to 101, 103, and 105 or a base sequence having at least 70% identity to the base sequence is preferably at least 2 bacteria in the intestinal bacterial group, more preferably at least 5 bacteria in the intestinal bacterial group, further preferably at least 10 bacteria in the intestinal bacterial group, more preferably at least 14 bacteria in the intestinal bacterial group, further preferably at least 15 bacteria in the intestinal

bacterial group, more preferably at least 16 bacteria in the intestinal bacterial group, further preferably at least 17 bacteria in the intestinal bacterial group, more preferably 18 intestinal bacteria each of which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69, 80, 85 to 92, 94, 96, 98 to 101, 103, and 105 or a base sequence having at least 70% identity to the base sequence, and particularly preferably 18 bacteria each of which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69, 80, 85 to 92, 94, 96, 98 to 101, 103, and 105 (bacteria which include a DNA composed of a base sequence set forth in SEQ ID NO: 69, bacteria which include a DNA composed of a base sequence set forth in SEQ ID NO: 80, bacteria which include a DNA composed of a base sequence set forth in SEQ ID NO: 85, bacteria which include a DNA composed of a base sequence set forth in SEQ ID NO: 86, bacteria which include a DNA composed of a base sequence set forth in SEQ ID NO: 87, bacteria which include a DNA composed of a base sequence set forth in SEQ ID NO: 88, bacteria which include a DNA composed of a base sequence set forth in SEQ ID NO: 89, bacteria which include a DNA composed of a base sequence set forth in SEQ ID NO: 90, bacteria which include a DNA composed of a base sequence set forth in SEQ ID NO: 91, bacteria which include a DNA composed of a base sequence set forth in SEQ ID NO: 92, bacteria which include a DNA

composed of a base sequence set forth in SEQ ID NO: 94,
bacteria which include a DNA composed of a base sequence
set forth in SEQ ID NO: 96, bacteria which include a DNA
composed of a base sequence set forth in SEQ ID NO: 98,
5 bacteria which include a DNA composed of a base sequence
set forth in SEQ ID NO: 99, bacteria which include a DNA
composed of a base sequence set forth in SEQ ID NO: 100,
bacteria which include a DNA composed of a base sequence
set forth in SEQ ID NO: 101, bacteria which include a DNA
10 composed of a base sequence set forth in SEQ ID NO: 103,
and bacteria which include a DNA composed of a base
sequence set forth in SEQ ID NO: 105).

Note that typical examples of 18 bacteria each of
which includes a DNA composed of a base sequence set forth
15 in any of SEQ ID NOs: 69, 80, 85 to 92, 94, 96, 98 to 101,
103, and 105 are accession bacterial strains shown in Table
5 below. All of the bacterial strains were deposited with
the National Institute of Technology and Evaluation (NITE,
2-5-8 Kazusakamatari, Kisarazu City, Chiba Prefecture,
20 Room 122) on March 2, 2020.

[Table 5]

No.	16S rDNA (SEQ ID NO:)	Identification Indication	Receipt Number	Accession Number
F01	69	f01_42H6	NITE ABP-03147	NITE BP-03147
F12	80	f12_42H4	NITE ABP-03148	NITE BP-03148
F17	85	f17_42I7	NITE ABP-03149	NITE BP-03149
F18	86	f18_42I2	NITE ABP-03150	NITE BP-03150
F19	87	f19_43G2	NITE ABP-03151	NITE BP-03151
F20	88	f20_43G1	NITE ABP-03152	NITE BP-03152
F21	89	f21_42A8	NITE ABP-03153	NITE BP-03153
F22	90	f22_43C3	NITE ABP-03154	NITE BP-03154
F23	91	f23_42K4	NITE ABP-03155	NITE BP-03155
F24	92	f24_42I4	NITE ABP-03156	NITE BP-03156
F26	94	f26_42K2	NITE ABP-03157	NITE BP-03157
F28	96	f28_43A3	NITE ABP-03158	NITE BP-03158
F30	98	f30_43A5	NITE ABP-03159	NITE BP-03159
F31	99	f31_43J5	NITE ABP-03160	NITE BP-03160
F32	100	f32_42A7	NITE ABP-03161	NITE BP-03161
F33	101	f33_43N2	NITE ABP-03162	NITE BP-03162
F35	103	f35_42L8	NITE ABP-03163	NITE BP-03163
F37	105	f37_42G1	NITE ABP-03164	NITE BP-03164

Note that as long as the antibacterial action and the like against drug-resistant bacteria or pro-inflammatory bacteria are not impaired, the intestinal bacteria according to the present invention also include bacteria (such as derivative strains and inducible strains) bred from each of these bacteria by mutation treatment, genetic recombination, genome editing, selection of natural mutant strains, and the like.

In the present invention, the "intestinal bacterium" which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 106 to 147 or a base sequence having at least 70% identity to the base sequence is preferably

at least 4 bacteria in the intestinal bacterial group,
more preferably at least 9 bacteria in the intestinal
bacterial group, further preferably at least 22 bacteria
in the intestinal bacterial group, more preferably at
5 least 34 bacteria in the intestinal bacterial group,
further preferably at least 39 bacteria in the intestinal
bacterial group, more preferably at least 41 bacteria in
the intestinal bacterial group, further preferably 42
intestinal bacteria each of which includes a DNA composed
10 of a base sequence set forth in any of SEQ ID NOs: 106 to
147 or a base sequence having at least 70% identity to the
base sequence, and particularly preferably 42 bacteria
each of which includes a DNA composed of a base sequence
set forth in any of SEQ ID NOs: 106 to 147. In addition,
15 the "intestinal bacterium" which includes a DNA composed
of a base sequence set forth in any of SEQ ID NOs: 106 to
147 or a base sequence having at least 70% identity to the
base sequence desirably has sensitivity to ampicillin.

Further, in the present invention, an aspect of the
20 "intestinal bacterium" is an intestinal bacterium that
exhibits resistance to at least one compound selected from
the group consisting of spectinomycin, and/or sensitivity
to at least one compound selected from the group consisting
of ampicillin, tylosin, and chloroform. In addition,
25 another aspect includes intestinal bacteria that exhibit
resistance to metronidazole and/or sensitivity to at least

one compound selected from the group consisting of vancomycin and tylosin.

In addition, as shown in Examples described later, the above-mentioned intestinal bacteria have been isolated
5 by the present inventors and are useful because they exert an antibacterial action against drug-resistant bacteria, pro-inflammatory bacteria, and the like. Therefore, the present invention may also provide the following.

(1) At least one bacterium which includes a DNA composed
10 of a base sequence set forth in any of SEQ ID NOs: 69 to 105 or a base sequence having at least 90% identity to the base sequence.

(2) At least one bacterium which includes a DNA composed
15 of a base sequence set forth in any of SEQ ID NOs: 69, 80, 85 to 92, 94, 96, 98 to 101, 103, and 105 or a base sequence having at least 90% identity to the base sequence.

(3) At least one bacterium specified by any of accession numbers NITE BP-03147 to 03164.

(4) At least one bacterium which includes a DNA composed
20 of a base sequence set forth in any of SEQ ID NOs: 1 to 147 or a base sequence having at least 90% identity to the base sequence.

(5) At least one bacterium which includes a DNA composed
25 of a base sequence set forth in any of SEQ ID NOs: 69 to 105 or a base sequence having at least 90% identity to the base sequence.

(6) At least one bacterium which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 106 to 147 or a base sequence having at least 90% identity to the base sequence.

5 (7) The bacterium according to any one of (1) to (6), which is a bacterium having an antibacterial action against drug-resistant bacteria, pro-inflammatory bacteria, or bacteria that induce proliferation or activation of Th1 cells in the intestinal tract.

10 <Antibacterial Composition and pharmaceutical Composition>

The composition of the present invention may be any as long as it contains the above-mentioned intestinal bacterium, and the bacterium may be a live bacterium or a
15 dead bacterium. In addition, when the compositions can be used in combination, and they are ingested or absorbed in combination as a result (in the case of combined compositions), the above-mentioned intestinal bacteria can also be present separately in two or more compositions.

20 The composition of the present invention can be in the form of a pharmaceutical composition, a quasi-drug composition, food and drink (including animal feed), or a reagent used for research purposes (such as in vitro or in vivo experiments).

25 Since the composition of the present invention exhibits antibacterial activity against drug-resistant

bacteria and the like, it is preferably used as a pharmaceutical composition, a quasi-drug composition, or food or drink for treating, preventing, or ameliorating diseases caused by the bacterium.

5 The composition of the present invention can be formulated by a known pharmaceutical method. For example, it can be used in the form of capsules, tablets, pills, liquids, powders, granules, fine granules, film coating preparations, pellets, troches, sublingual preparations,
10 chews, buccal preparations, pastes, syrups, suspensions, elixirs, emulsions, coating preparations, ointments, plasters, poultices, transdermal preparations, lotions, inhalants, aerosols, injections, suppositories, and the like, for the purpose of administration by oral,
15 parenteral (for example, intestinal, intramuscular, intravenous, tracheal, intranasal, transdermal, intradermal, subcutaneous, intraocular, vaginal, intraperitoneal, rectal, or inhalation), or multiple combinations of these routes.

20 In these formulations, they can be appropriately combined with a pharmacologically or food and drink acceptable carrier, specifically, sterilized water, saline, buffer solution, medium, vegetable oil, solvent, base, emulsifier, suspension, surfactant, stabilizer, flavor
25 agent, air freshener, excipient, vehicle, antiseptic, binder, diluent, isotonic agent, soothing agent, bulking

agent, disintegrant, buffer agent, coating agent, lubricant, colorant, sweetener, thickener, flavor modifier, solubilizer, or other additives.

5 In addition, in these formulations, from the viewpoint of e.g., more efficiently exerting antibacterial activity against drug-resistant bacteria and the like in the intestinal tract, particularly in a formulation intended for oral administration, the composition of the present invention may be combined with a composition that
10 enables efficient delivery into the intestinal tract. The composition capable of such delivery into the intestinal tract is not particularly limited, and a known composition can be appropriately employed, and examples thereof include pH-sensitive compositions, compositions that
15 suppress release to the intestinal tract (such as cellulosic polymers, acrylic acid polymers and copolymers, and vinyl acid polymers and copolymers), bioadhesive compositions that specifically adhere to the intestinal mucosa (such as polymers described in US Patent No.
20 6.368.586), protease inhibitor-containing compositions, and compositions specifically degraded by intestinal enzymes).

25 In addition, when the antibacterial composition of the present invention is used as a pharmaceutical composition, it may further contain a known substance used for treating, preventing, or ameliorating diseases caused

by drug-resistant bacteria and the like (such as other antibacterial agents, anti-inflammatory agents, and immunosuppressants), or may be used in combination with such a substance.

5 When the composition of the present invention is used as food and drink, the food and drink may be, for example, a health food, a functional food, a food for specified health use, a food with nutrient function claims, a food with functional claims, a dietary supplement, a food for
10 patients, or animal feed. Specific examples of the food and drink include liquid foods such as fermented beverages, oil-containing products, soups, milk beverages, refreshing beverages, tea beverages, alcoholic beverages, energy drinks, and jelly-like beverages, carbohydrate-containing
15 foods, processed livestock foods, and processed fishery foods; and processed vegetable foods, semi-solid foods, fermented foods, confectionery, retort products, microwave-compatible foods, and the like. It is also possible to list healthy food and drink prepared in the
20 form of powder, granules, tablets, capsules, liquid, paste, or jelly. The food and drink according to the present invention can be produced by a production technique known in the art. In the food and drink, components (such as nutrients) effective for ameliorating or preventing
25 diseases caused by drug-resistant bacteria and the like may be added. In addition, it may be a multifunctional

food and drink by combining it with other ingredients or other functional foods that exhibit functions other than the amelioration and the like.

5 The product (pharmaceutical product, quasi-drug, food and drink, reagent) of the composition of the present invention or the instruction manual thereof may be provided with an indication that it exhibits antibacterial activity against drug-resistant bacteria and the like, or is used for treating, ameliorating, or preventing diseases
10 caused by the drug-resistant bacteria and the like. Further, in relation to the food and drink, the product or the like of the composition of the present invention may be labeled with a health function as a food with health claims (food for specified health use, food with nutrient
15 function claims, food with functional claims) so that the form, target, and the like can be distinguished from general foods. Here, "the product or the instruction manual is labeled" means that a label is attached to the main body, container, packaging, or the like of the product,
20 or a label is attached to the instruction manual, package insert, promotional material, other printed matter, or the like that discloses product information. Further, the composition of the present invention may be in the form of a kit.

25 Further, as described above, a pharmaceutical composition can be produced by a known formulation

technique using the intestinal bacterium and the like of the present invention. Therefore, the present invention also provides the use of the intestinal bacterium and the like of the present invention for producing a pharmaceutical composition for treating, ameliorating, or preventing diseases caused by drug-resistant bacteria and the like.

<Treatment Method and the Like>

The present invention also provides a method for treating, ameliorating, or preventing diseases caused by drug-resistant bacteria and the like in a target, wherein the target is allowed to ingest the above-mentioned antibacterial composition or pharmaceutical composition, or the above-mentioned intestinal bacteria as an active ingredient thereof (hereinafter also collectively referred to as "the pharmaceutical composition and the like of the present invention or the active ingredient and the like thereof").

In the present invention, the "drug-resistant bacterium" means a bacterium that has resistance to antibacterial agents (such as antibiotics) and the drugs are ineffective or difficult to be effective. Further, the drug may be one drug or multiple drugs. That is, the drug-resistant bacteria according to the present invention also include multidrug-resistant bacteria. Such bacteria are not particularly limited, but examples thereof include

carbapenem-resistant Enterobacteriaceae (CRE, KPC-2-producing *Klebsiella pneumoniae*, and the like), vancomycin-resistant enterococci (VRE, bacteria with vancomycin-resistant gene (*vanA*), and the like),
5 *Clostridium difficile*, and *Campylobacter jejuni*. More specific examples include *Klebsiella pneumoniae* (ATCC BAA-1705), *Enterococcus faecium* (Orla-Jensen) Schleifer and Kilpper-Balz (ATCC 700221), *Clostridioides difficile* (Prevot) Lawson et al. (ATCC 43255, bacterial strain
10 designation: VPI 10463), *Clostridioides difficile* (Prevot) Lawson et al. (ATCC BAA-1382, bacterial strain designation: 630), and *Campylobacter jejuni* 81-176 (ATCC BAA2151).

The "diseases caused by drug-resistant bacteria"
15 include infectious diseases caused by drug-resistant bacteria. They also include diseases associated with the infection. Examples of such diseases include respiratory infectious diseases such as sepsis, peritonitis, meningitis, gastroenteritis, and pneumonia, urinary tract
20 infectious diseases, surgical site infectious diseases, soft tissue infectious diseases, and medical device-related infectious diseases (such as medical device-related bloodstream infectious diseases).

In the present invention, the "pro-inflammatory
25 bacterium" means a bacterium that induces inflammation in the intestinal tract, and examples thereof include

adherent-invasive *Escherichia coli* (AIEC). More specifically, AIEC LF82 can be mentioned.

The "diseases caused by pro-inflammatory bacteria" include diseases caused or involved in inflammation caused by the bacteria. Examples of such diseases include inflammatory bowel diseases (including chronic inflammatory bowel diseases such as Crohn's disease, ulcerative colitis, and inflammatory bowel disease).

The pharmaceutical composition and the like of the present invention or the active ingredient and the like thereof can be used for animals including humans, but there are no particular restrictions on animals other than humans, and various domestic animals, poultry, pets, laboratory animals, and the like can be targeted.

In addition, the ingestion target of the intestinal bacterium and the like of the present invention includes animals carrying the drug-resistant bacteria and the like, regardless of the onset of diseases caused by the drug-resistant bacteria and the like. From the viewpoint of prevention, an animal that does not possess the bacteria or is suspected of possessing them may be allowed to ingest the pharmaceutical composition and the like of the present invention or the active ingredient and the like thereof.

The method for ingesting the pharmaceutical composition and the like of the present invention or the active ingredient and the like thereof is not particularly

limited, and may be oral administration or parenteral administration (for example, administration into the intestinal tract), and in the case of oral administration, from the viewpoint of further improving the effects of the pharmaceutical composition and the like of the present invention or the active ingredient and the like thereof, it is preferable that the ingestion target of the pharmaceutical composition and the like of the present invention or the active ingredient and the like thereof has reduced production of gastric acid by ingestion of a proton pump inhibitor (PPI) or the like.

In addition, in the case of ingesting the pharmaceutical composition and the like of the present invention or the active ingredient and the like thereof, the amount ingested can be appropriately selected by those skilled in the art according to the target's age, weight, disease symptoms, health conditions, composition type (such as pharmaceutical product, and food and drink), ingestion method, and the like.

Preferable embodiments of the antibacterial composition and the pharmaceutical composition as well as the therapeutic method and the like of the present invention have been described above, but the present invention is not limited to the above embodiments.

As shown in Examples described later, regarding the ability to suppress intestinal colonization possessed by

the Th1 cell-inducible bacterium *Klebsiella* 2H7 strain (Kp2H7), the present inventors have succeeded in selecting 18 strains capable of exerting the same level of suppression ability as the 37 strains of intestinal bacteria derived from healthy subject #F. Therefore, the present invention may also provide the following aspects with respect to antibacterial compositions and pharmaceutical compositions as well as treatment methods and the like.

<1> An antibacterial composition against Th1 cell-inducible bacteria, containing an intestinal bacterium as an active ingredient, wherein the intestinal bacterium is at least one bacterium which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69, 80, 85 to 92, 94, 96, 98 to 101, 103, and 105 or a base sequence having at least 90% identity to the base sequence.

<2> The antibacterial composition according to <1>, which is a pharmaceutical composition.

<3> The antibacterial composition according to <1> or <2>, which is a pharmaceutical composition for treating, ameliorating, or preventing diseases caused by Th1 cells.

<4> A method for suppressing proliferation or activation of Th1 cells in a target, a method for suppressing immunity in the target, or a method for treating, ameliorating, or preventing diseases caused by Th1 cells in the target, including allowing the target to ingest the antibacterial

composition according to any one of <1> to <3> or at least one bacterium which includes a DNA composed of a base sequence set forth in any of SEQ ID NOs: 69, 80, 85 to 92, 94, 96, 98 to 101, 103, and 105 or a base sequence having
5 at least 90% identity to the base sequence.

The "Th1 cell-inducible bacterium" according to the present invention is a bacterium that normally exists in the human oral cavity, and induces the proliferation or activation of Th1 cells by colonizing in the intestinal
10 tract. It is preferably a bacterium that belongs to Klebsiella, more preferably belonging to Klebsiella pneumoniae or Klebsiella aeromobilis, and induces the proliferation or activation of Th1 cells in the intestinal tract. Further, the "Th1 cell-inducible bacterium" is
15 preferably a bacterium that easily colonizes in an intestinal environment in which the diversity has changed as compared with the healthy state due to the administration of an antibacterial agent. It is also a bacterium that easily colonizes in an intestinal
20 environment in which the diversity has changed as compared with the healthy state due to colitis or the like.

For an example of the "Th1 cell-inducible bacterium," PTL 1 can be referred to, and typical examples include Kp2H7 strain, KallE12 strain, 34E1 strain, BAA-1705 strain,
25 700603 strain, and 40B3 strain belonging to Klebsiella. Among these, the Kp2H7 strain or the KallE12 strain is

more preferable, and the Kp2H7 strain is particularly preferable. See Table 6 for details of these bacteria.

[Table 6]

Bacterial Name	Supplier	Information from Supplier	Registry number
KCTC2242	KCTC	http://kctc.kribb.re.kr/English/_SearchView.aspx?sn=2242	NCBI Taxonomy ID: 1049565
2552	ATCC	https://www.atcc.org/Products/All/BAA-2552.aspx	NCBI Taxonomy ID: 507522
KP-1	—	—	NCBI Taxonomy ID: 1365186
700721	ATCC	https://www.atcc.org/Products/All/700721.aspx	NCBI Taxonomy ID: 272620
13882	JCM	https://www.atcc.org/Products/All/13882.aspx	NCBI Taxonomy ID: 1913574
40B3	—	—	SAMD00083913
34E1	—	—	SAMD00083911
1705	ATCC	https://www.atcc.org/Products/All/BAA-1705.aspx	NCBI Taxonomy ID: 1276652
11E12	—	—	SAMD00083912
700603	ATCC	https://www.atcc.org/Products/All/700603.aspx	NCBI Taxonomy ID: 1276653
2H7	—	—	SAMD00083910

In addition, examples of the "Th1 cell-inducible bacterium" of the present invention include a bacterium which contains a DNA composed of a nucleotide sequence having 90% or more (91% or more, 92% or more, 93% or more, 5 94% or more, 95% or more, 96% or more, 97% or more, 98% or more, and 99% or more) identity to the nucleotide sequence encoding the 16S rRNA of Kp2H7 strain, KallE12 strain, 34E1 strain, BAA-1705 strain, 700603 strain, or 40B3 strain, and also include a bacterium which contains a DNA 10 composed of a nucleotide sequence having 70% or more (preferably 80% or more, more preferably 85% or more, further preferably 90% or more, and more preferably 94% or more (for example, 95% or more, 96% or more, 97% or more, 98% or more, and 99% or more)) homology or identity to the 15 nucleotide sequence specific to Kp2H7 strain, KallE12 strain, 34E1 strain, BAA-1705 strain, 700603 strain, or 40B3 strain.

In the present invention, the "Th1 cell" is a subgroup of CD4 positive helper T-cells (Th cells) and 20 means a cell that enhances cell-mediated immunity. In addition, the meaning of "activation of Th1 cells" includes the production of Th1 cytokines (such as IFN- γ) by the cells, induction of macrophages by the cytokines, activation of cells such as cytotoxic T-cells (CTL), and 25 enhancement of cell-mediated immunity by the activation. Further, the meaning of "induction of the proliferation or

activation of Th1 cells" also includes induction of differentiation from naive T-cells to Th1 cells which leads to proliferation or activation of Th1 cells.

5 The effect of inducing proliferation or activation of Th1 cells in the intestinal tract can be evaluated by quantitatively detecting Th1 cell-specific markers (such as CD4 and IFN- γ). Such quantitative detection can be performed by a known method, and can be performed by methods for detection using antibodies (immunological
10 methods) such as flow cytometry, imaging cytometry, ELISA method, radioimmunoassay, immunohistochemical staining, immunoprecipitation, immunoblotting, and antibody array analysis.

Regarding whether or not any bacterium or the like
15 has an effect of inducing proliferation or activation of Th1 cells in the intestinal tract, for example, it can be determined that the bacterium or the like has an effect of inducing proliferation or activation of Th1 cells in the intestinal tract when the proportion of IFN- γ^+ cells in
20 CD4⁺TCR β^+ T-cells in the intestinal tract detected by flow cytometry is 10% or more (it is preferable to determine, when the proportion is 25% or more, that the bacterium or the like has an effect of inducing proliferation or activation of Th1 cells in the intestinal tract, and it is
25 more preferable to determine, when the proportion is 30% or more, that the bacterium, substance, or the like has an

effect of inducing proliferation or activation of Th1 cells in the intestinal tract).

In the present invention, the "diseases caused by Th1 cells" mean diseases induced by proliferation or activation of Th1 cells, and examples thereof include inflammatory bowel diseases (including chronic inflammatory bowel diseases such as Crohn's disease, ulcerative colitis, and inflammatory bowel disease), type 1 diabetes, rheumatoid arthritis, experimental autoimmune encephalomyelitis (EAE), multiple sclerosis, autoimmune diseases such as systemic lupus erythematosus, and chronic inflammatory diseases. In addition, the "immunity" suppressed in the present invention includes not only mucosal immunity (such as intestinal immunity) but also systemic immunity. Furthermore, not only cell-mediated immunity but also humoral immunity is included.

In addition, when the antibacterial composition of the present invention is used as a pharmaceutical composition, it may further contain known substances (such as anti-inflammatory agents and immunosuppressants) used for the treatment, prevention, or amelioration of diseases caused by Th1 cells, or may be used in combination with such substances.

Note that for other phrases in <1> to <4>, refer as appropriate to the above-mentioned <Antibacterial Composition and Pharmaceutical Composition> and <Treatment

Method and the Like>.

<Composition for Examining Diseases Caused by Drug-Resistant Bacteria and the Like>

In the present invention, the presence of intestinal
5 bacteria that can suppress the colonization and the like
of drug-resistant bacteria and the like in the intestinal
tract has been clarified. Therefore, by detecting the
presence of the intestinal bacteria, it becomes possible
to examine diseases caused by drug-resistant bacteria and
10 the like.

Therefore, the present invention provides a
composition for examining diseases caused by the following
drug-resistant bacteria and the like.

A composition for examining diseases caused by drug-
15 resistant bacteria and the like, containing an antibody
that specifically recognizes the intestinal bacterium and
the like of the present invention.

A composition for examining diseases caused by drug-
resistant bacteria and the like, containing a
20 polynucleotide for detecting a nucleotide sequence
specific to the drug-resistant bacteria and the like of
the present invention.

In the present invention, the "antibody that
specifically recognizes the intestinal bacterium and the
25 like of the present invention" may be a polyclonal antibody,
a monoclonal antibody, or a functional fragment of

antibody (for example, Fab, Fab', F(ab')₂, variable region fragment (Fv), disulfide bond Fv, single chain Fv (scFv), sc(Fv)₂, diabody, multispecific antibody, or a polymer thereof) as long as it can specifically recognize the
5 bacterium. If the antibody of the present invention is a polyclonal antibody, it can be obtained by immunizing a host animal with an antigen (peptide, polynucleotide, sugar chain, lipid, or the like derived from the intestinal bacterium and the like of the present invention) and
10 purifying it from its antiserum by a conventional means (such as salting-out, centrifugation, dialysis, or column chromatography). In addition, the monoclonal antibody can be produced by a hybridoma method or a recombinant DNA method.

15 Further, as the antibody used for the examination of the present invention, it is possible to use an antibody bound with a labeling substance. By detecting the labeling substance, it is possible to directly measure the amount of antibody bound to the intestinal bacterium and the like
20 of the present invention or a substance derived from the bacterium. The labeling substance is not particularly limited as long as it can bind to an antibody and can be detected by a chemical or optical method, and examples thereof include fluorescent dyes (such as GFP), enzymes
25 (such as HRP), and radioactive substances.

In addition to the antibody component, the

examination composition of the present invention may contain additional components acceptable as a composition. Examples of the additional components include carriers, excipients, disintegrants, buffer agents, emulsifiers, suspensions, stabilizers, preservatives, antiseptics, saline, labeling substances, and secondary antibodies. In addition to the above examination compositions, it is possible to combine substrates required for detection of labeling substances, positive controls and negative controls, buffer solutions used for diluting or washing the sample, tubes or plates used for the reaction of the sample with the antibody of the present invention, and the like, or use as an examination kit for diseases caused by drug-resistant bacteria and the like is possible. In addition, when an unlabeled antibody is used as an antibody standard sample, it is possible to combine a labeled substance (such as secondary antibody, protein G, or protein A) that binds to the antibody. Further, the kit for examining diseases caused by drug-resistant bacteria and the like can include an instruction manual for use of the kit.

Further, the examination composition of the present invention can be combined with an apparatus for detecting the antibody of the present invention. Examples of the apparatus include a flow cytometry apparatus and a microplate reader.

In the present invention, the "polynucleotide for detecting a nucleotide sequence specific to the intestinal bacterium and the like of the present invention" is not particularly limited as long as the sequence specific to the bacterium is detected, and examples thereof include polynucleotides according to any of the following (a) and (b), each having a chain length of at least 15 nucleotides.

(a) A polynucleotide that is a pair of primers designed to sandwich the specific nucleotide sequence, and

(b) A polynucleotide that is a primer or probe that hybridizes to a nucleotide sequence containing the specific nucleotide sequence.

The polynucleotide of the present invention has a base sequence complementary to the nucleotide sequence of the intestinal bacterium and the like of the present invention. Here, "complementary" does not have to be perfectly complementary as long as it hybridizes. These polynucleotides usually have 80% or more, preferably 90% or more, more preferably 95% or more, and particularly preferably 100% homology to the nucleotide sequence.

The "chain length" of the polynucleotide of the present invention, when used as a primer, is usually 15 to 100 nucleotides, preferably 17 to 30 nucleotides, and more preferably 20 to 25 nucleotides. In addition, when used as a probe, it is usually 15 to 1000 nucleotides, and preferably 20 to 100 nucleotides.

The polynucleotide of the present invention may be a DNA or RNA, or may have its nucleotide replaced with an artificial nucleic acid such as LNA (registered trademark, crosslinked nucleic acid), ENA (registered trademark, 2'-O, 4'-C-Ethylene-bridged nucleic acids), GNA (glycerol nucleic acid), TNA (threose nucleic acid), or PNA (peptide nucleic acid) in part or all of it.

Note that the polynucleotide of the present invention can be chemically synthesized using a commercially available nucleotide automatic synthesizer or the like. Further, as the polynucleotide used for the examination of the present invention, it is possible to use a polynucleotide bound with a labeling substance. The labeling substance is not particularly limited as long as it can bind to a polynucleotide and can be detected by a chemical or optical method, and examples thereof include fluorescent dyes (such as DEAC, FITC, R6G, TexRed, and Cy5) as well as dyes such as DAB (chromogen), enzymes, and radioactive substances in addition to fluorescent dyes.

In addition to the above-mentioned polynucleotide, the examination composition of the present invention may contain additional pharmacologically acceptable components. Examples of the additional components include buffer agents, emulsifiers, suspensions, stabilizers, antiseptics, and saline.

In addition to the above examination compositions,

it is possible to combine standard samples such as substrates required for detection of labeling substances added to the polynucleotide, positive controls and negative controls, and buffer solutions used for diluting or washing the sample, thereby combining tubes or plates used for the reaction of the sample with the polynucleotide of the present invention, or use as an examination kit for diseases caused by drug-resistant bacteria and the like is possible. Moreover, the kit for examining diseases caused by drug-resistant bacteria and the like can include an instruction manual for use of the kit.

In addition, the examination composition of the present invention can be combined with an apparatus for detecting a nucleotide sequence specific to the intestinal bacterium and the like of the present invention. Examples of the apparatus include a thermal cycler, a sequencer, and a microarray.

Furthermore, the present invention also provides a method for examining diseases caused by drug-resistant bacteria and the like using the above-mentioned antibody, polynucleotide, or examination composition. Specifically, the present invention provides

a method for examining diseases caused by drug-resistant bacteria and the like, including the steps of contacting the antibody, polynucleotide, or examination composition with a sample isolated from a subject, and

detecting the presence or absence of an intestinal bacterium and the like of the present invention in the intestinal tract by the contact.

The subject is not particularly limited, and examples thereof include animals such as humans suspected of suffering from a disease caused by drug-resistant bacteria and the like. Further, the sample isolated from the subject is not particularly limited, but a fecal sample of a subject, a culture thereof, or a polypeptide, polynucleotide, sugar chain, lipid or the like extracted therefrom is preferably used in the method of the present invention.

As the method for detecting the presence or absence of an intestinal bacterium and the like of the present invention by contacting the sample with the antibody of the present invention or an examination composition containing the same, examples thereof include methods for detection using antibodies (immunological methods) such as ELISA method, immunoblotting, antibody array analysis, immunohistochemical staining, flow cytometry, imaging cytometry, radioimmunoassay, and immunoprecipitation.

Further, as the method for detecting the presence or absence of an intestinal bacterium and the like of the present invention by contacting the sample with the polynucleotide of the present invention or an examination composition containing the same, it is possible to use PCR

(RT-PCR, real-time PCR, and quantitative PCR), DNA microarray analysis, Northern blotting, 16s rRNA sequencing, next-generation sequencing (synthetic sequencing (sequencing-by-synthesis, for example, sequencing by Solexa Genome Analyzer or Hiseq (registered trademark) 2000 manufactured by Illumina), pyrosequencing (for example, sequencing by a sequencer GSLX or FLX manufactured by Roche Diagnostics (454) (so-called 454 sequencing)), and ligase reaction sequencing (for example, sequencing by Solid (registered trademark) or 5500xl manufactured by Life Technologies)), bead array method, in situ hybridization, dot blotting, RNase protection assay, mass spectrometry, genomic PCR method, and Southern blotting.

In the present invention, the "examination" of a disease caused by drug-resistant bacteria and the like includes not only the presence or absence of the onset of the disease but also the risk of the onset of the disease, and if the presence of an intestinal bacterium and the like of the present invention is detected in the intestinal tract by the above-mentioned method, it can be determined that a disease caused by drug-resistant bacteria and the like has not developed or the risk of developing the disease is low.

Diagnosis of a disease caused by drug-resistant bacteria and the like in a subject is usually made by a

doctor (including a person who has been instructed by a doctor), but the data obtained by the method of the present invention is useful for diagnosis by a doctor. Therefore, the method of the present invention can also be expressed
5 as a method of collecting and presenting data useful for diagnosis by a doctor.

Further, the present invention can also provide a companion diagnostic method using the above-mentioned examination method and a drug thereof. That is, the
10 present invention also provides the following.

A method for determining the effectiveness of the pharmaceutical composition and the like of the present invention or the active ingredient and the like thereof in the treatment, amelioration, or prevention of diseases
15 caused by drug-resistant bacteria and the like, including the steps of contacting the antibody, polynucleotide, or examination composition with a sample isolated from a subject, and detecting the presence or absence of the intestinal bacteria and the like by the contact, wherein
20 in the step, when the presence of the bacteria is not detected, it is determined that the pharmaceutical composition and the like of the present invention or the active ingredient and the like thereof in the subject is highly effective in treating, ameliorating, or preventing
25 the disease.

A method for treating, ameliorating, or preventing

diseases caused by drug-resistant bacteria and the like, including a step of allowing a patient determined by the determination method to have high effectiveness of the pharmaceutical composition and the like of the present invention or the active ingredient and the like thereof to ingest the pharmaceutical composition and the like or the active ingredient and the like thereof.

A composition for treating, ameliorating, or preventing diseases caused by drug-resistant bacteria and the like, containing the intestinal bacterium and the like of the present invention as an active ingredient, wherein the composition is ingested by a subject determined to have high effectiveness by the determination method.

<Method for Screening Intestinal Bacteria Having Antibacterial Activity against Drug-Resistant Bacteria and the Like in Intestinal Tract>

It has also been clarified for the first time by the present inventors that among intestinal bacteria, there are bacteria that suppress the colonization of drug-resistant bacteria and the like in the intestinal tract. Therefore, the present invention provides a method for screening intestinal bacteria having antibacterial activity against drug-resistant bacteria and the like, including the following steps.

A step of allowing a non-human germ-free animal to ingest drug-resistant bacteria and the like and test

intestinal bacteria in the intestinal tract,

a step of detecting the drug-resistant bacteria and the like in the intestinal tract of the non-human germ-free animal, and

5 a step of determining the test intestinal bacteria as intestinal bacteria having antibacterial activity against the drug-resistant bacteria and the like when the number of bacteria detected in the step is reduced as compared with the case where the test intestinal bacteria
10 are not ingested.

The "drug-resistant bacteria and the like" are as described above. The "non-human germ-free animal" means a non-human animal that is born and grown under germ-free conditions. Examples of non-human animals include, but
15 are not limited to, mice, rats, monkeys, pigs, cows, horses, sheep, goats, chickens, ducks, ostriches, domestic ducks, dogs, cats, rabbits, and hamsters. Moreover, in these animals, mice are preferably used.

The test intestinal bacteria to be ingested by non-
20 human germ-free animals may be any bacteria present in the intestines of animals, examples of such animals include humans and non-human animals (such as mice, rats, monkeys, pigs, cows, horses, sheep, goats, chickens, ducks, ostriches, domestic ducks, dogs, cats, rabbits, and
25 hamsters). In addition, the test intestinal bacteria to be ingested by non-human germ-free animals may be isolated

intestinal bacteria, and examples thereof include a sample containing intestinal bacteria (for example, a fecal sample of the animal or a culture thereof).

Further, the method for "ingesting" the test intestinal bacteria and drug-resistant bacteria and the like to a non-human animal is not particularly limited, and is usually performed by oral administration, but may be parenteral administration (for example, administration into the intestinal tract). In addition, the test intestinal bacteria and drug-resistant bacteria and the like may be ingested at the same time, the drug-resistant bacteria and the like may be ingested by the non-human animal after the test intestinal bacteria are ingested by the animal, or the test intestinal bacteria may be ingested by the non-human animal after the drug-resistant bacteria and the like are ingested by the animal.

The "detection" of drug-resistant bacteria and the like in the intestinal tract can be performed by detecting a nucleotide sequence specific to the drug-resistant bacteria and the like. Examples of the detection method include PCR (RT-PCR, real-time PCR, and quantitative PCR), DNA microarray analysis, Northern blotting, 16s rRNA sequencing, next-generation sequencing (synthetic sequencing (sequencing-by-synthesis, for example, sequencing by Solexa Genome Analyzer or Hiseq (registered trademark) 2000 manufactured by Illumina), pyrosequencing

(for example, sequencing by a sequencer GSIX or FLX manufactured by Roche Diagnostics (454) (so-called 454 sequencing)), and ligase reaction sequencing (for example, sequencing by Solid (registered trademark) or 5500xl manufactured by Life Technologies)), bead array method, in situ hybridization, dot blotting, RNase protection assay, mass spectrometry, genomic PCR method, and Southern blotting.

Further, the "detection" of drug-resistant bacteria and the like in the intestinal tract can be performed, for example, by detecting an amino acid sequence specific to the drug-resistant bacteria and the like. Examples of the detection method include methods for detection using antibodies (immunological methods) such as ELISA method, immunoblotting, antibody array analysis, immunohistochemical staining, flow cytometry, imaging cytometry, radioimmunoassay, and immunoprecipitation. The timing of detection is not particularly limited, and those skilled in the art can appropriately adjust the timing according to the type and the like of animal to be used.

Note that if the screening method of the present invention fails to select intestinal bacteria having antibacterial activity against drug-resistant bacteria and the like in a single trial, an intestinal sample containing the obtained bacteria is ingested by a new non-human germ-free animal as the next test intestinal bacteria, and the

above-mentioned screening is performed multiple times, making it possible to isolate the intestinal bacteria having the antibacterial activity.

[Examples]

5 (Example 1)

As shown in the upper part of Fig. 1, germ-free mice were administered with Klebsiella 2H7 strains (Kp2H7), and one week later, fecal samples of healthy volunteers were administered.

10 Specifically, the germ-free mice used were C57BL/6N at 4 to 8 weeks of age (CLEA Japan, Inc.), which were kept in a rearing vinyl isolator (germ-free isolator) (manufactured by ICM Co., Ltd.; ICM-1B) under the conditions of free-drinking water and feeding for one week
15 or longer to acclimate them to the environment. The age at the start of the experiment was 8 to 14 weeks of age. The same applies to the other Examples in the present specification.

20 A bacterial solution of Klebsiella was placed in an LB liquid medium and cultured overnight at 37°C to adjust the OD value to 1.2 (equivalent to 1×10^9 CFU/mL), and 200 μ L/individual (equivalent to 2×10^8 CFU/individual) of bacterial solution was administered into the stomach of mice using a sonde.

25 For the fecal samples, feces provided by Japanese healthy subject volunteers (#A, #F, #I, #J, and #K) were

diluted 5-fold by weight with a glycerol PBS solution (final concentration of glycerol: 20% by volume), which was filtered through a 100 μ m diameter filter, and the resultant was stored as a stock solution at -80°C. At the
5 time of fecal administration, the stock solution was diluted 10-fold with PBS in an anaerobic chamber, and administered in an amount of 200 μ L/individual into the stomach of each mouse using a sonde.

Mouse fecal samples were dissolved in a solution of
10 glycerol (final concentration 20%) and EDTA (final concentration 10 mM) mixed in PBS at a proportion of 50 mg feces/mL. The fecal lysate was seeded after appropriate dilution in DHL medium containing 50 mg/L ampicillin and 50 mg/L spectinomycin, and after culturing at 37°C
15 overnight, the number of colonies was counted and the number of CFUs per 1 g of feces was calculated.

In the other Examples in the present specification, bacterial solution of Klebsiella and feces were administered in the same manner, and CFUs were counted in
20 the same manner.

As a result, as shown in the lower part (graph) of Fig. 1, five types of feces were used, and a significant decrease in the bacterial amount of Kp2H7 was observed in all the samples.

25 (Example 2) Isolation of Bacteria from Feces of Healthy Subject Volunteers

Each of the frozen fecal samples derived from feces of healthy subjects F, K, and I (feces F, feces K, and feces I) prepared in Example 1 was thawed at room temperature, diluted with PBS, and cultured in an agar plate of EG medium, modified GAM agar medium (Nissui Pharmaceutical Co., Ltd.; 05426), REINFORCED CLOSTRIDIAL AGAR (RCM AGAR) (Thermo Fisher Scientific Inc; CM0151), or Schaedler blood medium (manufactured by Wako; 517-45805) under an anaerobic environment of at 37°C and 10% CO₂, and the colonies formed were isolated. From feces F, 37 strains were isolated, 42 strains were isolated from feces I, and 47 strains were isolated from feces K. After that, the feces K was isolated again, and finally 68 strains were isolated from the feces K.

For the isolated bacteria, the gene sequence was analyzed and the bacterial species was estimated by 16S rDNA analysis by the Sanger method. Sequence analysis was performed using 3130 DNA Analyzer manufactured by Thermo Fisher Scientific and a primer set having the following sequences.

27 Forward-mod: 5'-AGRGTTTGATYMTGGCTCAG-3' (SEQ ID NO: 148)

1492 Reverse: 5'-GGYTACCTTGTTACGACTT-3' (SEQ ID NO: 149)

Here, R: A or G, Y: C or T, and M: A or C.

Furthermore, the genome sequences of the 37 bacterial strains derived from feces F were determined using a next-

generation sequencer. That is, genome sequencing was performed using MiSeq manufactured by Illumina and Sequel manufactured by Pacific Biosciences, and all genome sequences were obtained by hybrid assembly using Unicycler.

By extracting the 16S rRNA sequence using RNAmmer for each of these genome sequences, a more accurate sequence was obtained, including both terminal sequences that could not be determined by the 16S rDNA sequence determined by the Sanger method.

Tables 1 to 4 show the results of this analysis. In addition, Fig. 2 shows the results of 16S meta-analysis of three types of feces derived from donors F, I, and K.

(Example 3)

As shown in the upper part of Fig. 3, Kp2H7 was administered to germ-free mice, and one week later, mixed isolated bacteria (37 strains derived from feces F (F37mix), 42 strains derived from feces I (I42mix), and 47 strains derived from feces K (K47mix)) or feces I were administered.

The isolated bacteria were cultured in mGAM liquid medium, EG medium, or CM0149 medium at 37°C in an anaerobic chamber for 24 to 48 hours, and mixed. The mixed solution was concentrated 5 times, and 200 μ L/individual (equivalent to total bacterial amount 1×10^9 CFU/animal) of bacterial solution was administered into the stomach using a sonde. The mixed isolated bacterial strains in

the following Examples were also administered in the same manner.

As a result, as shown in the lower part (graph) of Fig. 3, it was clarified that the 37 strains derived from feces F had the same activity as feces I in terms of the ability to eliminate *Klebsiella* from the mouse intestinal tract.

(Example 4)

As shown in the upper part of Fig. 4, Kp2H7 was administered to germ-free mice, and one week later, the mixed isolated bacteria (37 strains derived from feces F and 68 strains derived from feces K) were administered. As a result, as shown in the lower part (graph) of Fig. 4, 37 strains derived from feces F and 68 strains derived from feces K equally eliminated *Klebsiella* from the mouse intestinal tract.

(Example 5)

As shown in the upper part of Fig. 5, Kp2H7 was administered to germ-free mice, and one week later, mixed isolated bacteria (F37mix) were administered, and 200 mg/L of ampicillin was administered by drinking water after the administration of the isolated bacteria. In order to investigate the change in the bacterial amount of each of the administered bacteria, PCR was performed using a specific primer of each of the bacteria. Table 7 shows the primers used in the analysis.

[Table 7]

	Forward Primer Name	SEQ ID NO:	Reverse Primer Name	SEQ ID NO:
f01	f01_42H6_Fw1	150	f01_42H6_Rv1	151
f02	f02_42I8_Fw1	152	f02_42I8_Rv3	153
f05	f05_43D4_Fw4	154	f05_43D4_Rv2	155
f06	f06_42F4_Fw1	156	f06_42F4_Rv1	157
f07	f07_42J6_Fw1	158	f07_42J6_Rv1	159
f08	f08_42B5_Fw1	160	f08_42B5_Rv1	161
f09	f09_43F1_Fw1	162	f09_43F1_Rv1	163
f10	f10_43J3_Fw1	164	f10_43J3_Rv1	165
f12	f12_42H4_Fw1	166	f12_42H4_Rv1	167
f13	f13_42H8_Fw1	168	f13_42H8_Rv1	169
f14	f14_42L4_Fw1	170	f14_42L4_Rv1	171
f16	f16_43M1_Fw2	172	f16_43M1_Rv2	173
f17	f17_42I7_Fw3	174	f17_42I7_Rv3	175
f18	f18_42I2_Fw5	176	f18_42I2_Rv5	177
f19	f19_43G2_Fw2	178	f19_43G2_Rv2	179
f20	f20_43G1_Fw2	180	f20_43G1_Rv2	181
f21	f21_42A8_Fw2	182	f21_42A8_Rv6	183
f22	f22_43C3_Fw2	184	f22_43C3_Rv2	185
f24	f24_42I4_Fw1	186	f24_42I4_Rv1	187
f25	f25_42J1_Fw2	188	f25_42J1_Rv2	189
f26	f26_42K2_Fw1	190	f26_42K2_Rv1	191
f27	f27_42J5_Fw2	192	f27_42J5_Rv2	193
f28	f28_43A3_Fw1	194	f28_43A3_Rv1	195
f29	f29_43J8_Fw2	196	f29_43J8_Rv2	197
f30	f30_43A5_Fw1	198	f30_43A5_Rv1	199
f31	f31_43J5_Fw1	200	f31_43J5_Rv2	201
f32	f32_42A7_Fw1	202	f32_42A7_Rv2	203
f33	f33_43N2_Fw1	204	f33_43N2_Rv1	205
f34	f34_43K4_Fw1	206	f34_43K4_Rv1	207
f35	f35_42L8_Fw2	208	f35_42L8_Rv2	209
f37	f37_42G1_Fw3	210	f37_42G1_Rv4	211

As a result, as shown in the lower part of Fig. 5, the bacterial amount of *Klebsiella* was transiently increased by the administration of ampicillin, but decreased again thereafter.

5 In addition, the change over time of the abundance ratio of each isolated bacterium in the total bacterial amount was analyzed together with that of Kp2H7. Figs. 6A to 6H show the results obtained. Furthermore, the bacterial amount of *Klebsiella* and the Spearman's rank correlation coefficient of each bacterium were calculated and arranged in order of high positive correlation. Fig. 10 7 shows the results obtained.

As shown in Fig. 7, it was found that the strains belonging to *Bacteroidetes* moved regardless of the movement of *Klebsiella*. On the other hand, many of the 15 strains having a negative correlation belonged to the genus *Furmicutes*.

(Example 6)

As shown in Fig. 9, 37 the strains derived from feces 20 F were divided into 8 strains belonging to *Bacteroidetes* (F8mix) and the 29 other bacterial strains (F29mix), and mixed respectively, and after Kp2H7 was colonized in germ-free mice, the mixed isolated bacteria were administered. Note that the phylogenetic trees shown in Figs. 9, 10, and 25 12 were prepared by the Neighbor-joining method using MEGA X with the DNA base sequence of the 16S rDNA analysis

result of the isolated bacteria by the Sanger method.

As a result, as shown in Fig. 8, F37mix and F29mix equally eliminated *Klebsiella* from the mouse intestinal tract. On the other hand, in the F8mix administration group belonging to *Bacteroidetes*, the bacterial amount of *Klebsiella* did not change, suggesting that F8mix is not related to the elimination of *Klebsiella*.

(Example 7)

From the 37 strains, 18 strains were selected by excluding strains belonging to *Bacteroidetes*, strains duplicate at the 16S rDNA level, strains disappeared by ampicillin administration, and strains showing behavior unrelated to *Klebsiella* (see Fig. 10).

Then, as shown in the upper part of Fig. 11, the Kp2H7 strains were administered to germ-free mice, and one week later, mixed isolated bacteria (37 strains derived from feces F (F37mix), 18 strains (F18mix), and 42 strains derived from feces I (I42mix)) were administered.

As a result, as shown in the lower part (graph) of Fig. 11, it was clarified that the 18 strains shown in Fig. 10 was also able to exhibit the ability to eliminate *Klebsiella* strains as much as the 37 strains.

(Example 8)

The 18 strains shown in Fig. 10 were divided into 4 groups based on the phylogenetic tree (*Blautia*, *Lachnospirillum*, other *Firmicutes*, and other *Phyla*) (see

Fig. 12). Furthermore, these 4 groups were subtracted from the 18 bacterial strains (F18mix) to prepare bacterial strain groups of F15mix (F18mix-other phyla), F12mix (F18mix-Lachnoclostridium), F14mix (F18mix-Blautia), F13mix (F18mix-other Firmicutes). In addition, a bacterial strain set was also prepared by mixing 13 strains (F13mix (F31-18mix)), which were obtained by removing the duplicated strains from the 37 strains and further removing the above-described 18 strains from the remaining 31 strains. Then, as shown in the upper part of Fig. 13, Kp2H7 was administered to germ-free mice, and one week later, each isolated bacterium mixed as described above was administered.

As a result, as shown in the lower part (graph) of Fig. 13, F18mix best eliminated *Klebsiella*. However, the bacterial amount of *Klebsiella* significantly increased regardless of which group was excluded. In addition, as shown in Fig. 14, also in terms of the CFU of fecal *Klebsiella* in each administration group at the time of day 28 in the experiment shown in Fig. 13, F18mix was able to reduce *Klebsiella* statistically significantly by more than any other group excluding F37mix.

From the above, it was suggested that all of the four groups shown in Fig. 10 were involved in the elimination of *Klebsiella*, and eliminated *Klebsiella* as a community.

Further, in the experiment shown in Fig. 13,

lymphocytes in the lamina propria of the large intestine were extracted from mice in the F37mix, F18mix, and F31-18mix groups and subjected to analysis by flow cytometry.

As a result, as shown in Fig. 15, it was clarified that the proportion of CD4+IFN γ + cells was high in the F13mix (F31-18mix) group, and that the induction of Th1 cells was suppressed in F37mix and F18mix.

(Example 9)

As shown in Example 8, in the experiment of removing the bacteria of each group from F18mix, F15mix obtained by removing the 3 strains of other Phyla had the lowest ability to eliminate Klebsiella. Therefore, paying attention to this group, the present inventors prepared groups each obtained by removing any one of these 3 strains (E. coli, Bifidobacterium, and Fusobacterium) from the 18 strains derived from feces F. Then, as shown in the upper part of Fig. 16, the Kp2H7 strains were administered to germ-free mice, and one week later, isolated bacteria mixed as described above, F18mix, or F15mix were administered.

As a result, as shown in the lower part (graph) of Fig. 16, by removing any of the above three strains, an increase of about 1 log in the bacterial amount of Klebsiella was observed, suggesting that all of the strains were involved in the elimination of Klebsiella.

(Example 10)

In order to explore the mechanism of eliminating Klebsiella of F37mix, the present inventors focused on whether host immunity was involved in this mechanism. Therefore, as shown in the upper part of Fig. 17, the
5 Klebsiella 2H7 strain was administered to a germ-free Rag2-
/-γc-/- mouse, MyD88-/-Triff-/- mouse, or wild-type mouse, and one week later, mixed F37mix was administered.

As a result, as shown in the lower part (graph) of Fig. 17, the same degree of elimination of the Klebsiella
10 2H7 strain by F37mix was observed in all types of mice. This suggests that the major innate immunity and acquired immunity of the host are not involved in the elimination of Klebsiella.

Next, the effect of eliminating the isolated
15 bacterial strains on pathogens and drug-resistant bacteria other than the Kp2H7 strain was evaluated. Note that in the analysis, the primers shown in Table 8 were used as primers specific to the bacteria.

[Table 8]

Target Bacteria and the Like	Primer Name	SEQ ID NO:
CRE	KPC-F684	212
	KPC-R860	213
VRE (vanA)	VanABF_JCM1998	214
	VanAR_JCM1998	215
ESBL	ESBL-F_AAC2001	216
	ESBL-R_AAC2001	217
LF82-specific pMT gene	LF82-F	218
	LF82-R	219
C.jejuni	C.jejuni_F	220
	C.jejuni_R	221
Universal	Universal_Fw_1387	222
	Universal_Rv_1492	223
C.diff	tpi-F	224
	tpi-R	225
C.upsaliensis	Cupsaliensis_2F	226
	Cupsaliensis_2R	227
C.upsaliensis	Cupsaliensis_3F	228
	Cupsaliensis_3R	229
Kp-P1	khe-Fw	230
	khe-RV	231
E.gallinarum	Eg_2F	232
	Eg_2R	233
P.mirabilis	P.mirabilis_NatMicro_F	234
	P.mirabilis_NatMicro_R	235

(Example 11)

First, as shown in the upper part of Fig. 18,

5 carbapenem-resistant Klebsiella (CRE) was administered to

germ-free mice, and one week later, mixed F37mix, K68mix, or I42mix was administered. The results obtained are shown at the lower part of Fig. 18.

Note that a bacterial solution of CRE was placed in an LB liquid medium and cultured overnight at 37°C to adjust the OD value to 1.2 (equivalent to 1×10^9 CFU/mL), and 200 μ L/individual (equivalent to 2×10^8 CFU/individual) of bacterial solution was administered into the stomach of mice using a sonde. Regarding the CFU count for CRE, DHL medium containing 30 mg/L ampicillin and 30 mg/L spectinomycin was used as a selective medium and cultured overnight at 37°C under aerobic conditions.

In addition, the mice one month after administration of the mixed isolated bacteria were dissected, and the large intestine was fixed with 4% PFA and embedded in paraffin to prepare sliced sections. These sections were stained with hematoxylin solution and eosin solution to observe inflammatory images of the tissues. Fig. 19 shows the obtained results.

As shown in the lower part (graph) of Fig. 18, F37mix and K68mix showed equivalent CRE elimination ability, and I42mix was slightly inferior to them. In addition, as shown in Fig. 19, no inflammatory findings such as ulceration or infiltration of inflammatory cells were observed in any of the mice. From this, it was clarified that the induction of inflammation in the large intestine

was suppressed by administering each of the isolated bacterial mixed groups.

(Example 12)

As shown in the upper part of Fig. 20, vancomycin-resistant *Enterococcus faecium* (VRE) was administered to germ-free mice, and one week later, mixed F37mix, K68mix, or I42mix was administered.

Note that a bacterial solution of VRE was placed in an LB liquid medium and cultured overnight at 37°C to adjust the OD value to 1.2 (equivalent to 1×10^9 CFU/mL), and 200 μ L/individual (equivalent to 2×10^8 CFU/individual) of bacterial solution was administered into the stomach of mice using a sonde. Regarding the CFU count for VRE, VRE medium (Nippon Becton) was used and cultured overnight at 37°C under aerobic conditions.

As a result, as shown in the lower part (graph) of Fig. 20, K68mix exhibited the highest elimination ability on VRE. In addition, as shown in Fig. 21, no inflammatory findings such as ulceration or infiltration of inflammatory cells were observed in any of the mice. From this, it was clarified that the induction of inflammation in the large intestine was suppressed by administering each of the isolated bacterial mixed groups.

(Example 13)

As shown in the upper part of Fig. 22, adhesion-invasive *E.coli* (AIEC LF82) was administered to germ-free

mice, and one week later, mixed F37mix, K68mix, and I42mix were administered.

Note that a bacterial solution of AIEC LF82 was placed in an LB liquid medium and cultured overnight at 37°C to adjust the OD to 1.2 (equivalent to 1×10^9 CFU/mL), and 200 μ L/individual (equivalent to 2×10^8 CFU/individual) of bacterial solution was administered into the stomach of mice using a sonde. Regarding the CFU count for AIEC LF82, MacConkey medium containing 1 mg/L cefotaxime was used as a selective medium and cultured overnight at 37°C under aerobic conditions to calculate the CFU.

As a result, as shown in the lower part (graph) of Fig. 22, F37mix had the highest elimination ability on AIEC LF82.

(Example 14)

As shown in the upper part of Fig. 23, ESBL-producing *Klebsiella* (Kp-ESBL) (ATCC 700721) was administered to germ-free mice, and one week later, mixed F37mix, K68mix, I42mix, or feces F was administered.

Note that a bacterial solution of Kp-ESBL was placed in an LB liquid medium and cultured overnight at 37°C to adjust the OD to 1.2 (equivalent to 1×10^9 CFU/mL), and 200 μ L/individual (equivalent to 2×10^8 CFU/individual) of bacterial solution was administered into the stomach of mice using a sonde. Regarding the CFU count for Kp-ESBL,

DHL medium containing 30 mg/L ampicillin and 30 mg/L spectinomycin was used as a selective medium and cultured overnight at 37°C under aerobic conditions for calculation.

As a result, as shown in the lower part (graph) of Fig. 23, F37mix and K68mix showed the same Kp-ESBL elimination ability as feces F.

(Example 15)

As shown in the upper part of Figs. 24 and 25, *Campylobacter jejuni* 81-176 (ATCC BAA2151) was administered to germ-free mice, and one week later, mixed F37mix, K68mix, I42mix, or feces F was administered.

The bacterial solution of *Campylobacter jejuni* was placed in a TS liquid medium, placed in an anaerobic jar together with a slightly aerobic AnaeroPack, and cultured at 42°C for 48 hours, and the bacterial solution was administered into the stomachs of the mice using a sonde.

For *Campylobacter jejuni*, CFU and qPCR were used to show the bacterial amount.

CHROMagar *Campylobacter* was used for CFU counting, which was placed in an anaerobic jar together with a slightly aerobic AnaeroPack and cultured at 42°C for 48 hours. Fig. 24 shows the obtained results.

The qPCR measurement was performed according to the following procedure.

LightCycler (registered trademark) 480II (Roche; 05015243001) and Thunderbird (registered trademark) SYBR

(registered trademark) qPCR Mix (TOYOBO; QPS-201X5) were used to amplify and quantify with *Campylobacter jejuni* genome-specific primers and universal bacterial primers, and the calculated DNA concentration ratio was used as the abundance ratio of *Campylobacter jejuni*. Fig. 25 shows the obtained results.

Note that the primer sets set forth in SEQ ID NOs: 220 and 221 were used as *Campylobacter jejuni* genome-specific primers for qPCR, and the primer sets set forth in SEQ ID NOs: 222 and 223 were used as universal bacterial primers.

In addition, the bacterial genome was extracted by the following steps.

A PBS solution containing EDTA and glycerol (final concentration of EDTA: 10 mM, and final concentration of glycerol: 20% by volume) was added 5 times by weight to 10 mg of mouse feces, and the mixture was crushed and suspended with vigorous shaking and stirring. Added to 100 μ L of sample solution was 800 μ L of 10 mM Tris/10 mM EDTA buffer solution (pH 8.0, also referred to as "TE10" hereinafter) having 15 mg of lysozyme (manufactured by Sigma-Aldrich, Lysozyme from chicken egg white; L4919) and 5 μ L of RNase (manufactured by Thermo Fisher Scientific, PureLink RNase A (20 mg/mL); 12091-021) dissolved therein, and the mixture was shaken at 37°C for 1 hour. Subsequently, 2,000 U of Achromopeptidase (registered

trademark) (Wako; 015-09951) was added, and the mixture was shaken at 37°C for 30 minutes for lysis. Then, 50 µL of a 20% SDS TE10 solution and 50 µL of a TE10 solution having Proteinase K (Roche, Proteinase K, recombinant, PCR Grade; 0311585.101) at a final concentration of 20 mg/ml dissolved therein were added, and the mixture was shaken at 55°C for 60 minutes. Next, DNA was obtained from 400 µL of the solution using Maxwell (registered trademark) RSC Cultured Cells DNA Kit (Promega).

As shown in Figs. 24 and 25, for *Campylobacter jejuni*, all the mixed bacteria had a good bacterial elimination ability equivalent to that of feces F.

(Example 16)

As shown in the upper part of Fig. 26, *Clostridium difficile* (St. 630) was administered to germ-free mice, and one week later, mixed F37mix, K68mix, I42mix, K47mix, or feces F was administered. Note that K47mix denotes 47 strains isolated from the fecal sample derived from #K, and is duplicated with the 68 bacterial strains (K1 to K46 shown in Tables 1 and 2) except for 1 bacterial strain.

The bacterial solution of *C. difficile* was subjected to spore formation, adjusted to around 1×10^5 cells, and administered into the stomachs of the mice using a sonde. The spore formation was performed by culturing in Clospore medium for 8 days in an anaerobic chamber at 37°C and washing the medium with PBS, followed by treatment with

sonication, addition of Lysoizyme and trypsin, and treatment at 45°C for 6 hours and then at 70°C for 10 minutes.

For *C. difficle*, qPCR was used to quantify the bacterial amount. As the primers for qPCR, the primer set shown in SEQ ID NOs: 224 and 225 was used.

As a result, as shown in the lower part of Fig. 26, a high elimination ability was observed in K68mix and K47mix for *C. difficile*.

[Industrial Applicability]

As described above, by suppressing the colonization of drug-resistant bacteria and pro-inflammatory bacteria in the intestinal tract, the present invention makes it possible to treat, ameliorate, or prevent diseases caused by these bacteria. Therefore, the present invention is extremely useful in the development, treatment, amelioration, prevention, and the like of pharmaceutical products relating to infectious diseases caused by drug-resistant bacteria or pro-inflammatory bacteria.

5 Throughout this specification and the claims which follow,
unless the context requires otherwise, the word "comprise",
or variations such as "comprises" or "comprising", will be
understood to imply the inclusion of a stated integer or
group of integers or steps but not the exclusion of any other
10 integer or group of integers or steps.

The reference to any prior art in this specification is not,
and should not be taken as, an acknowledgement or any form of
suggestion that the prior art forms part of the common
15 general knowledge in Australia.

THE CLAIMS DEFINING THE INVENTION ARE AS FOLLOWS:

[Claim 1]

An antibacterial composition against drug-resistant bacteria or pro-inflammatory bacteria, wherein

5 said composition has active ingredients, and

the active ingredients are:

(1) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 69 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 69;

(2) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 80 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 80;

(3) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 85 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 85;

(4) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 86 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 86;

(5) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 87 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 87;

(6) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 88 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 88;

(7) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 89 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 89;

(8) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 90 or a nucleotide sequence sharing

at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 90;

(9) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 91 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 91;

(10) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 92 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 92;

(11) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 94 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 94;

(12) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 96 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 96;

(13) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 98 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 98;

(14) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 99 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 99;

(15) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 100 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 100;

(16) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 101 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 101;

(17) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 103 or a nucleotide sequence sharing

at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 103; and,

(18) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 105 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 105.

[Claim 2]

The antibacterial composition according to claim 1, which is a pharmaceutical composition.

[Claim 3]

The antibacterial composition according to claim 1, which is a pharmaceutical composition for treating, ameliorating, or preventing an infectious disease or an inflammatory disease.

[Claim 4]

A method of treating, ameliorating, or preventing a disease caused by drug-resistant bacteria or pro-inflammatory bacteria, wherein the method comprises a step of administering an effective amount of the antibacterial composition according to claim 1 to a subject.

[Claim 5]

Use of active ingredients in a manufacture of a medicament for treating, ameliorating, or preventing a disease caused by drug-resistant bacteria or pro-inflammatory bacteria, wherein the active ingredients are:

(1) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 69 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 69;

(2) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 80 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of

sequence of SEQ ID NO: 80;

(3) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 85 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 85;

(4) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 86 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 86;

(5) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 87 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 87;

(6) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 88 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 88;

(7) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 89 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 89;

(8) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 90 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 90;

(9) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 91 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 91;

(10) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 92 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 92;

(11) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 94 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of

sequence of SEQ ID NO: 94;

(12) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 96 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 96;

(13) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 98 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 98;

(14) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 99 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 99;

(15) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 100 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 100;

(16) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 101 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 101;

(17) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 103 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 103; and,

(18) a bacterium having a DNA composed of the nucleotide sequence of SEQ ID NO: 105 or a nucleotide sequence sharing at least 90% identity with the nucleotide sequence of sequence of SEQ ID NO: 105.

Fig. 1

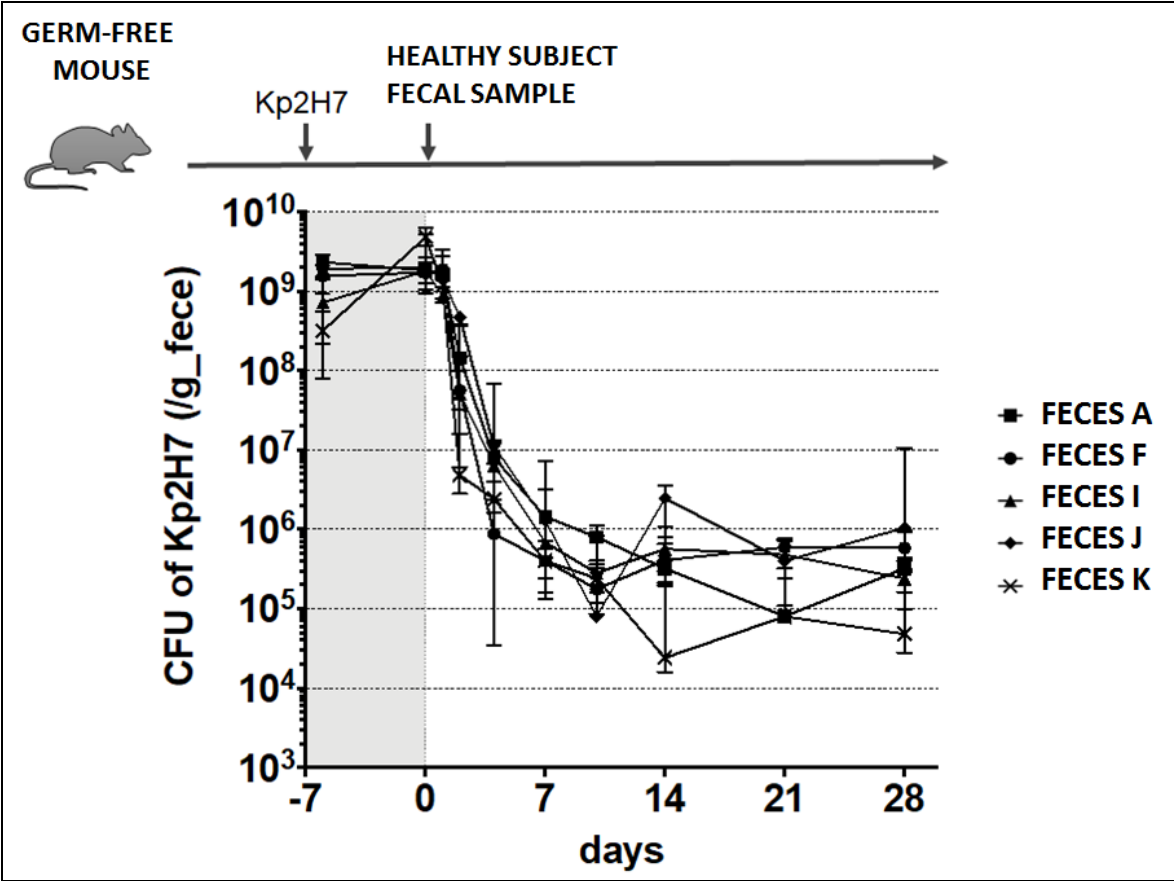


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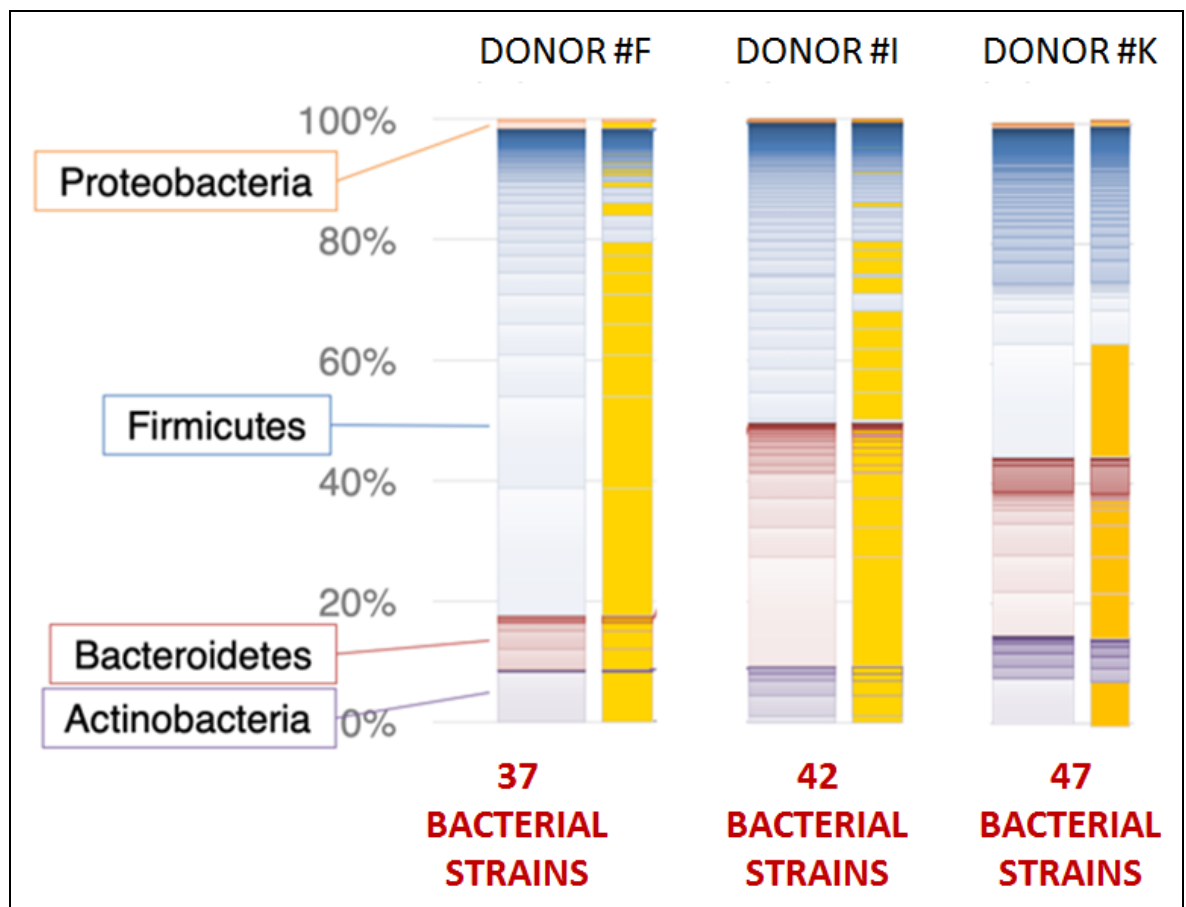


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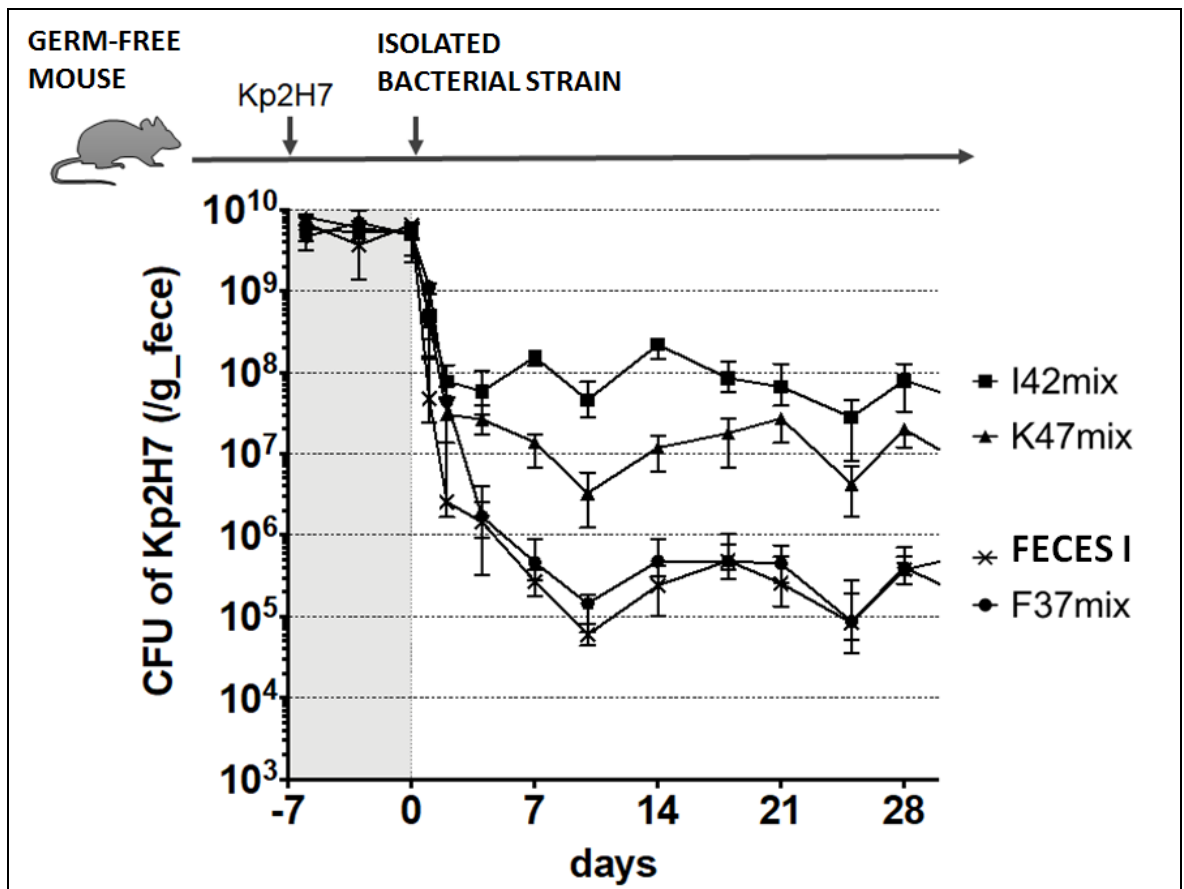


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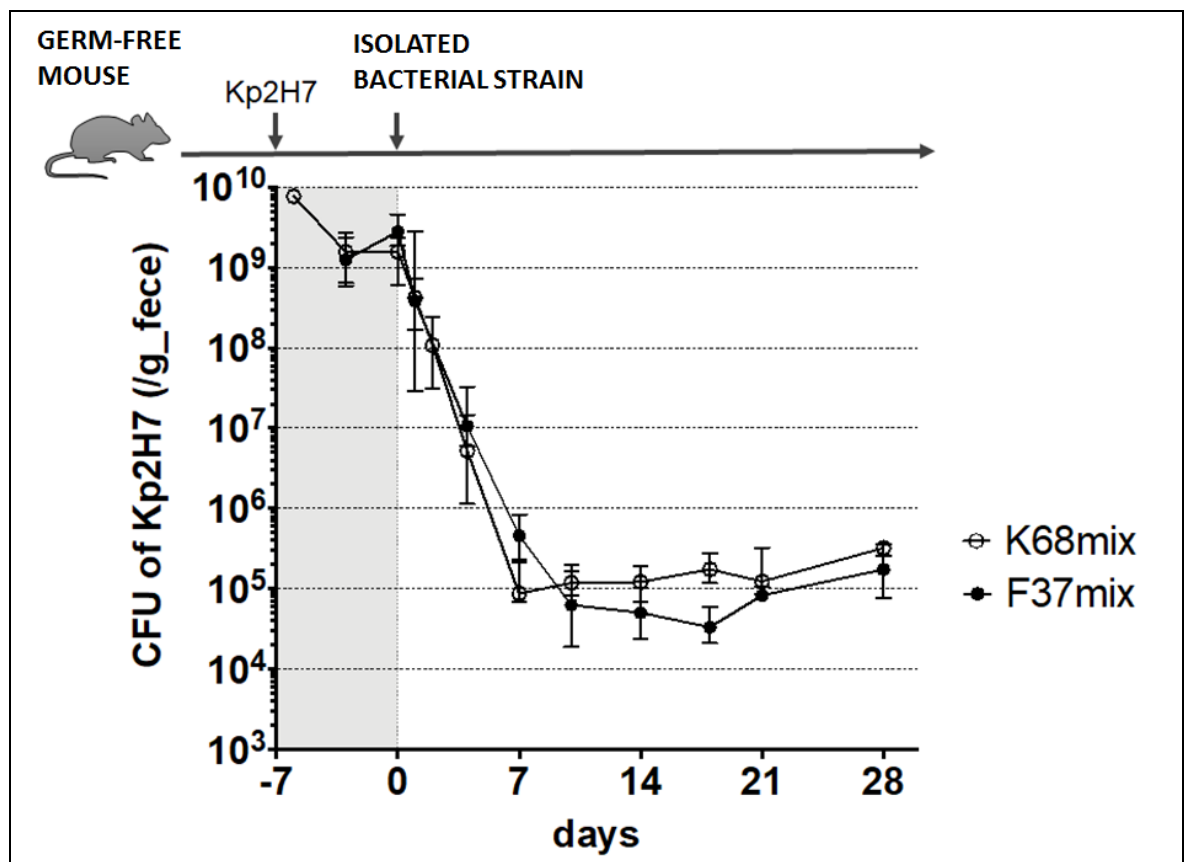


Fig. 5

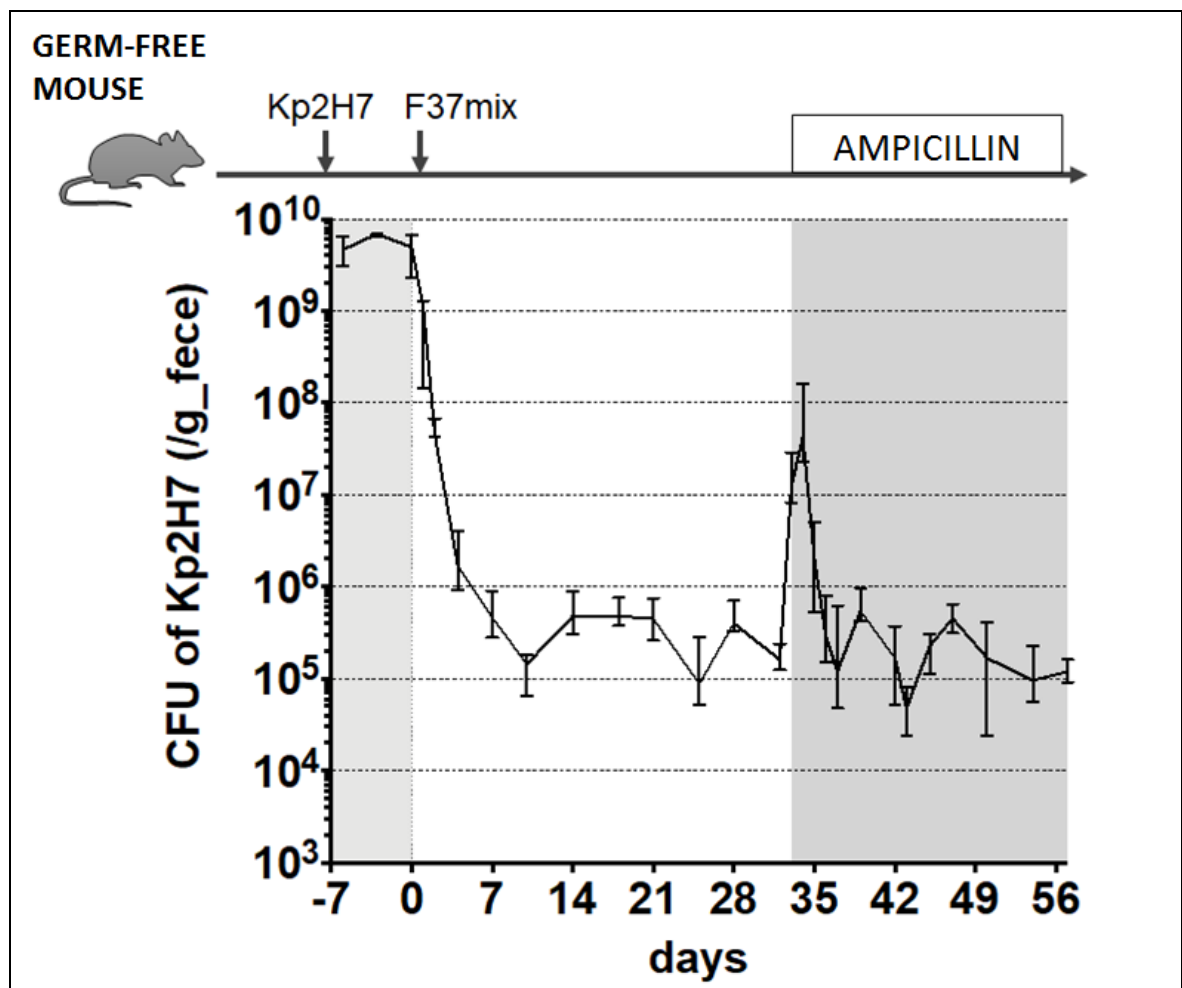


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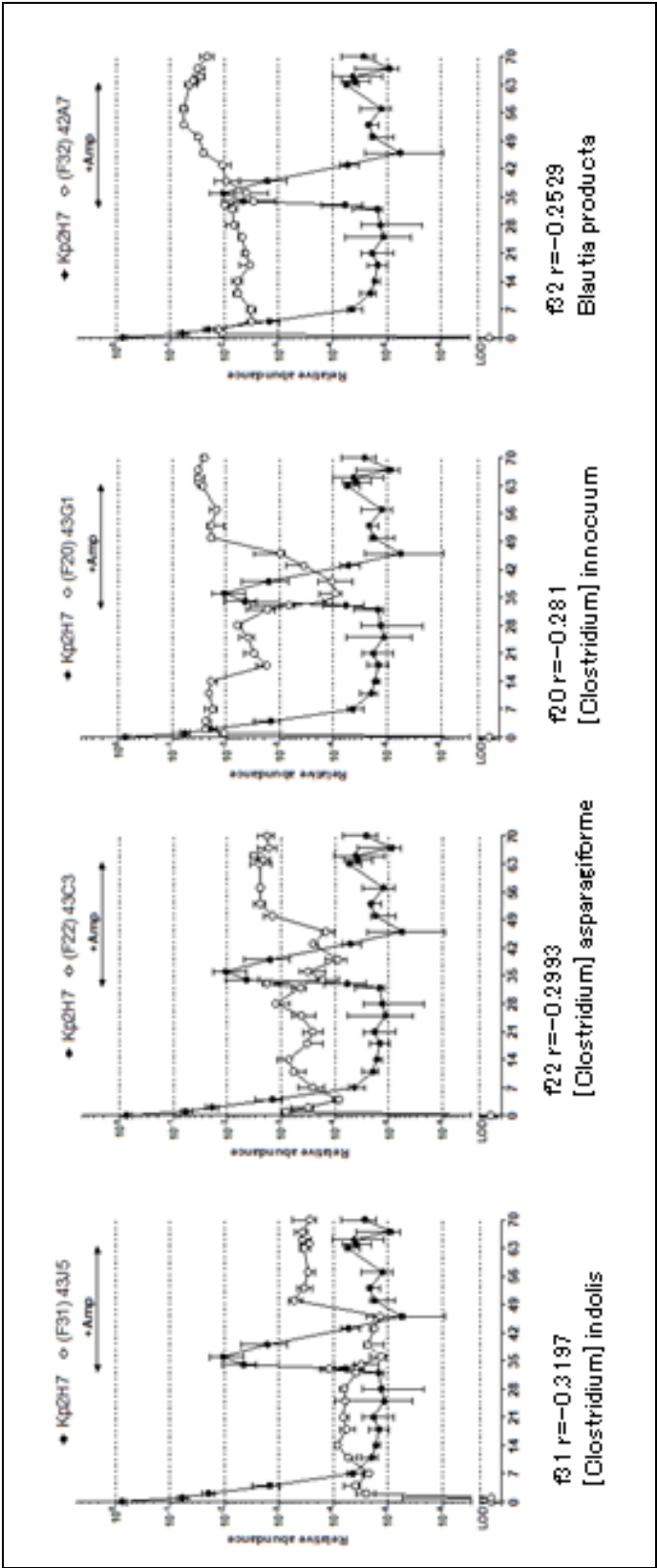


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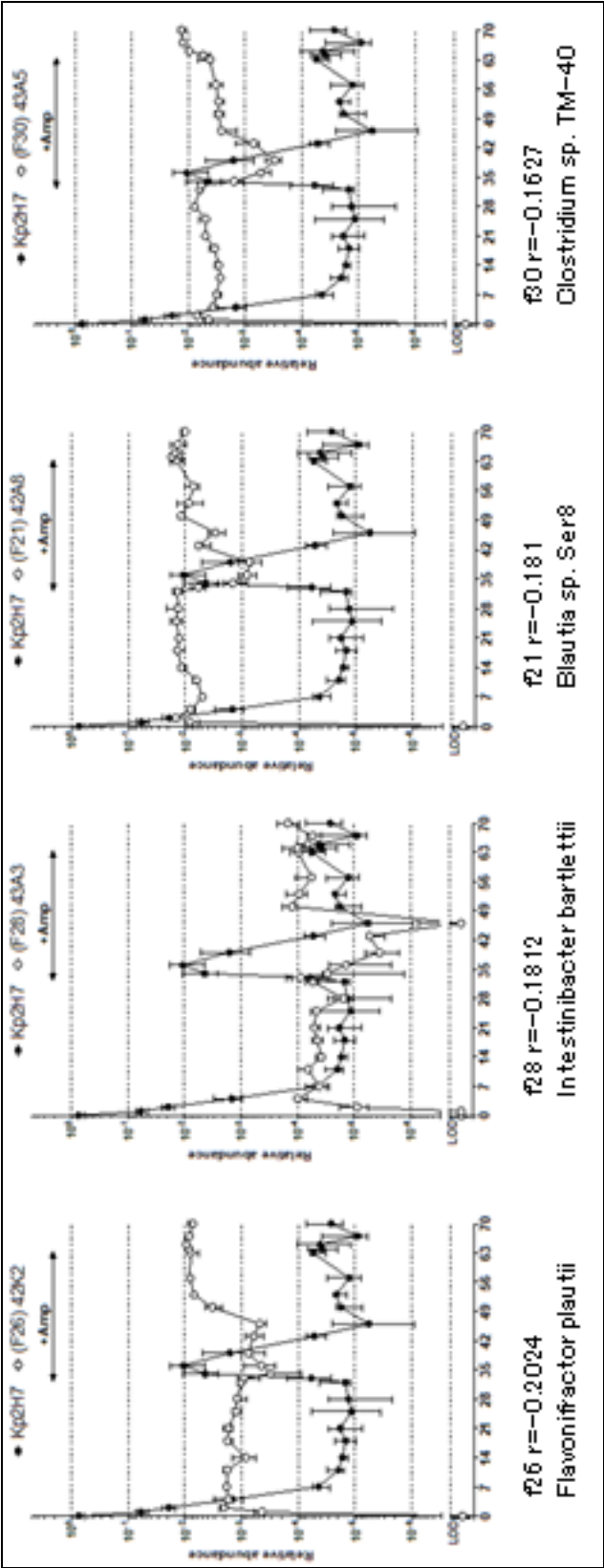


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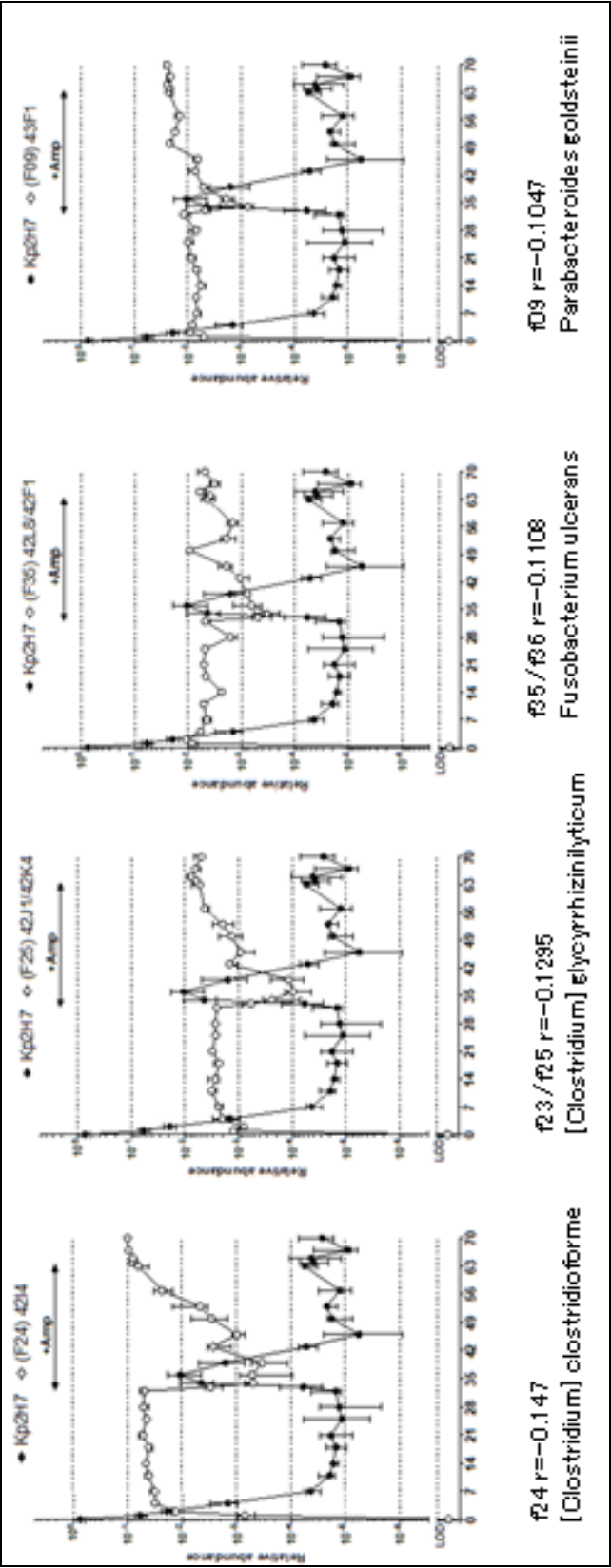


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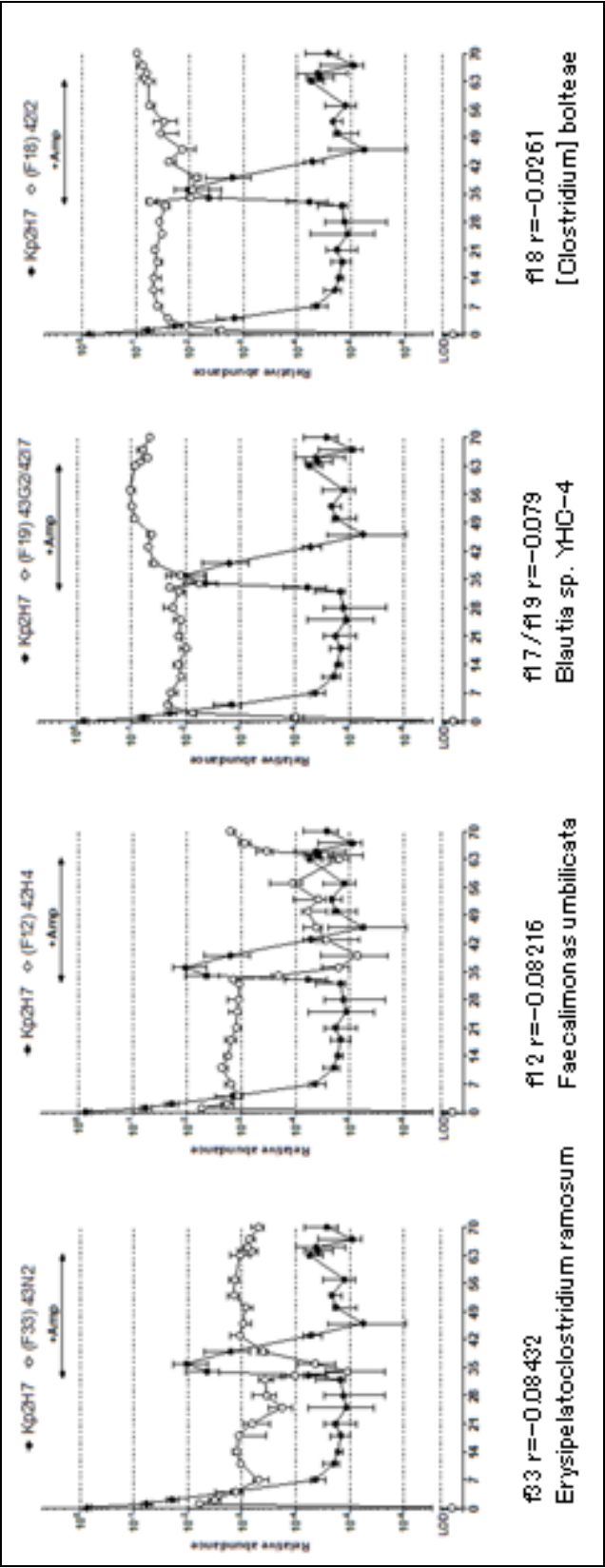


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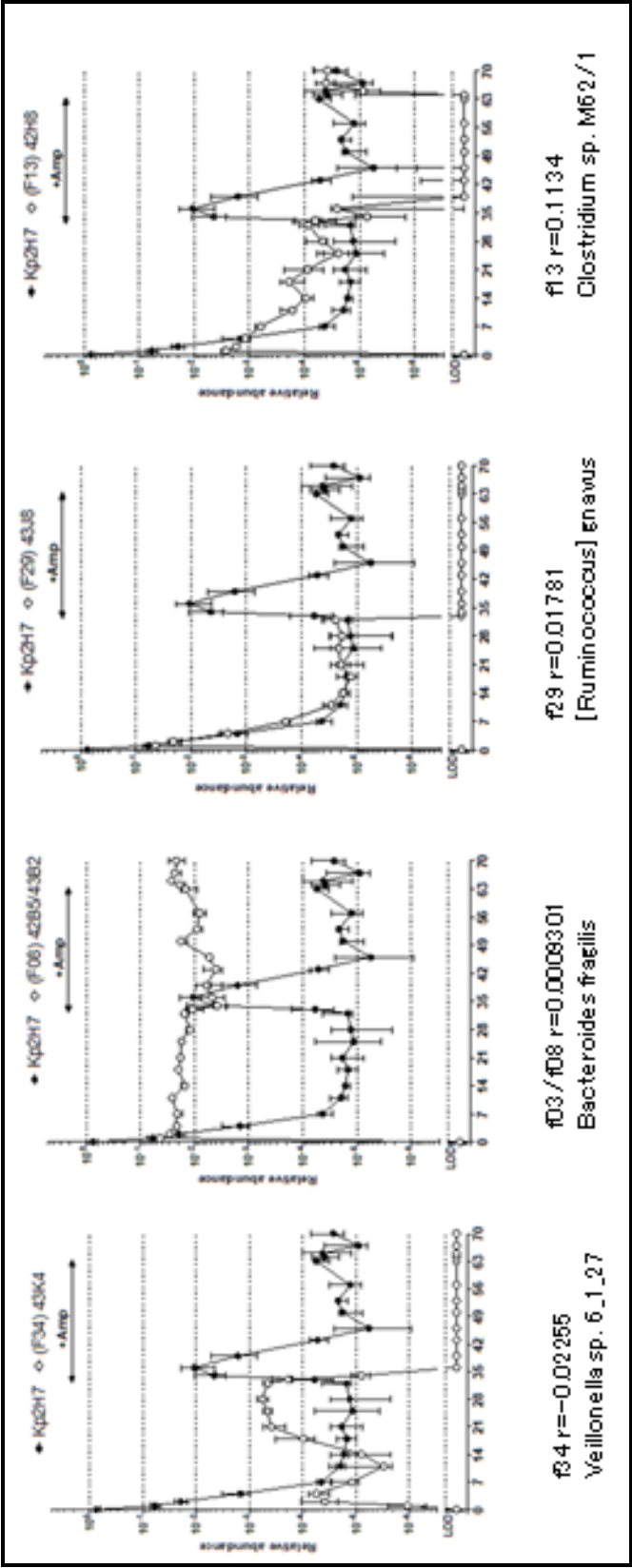


Fig. 6F

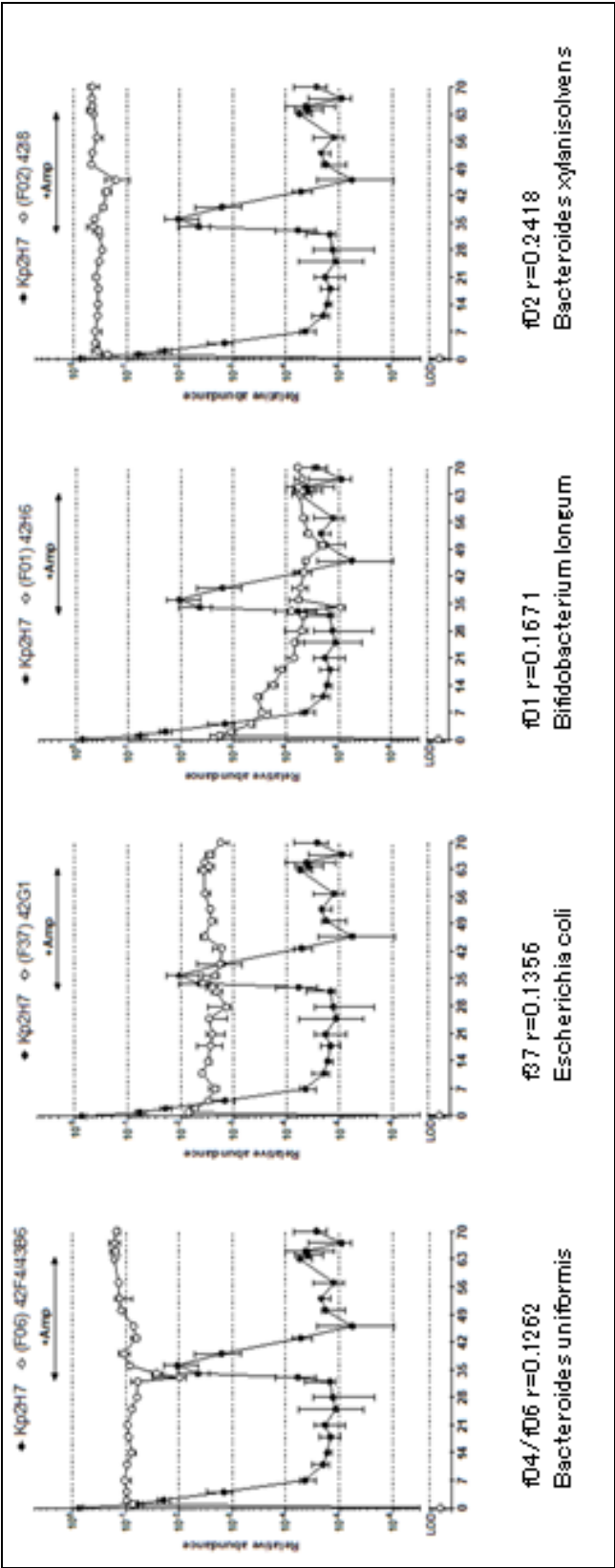


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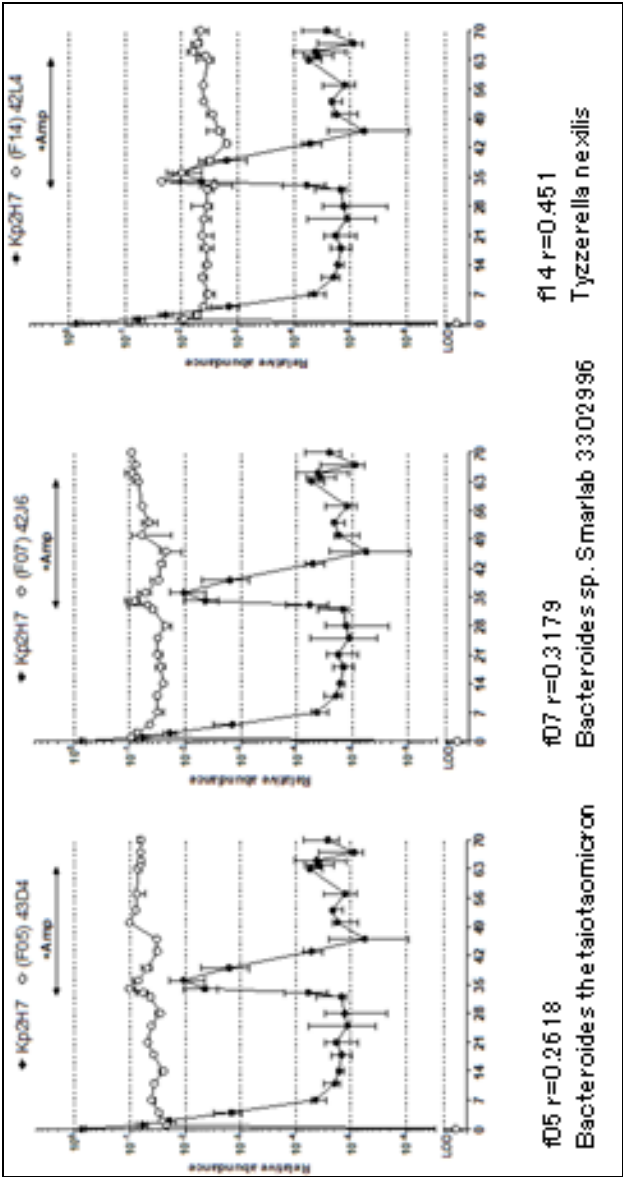


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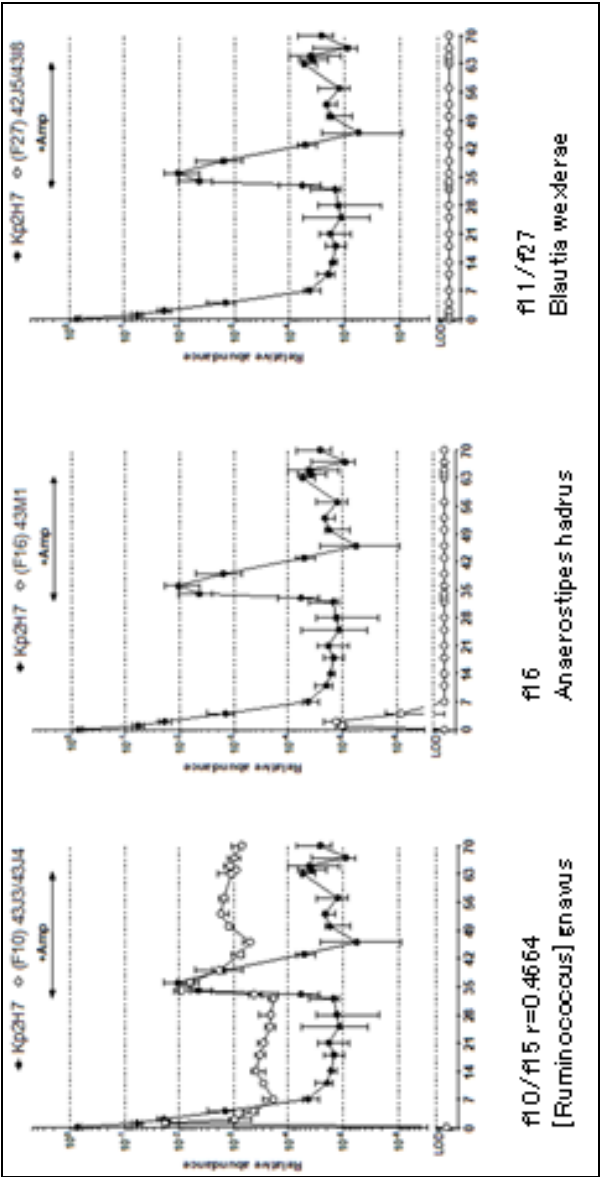


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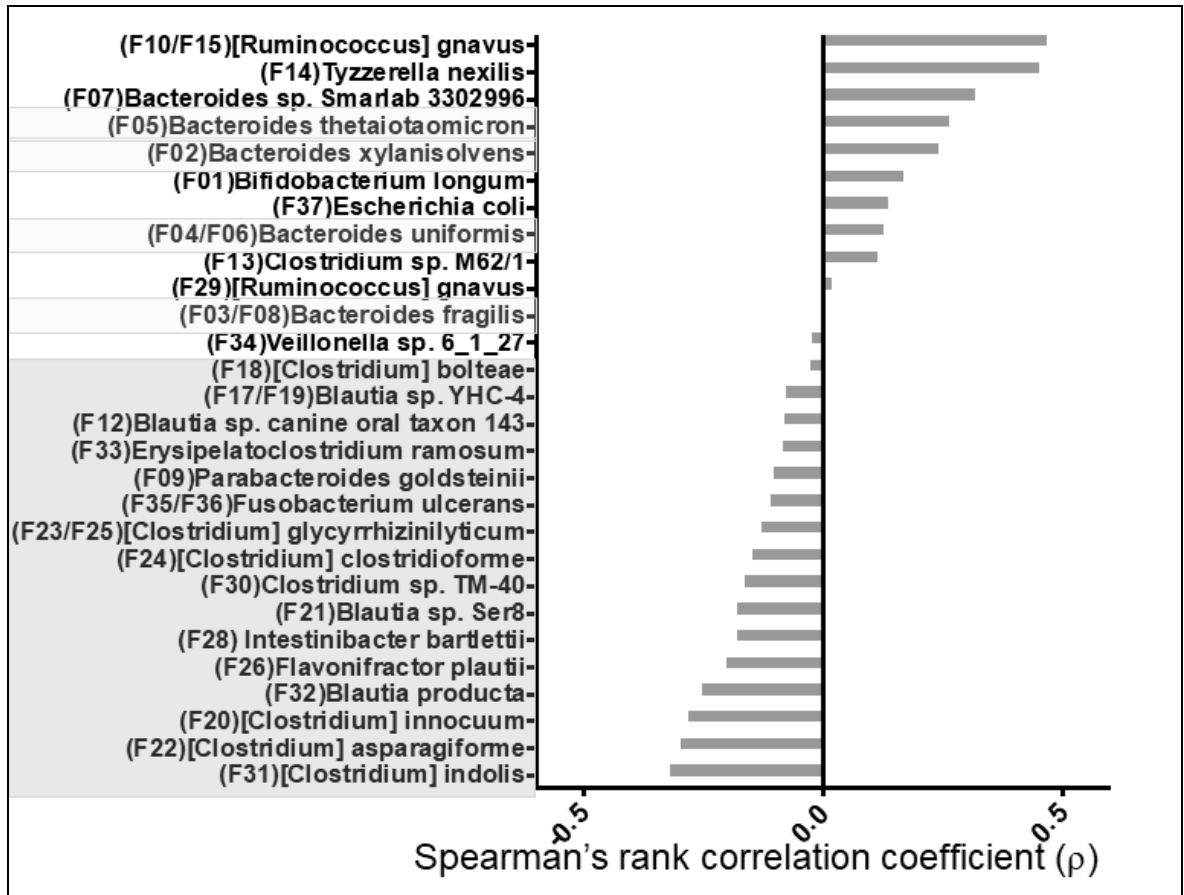


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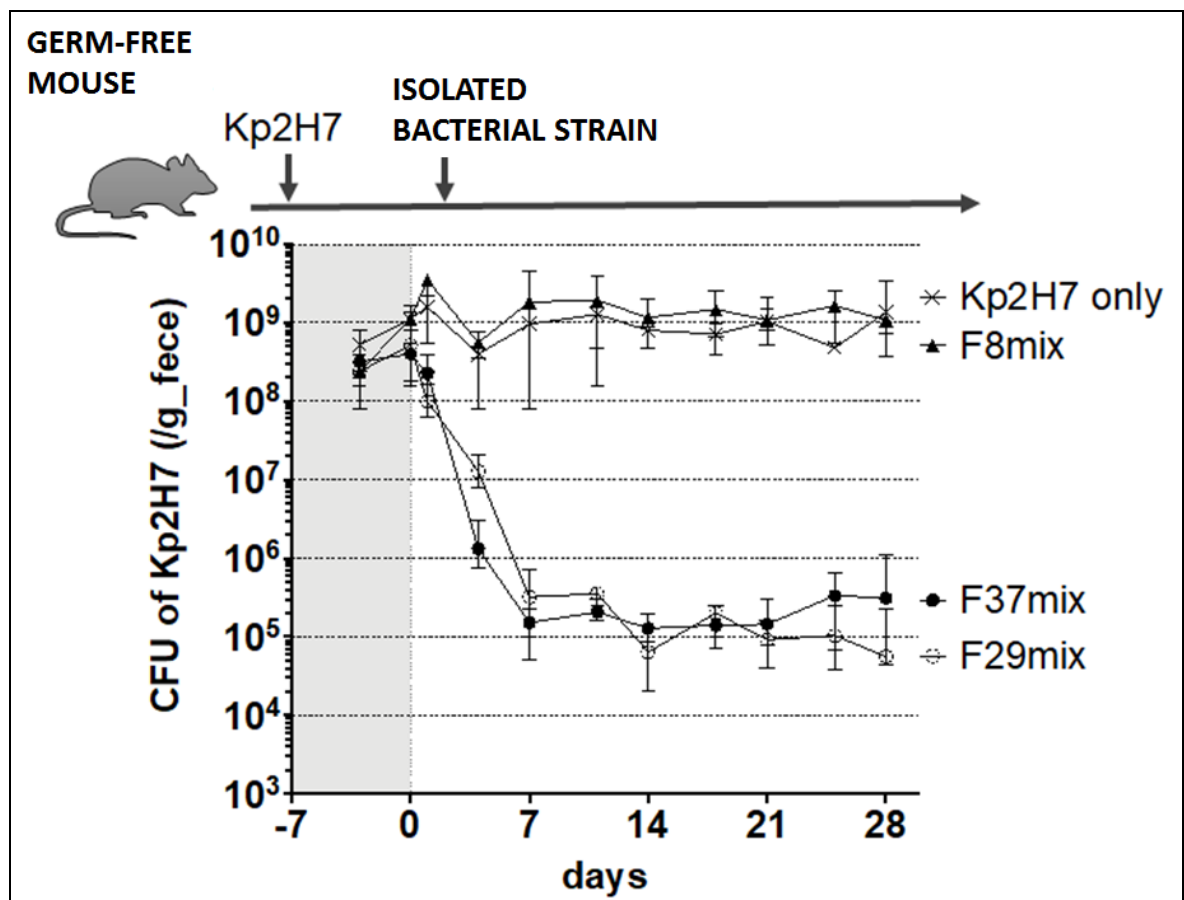


Fig. 9

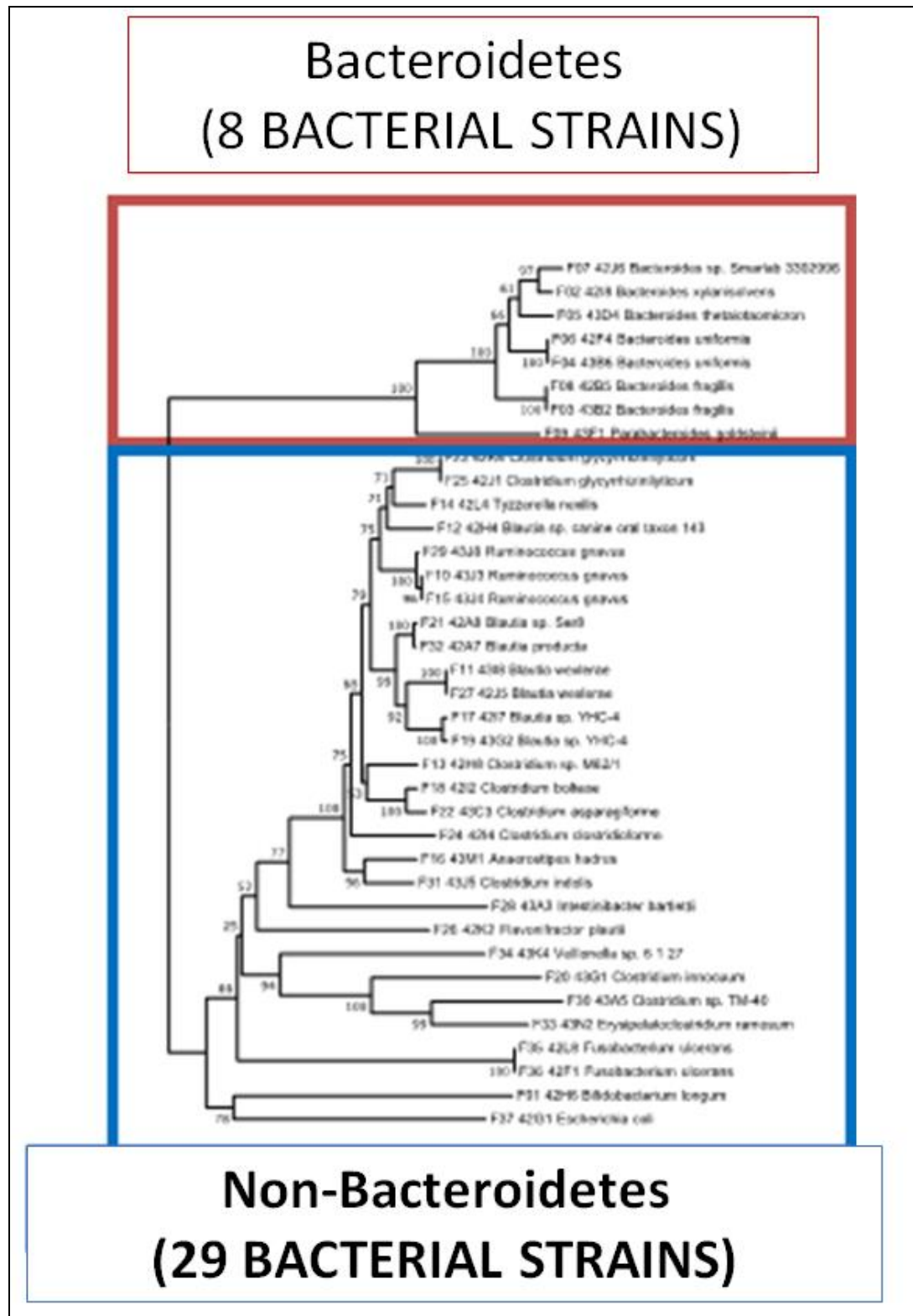


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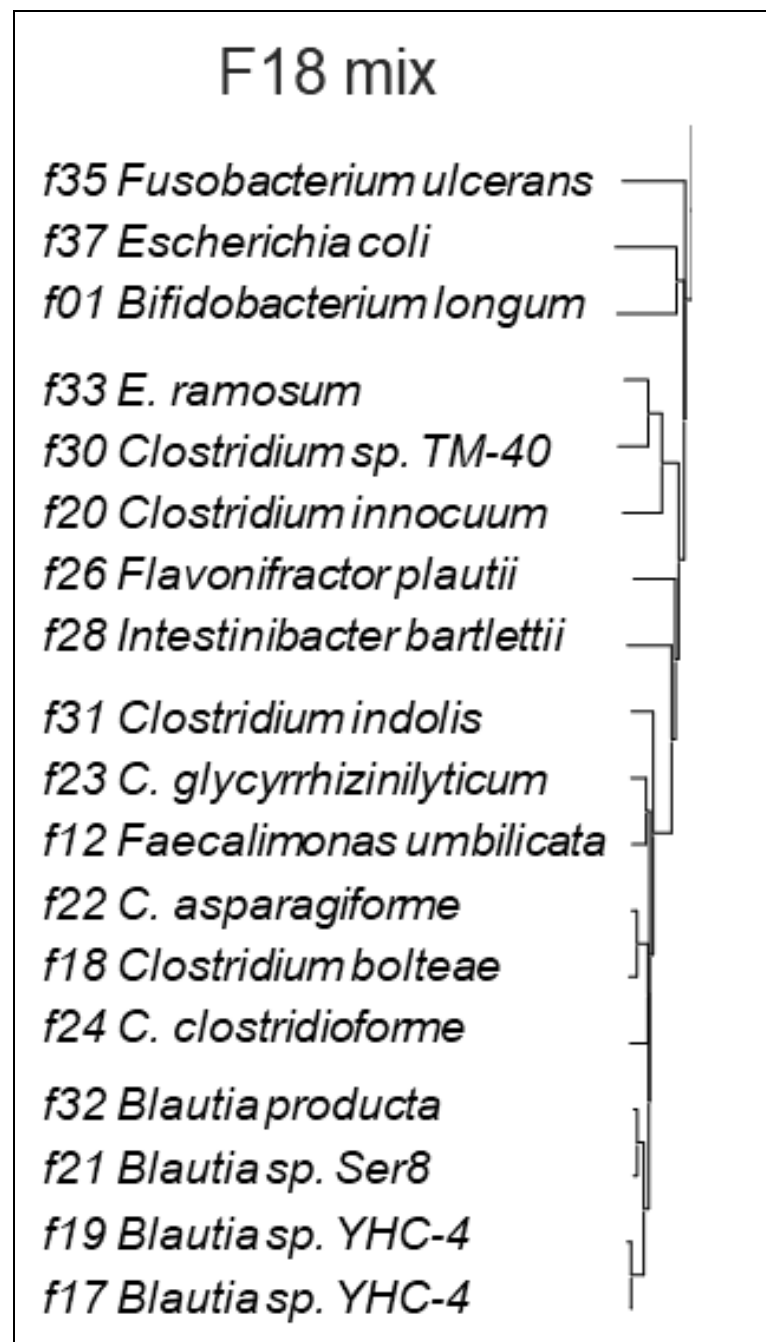


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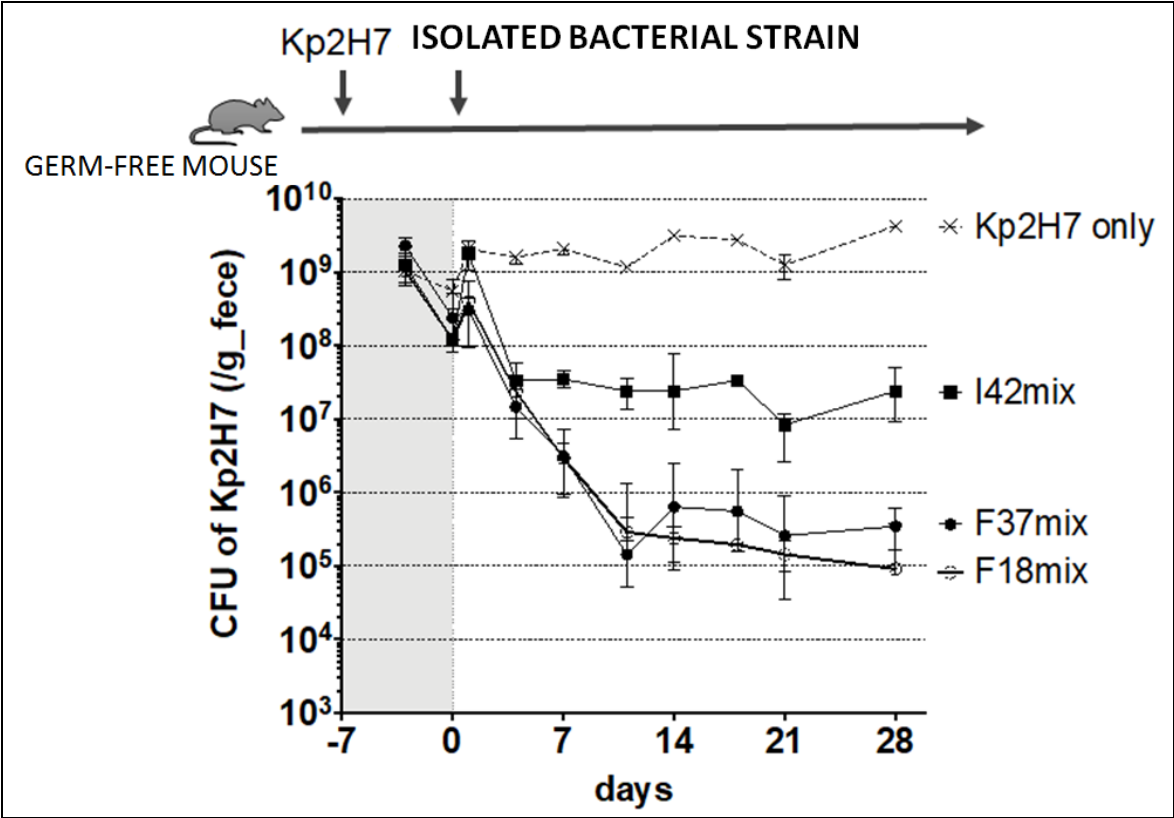


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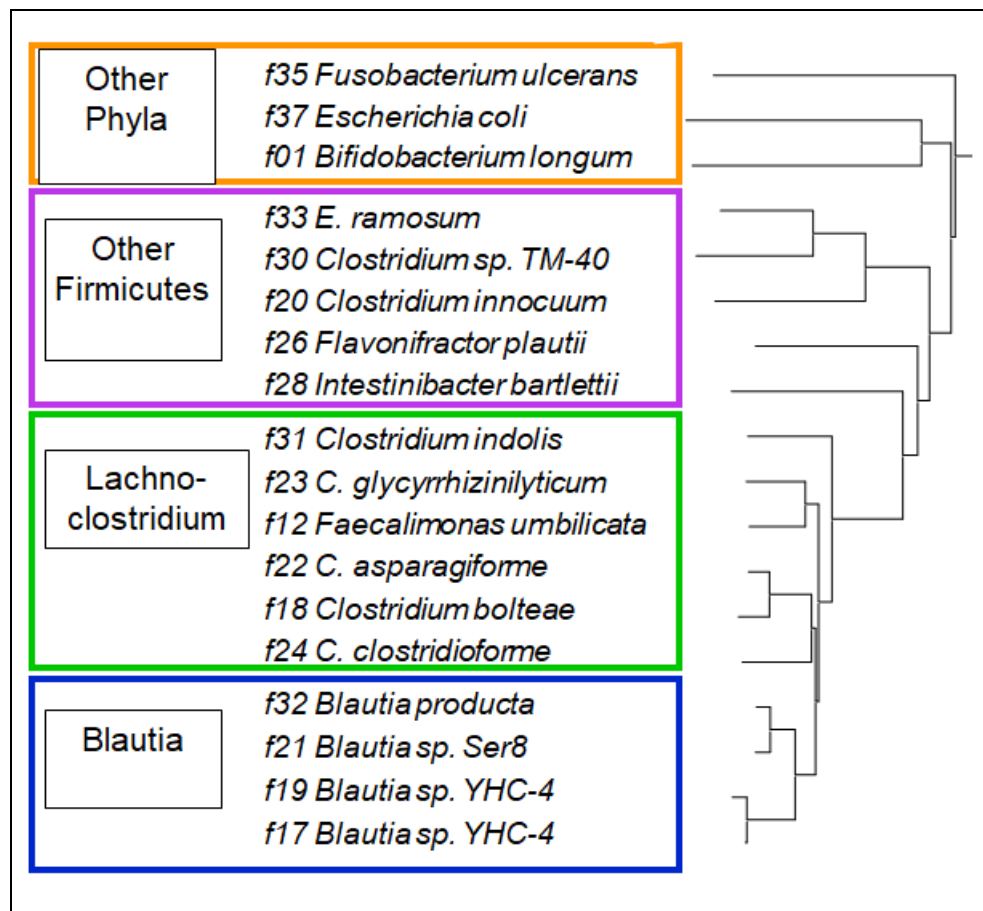


Fig. 13

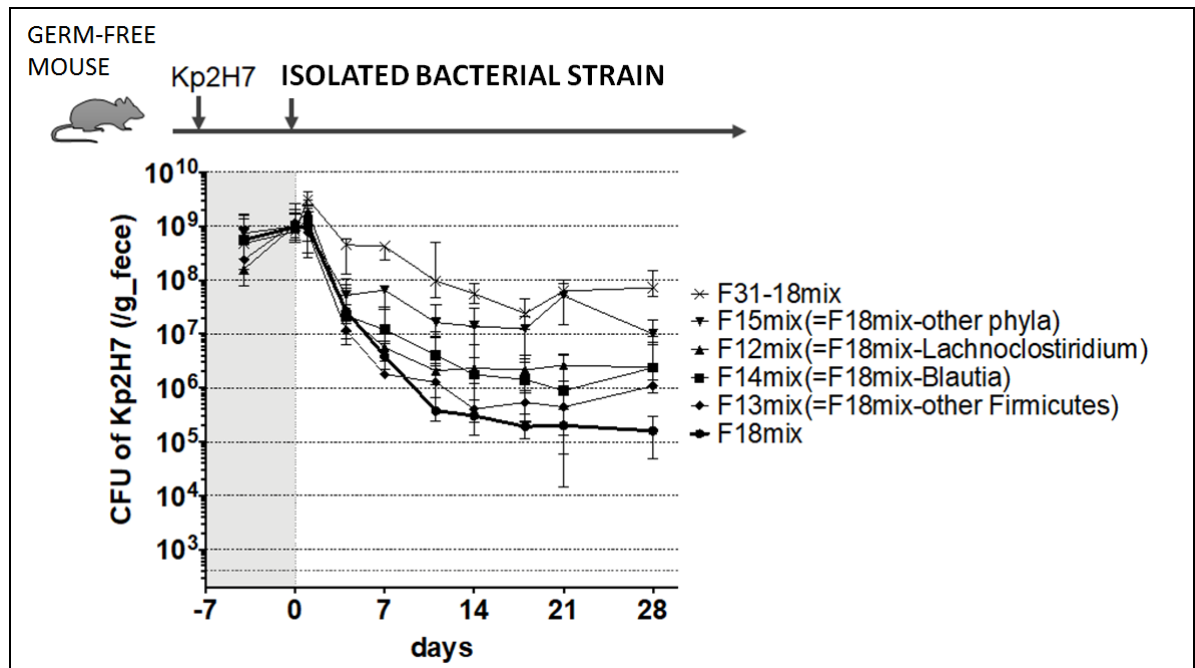


Fig. 14

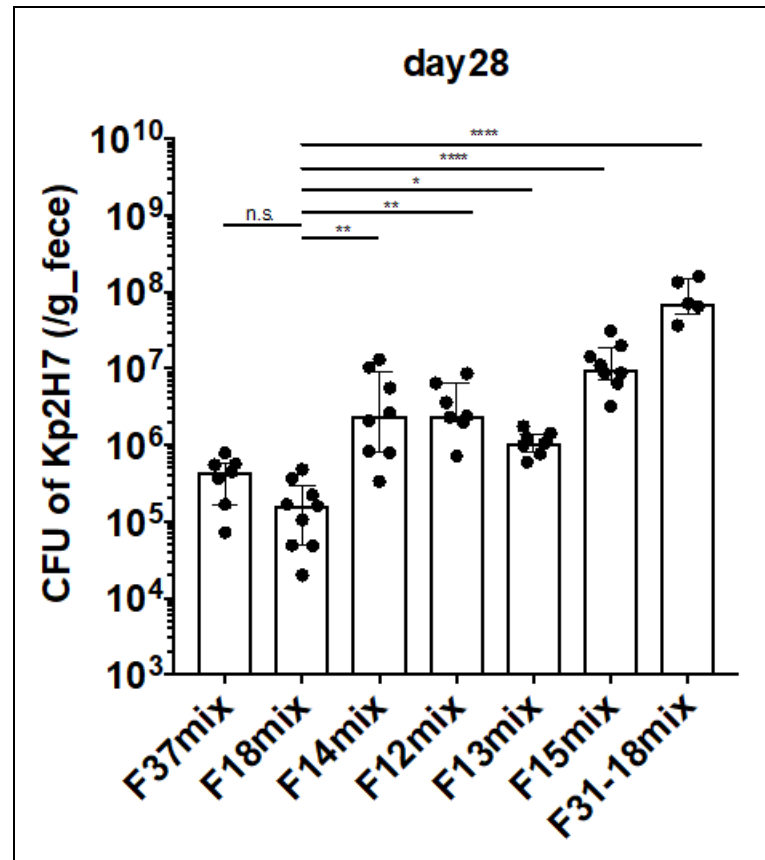


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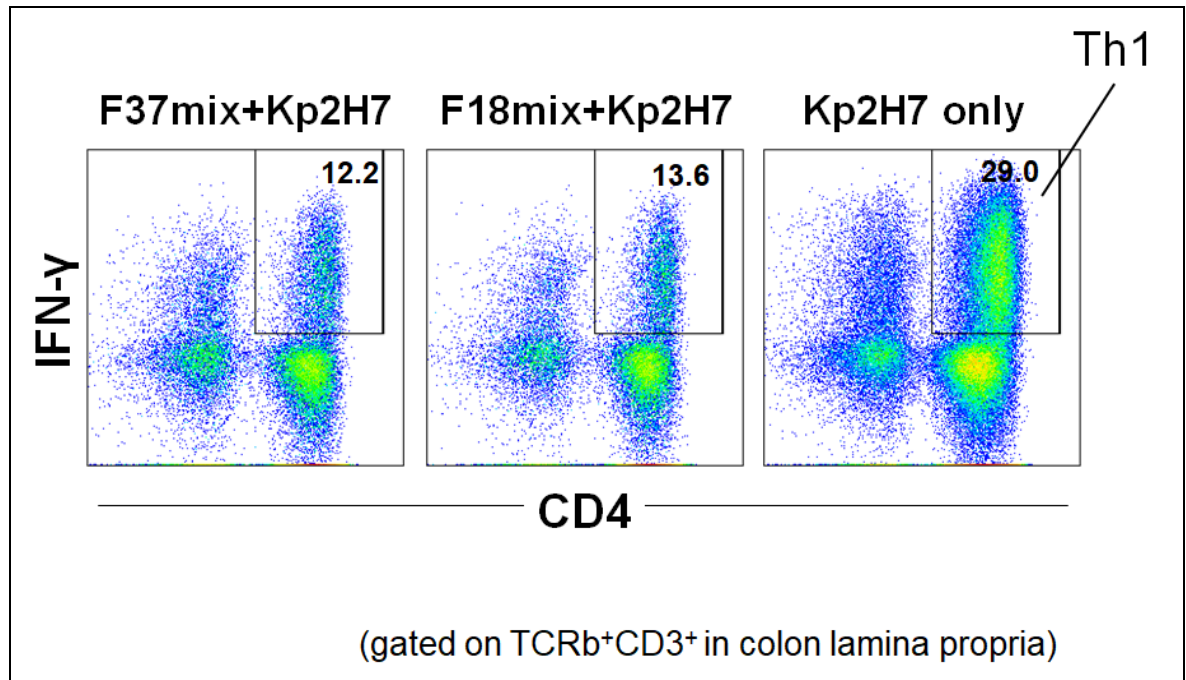


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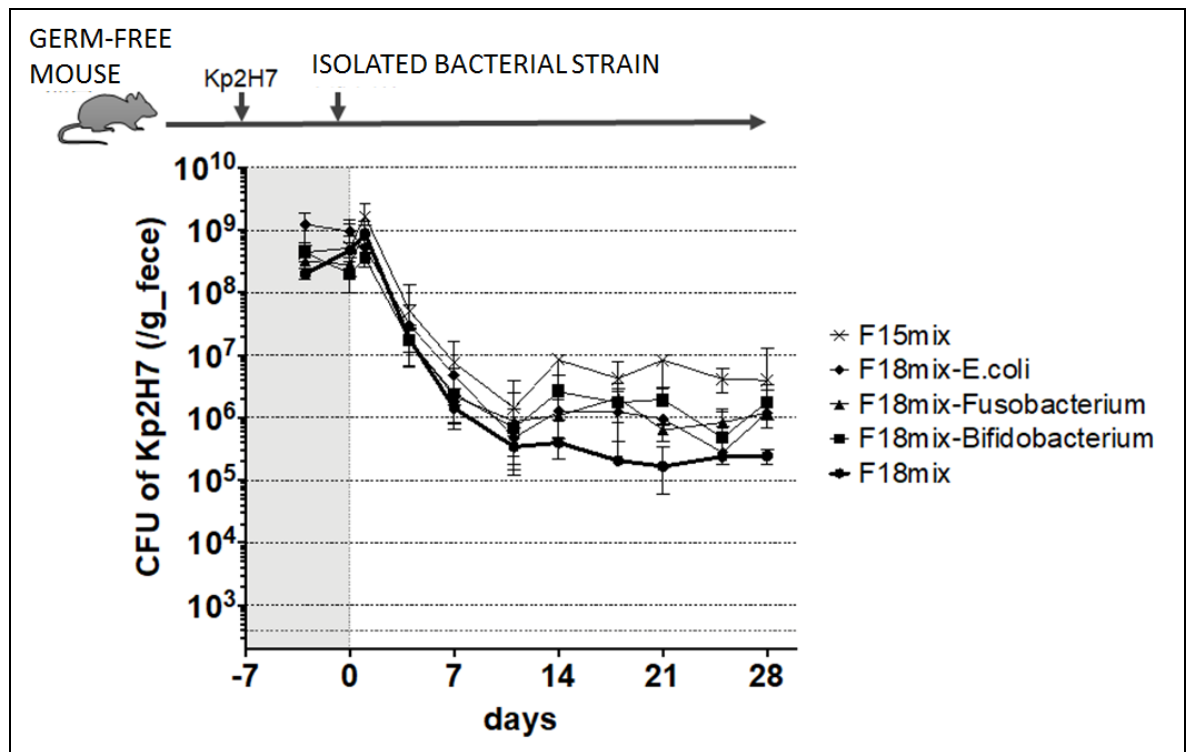


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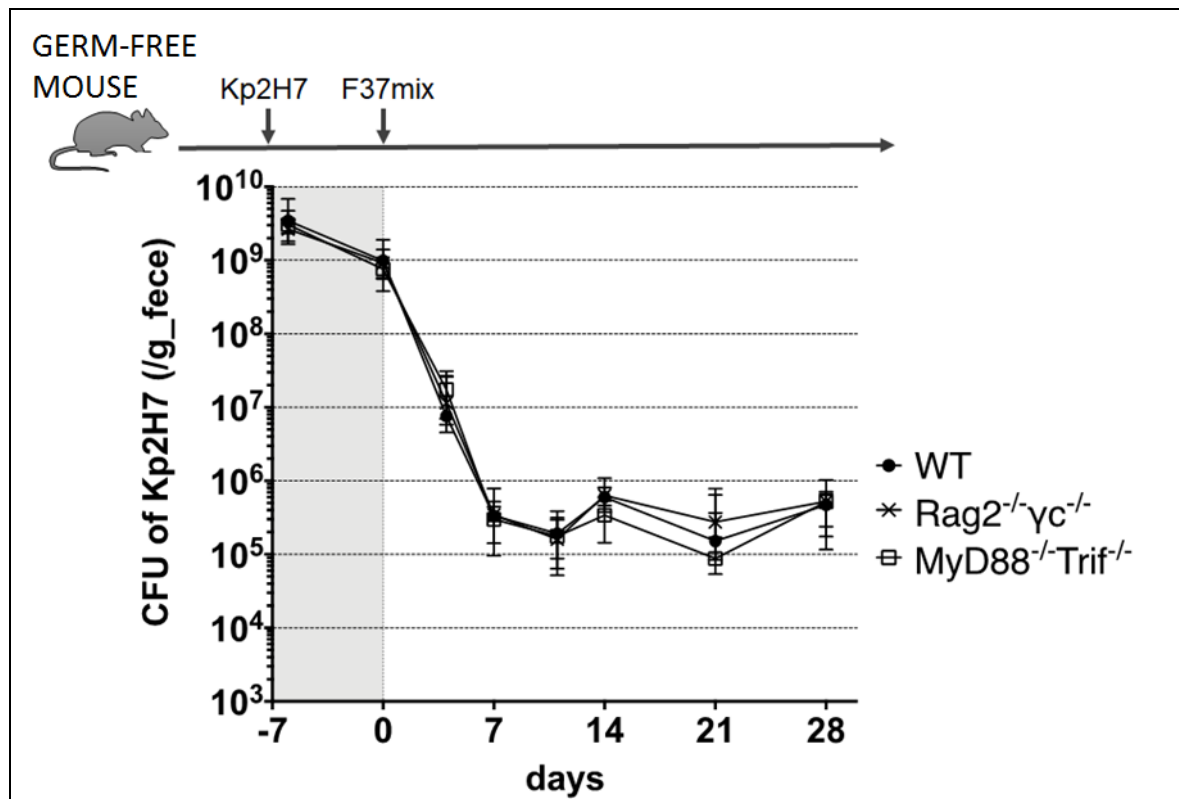


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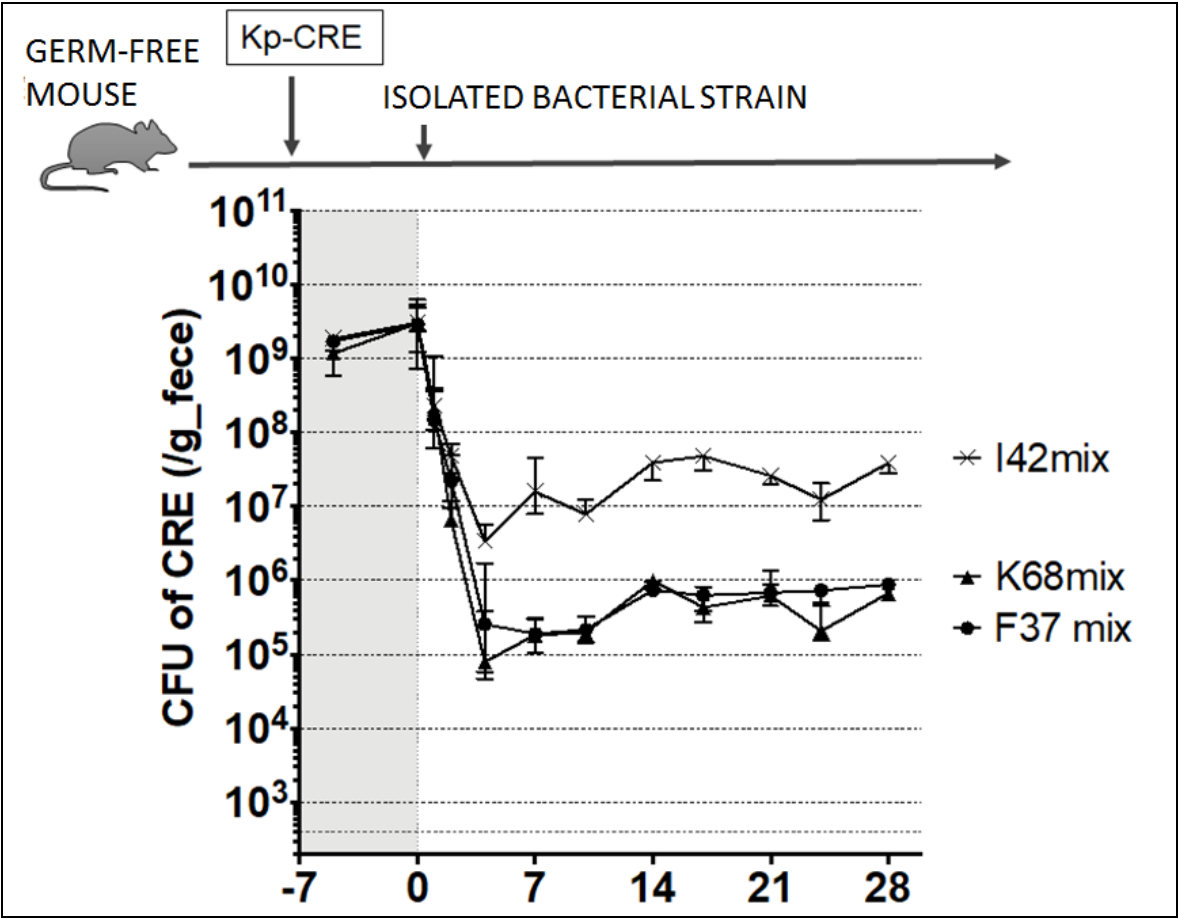


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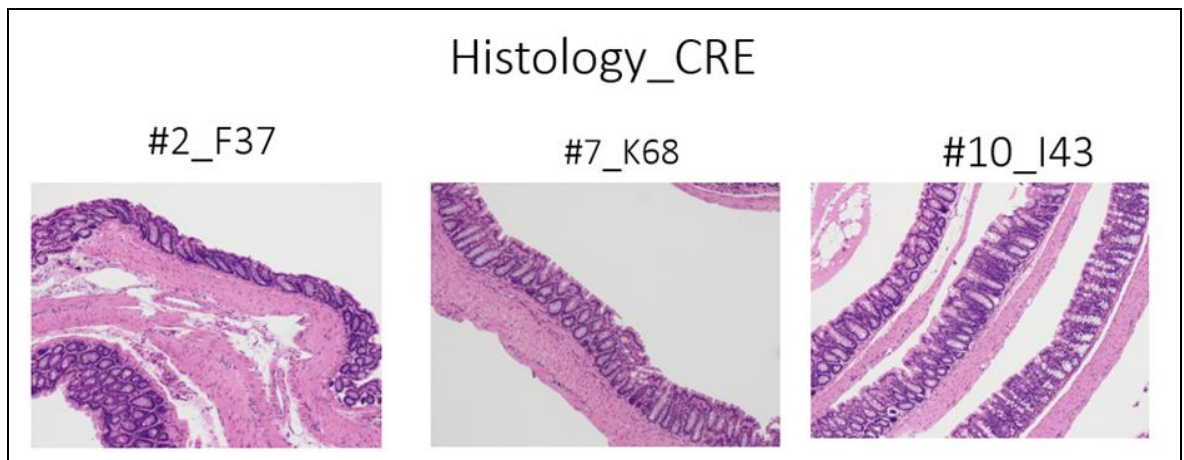


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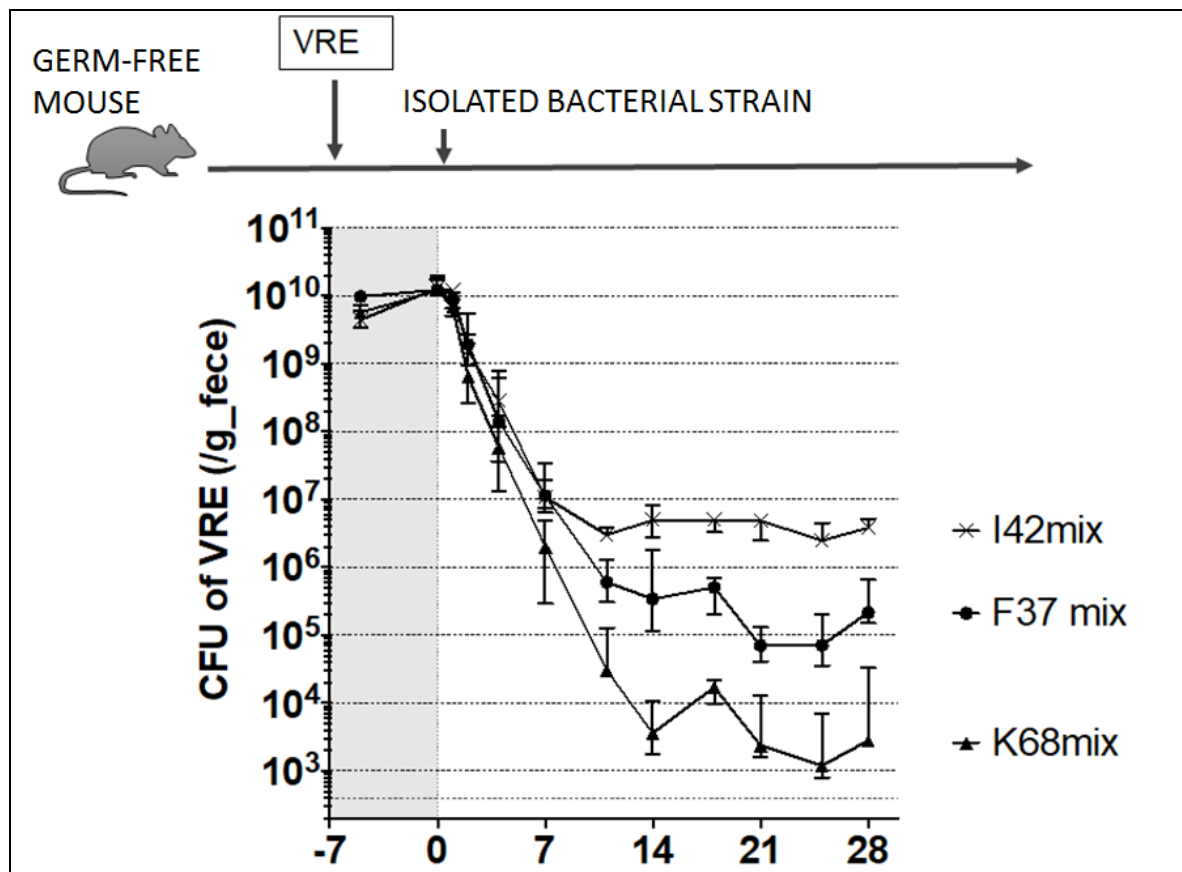


Fig. 21



Fig. 22

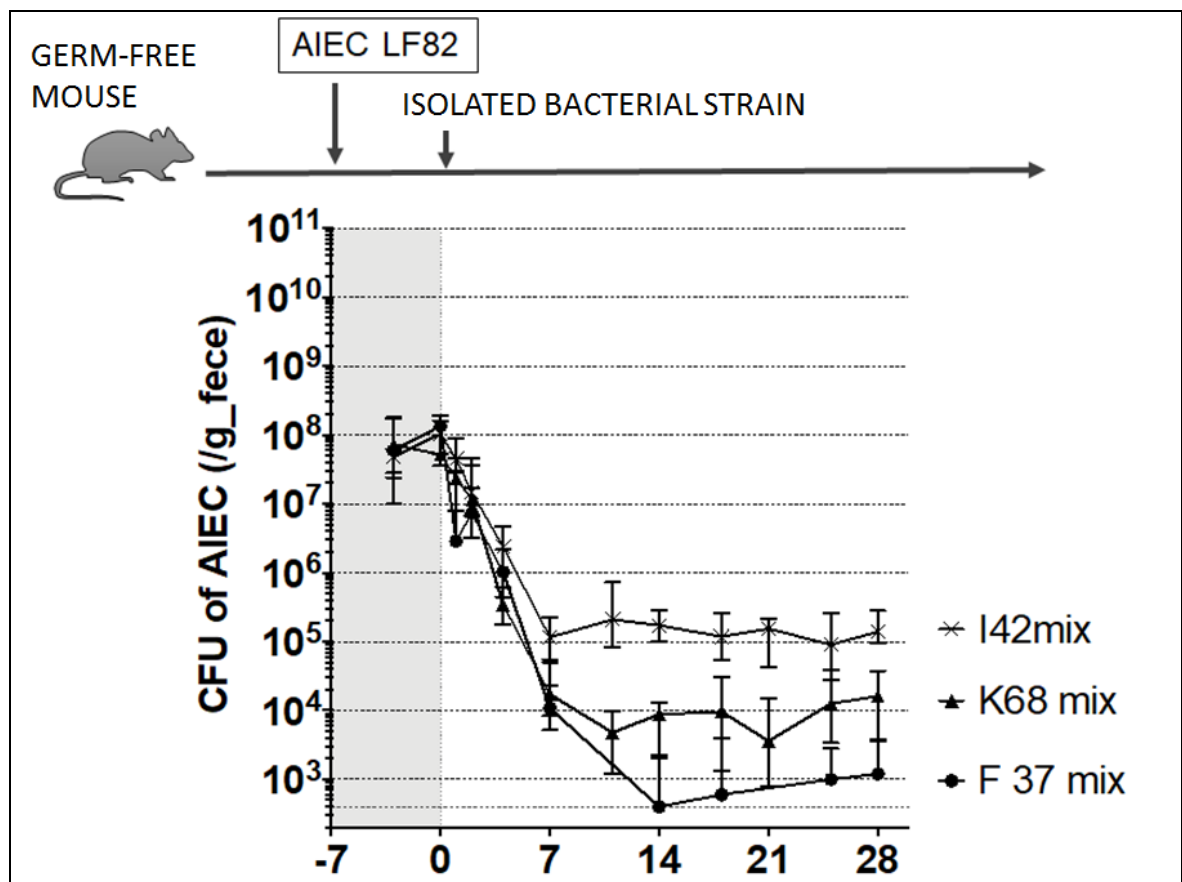


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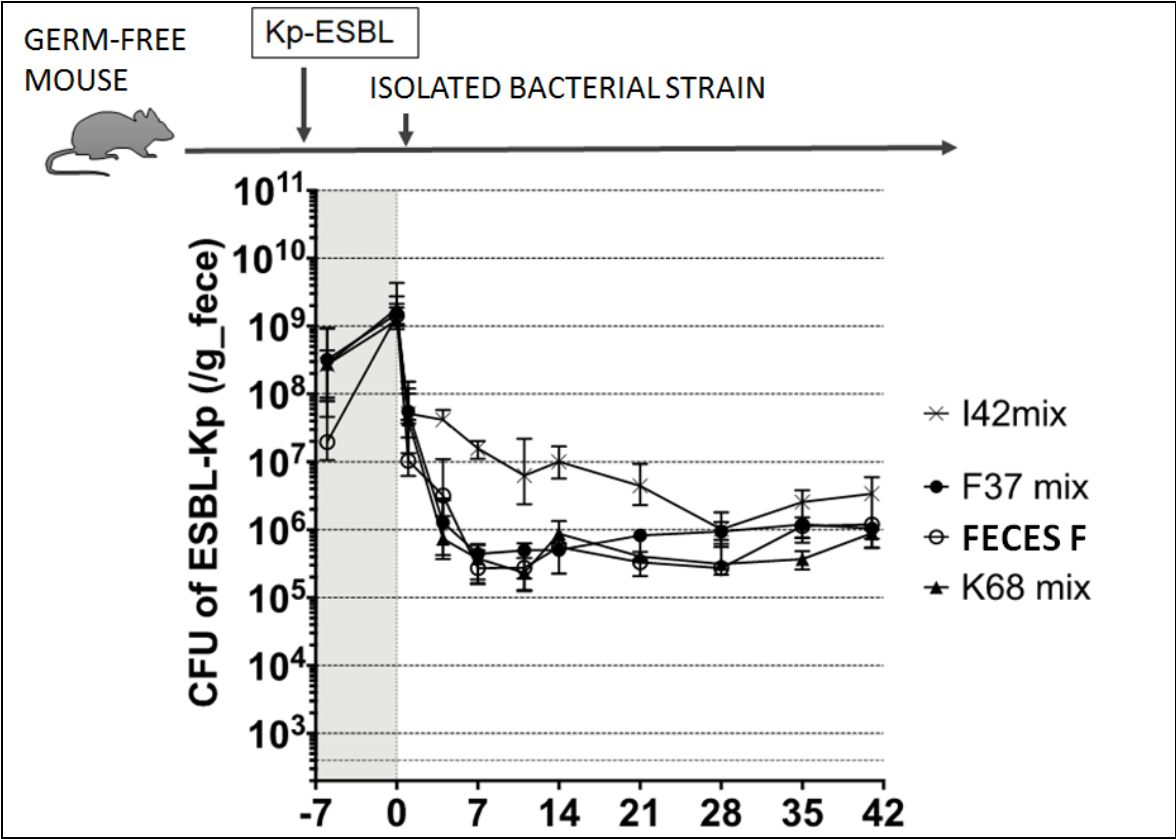


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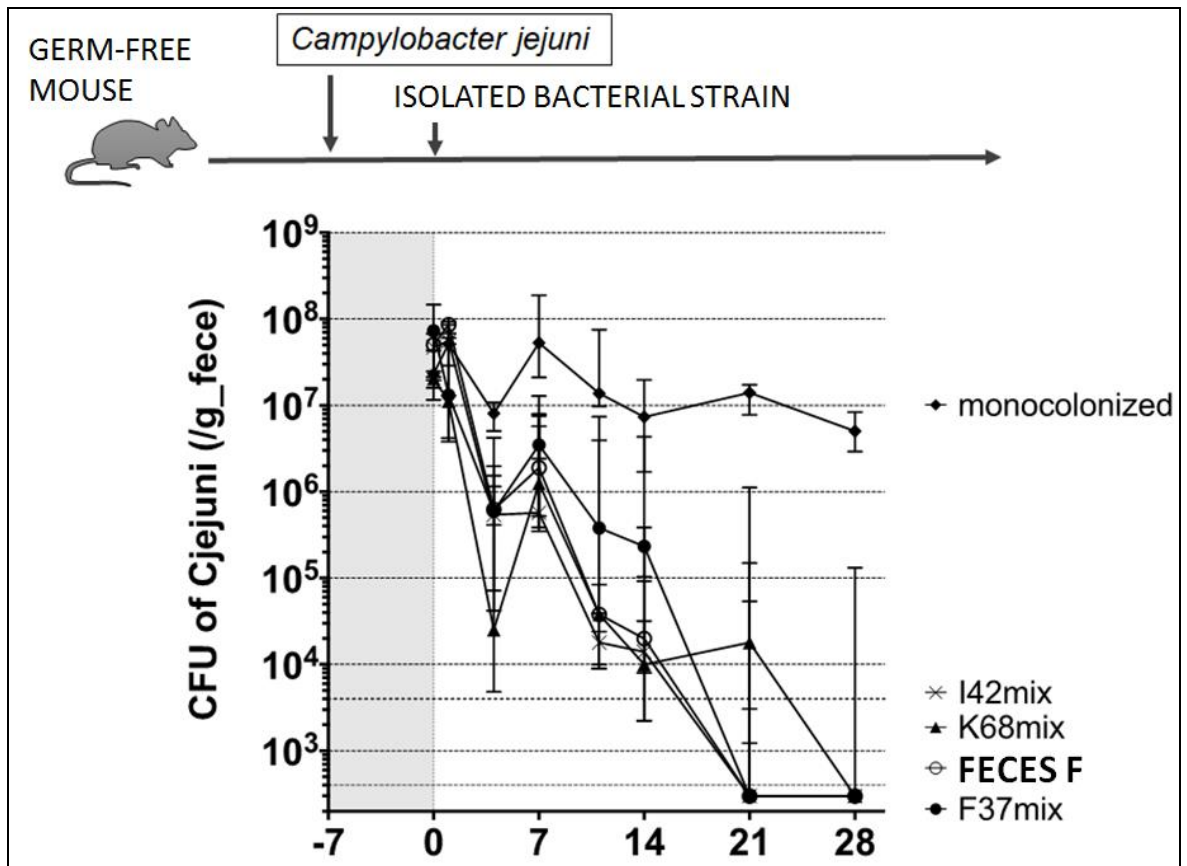


Fig. 25

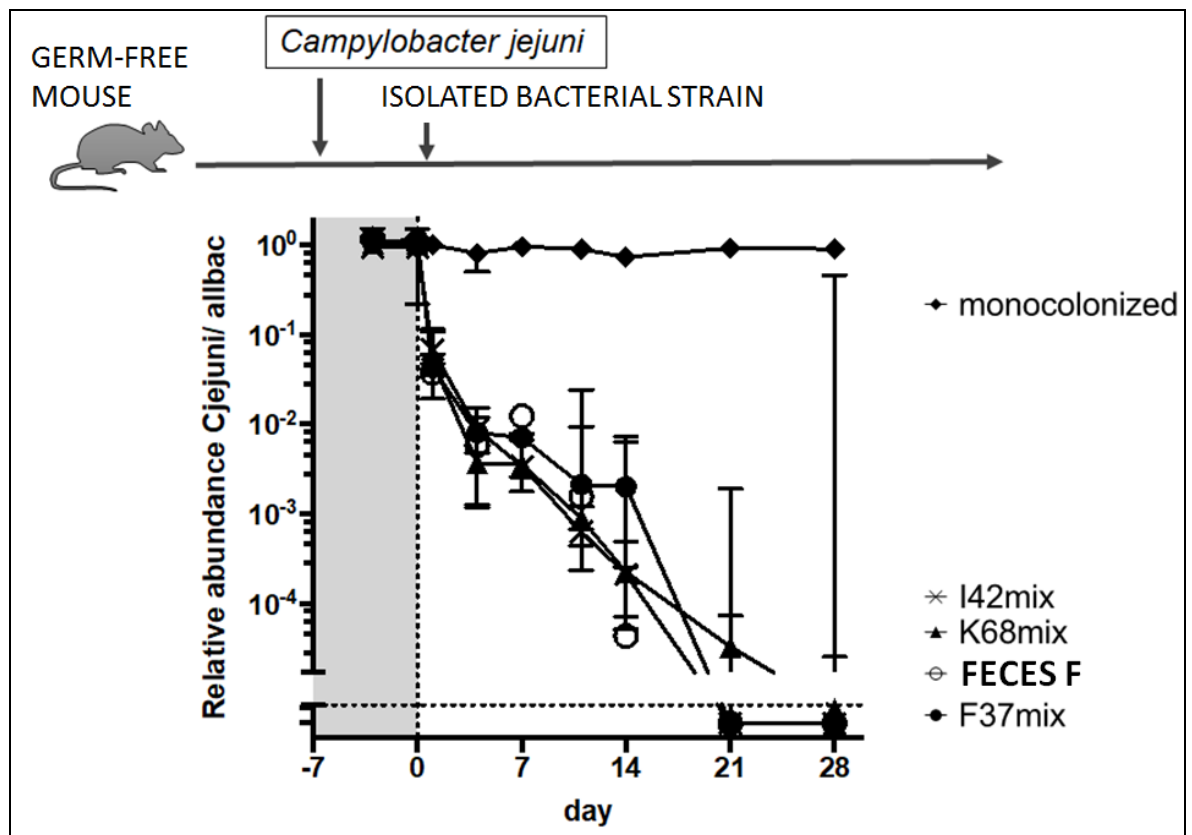
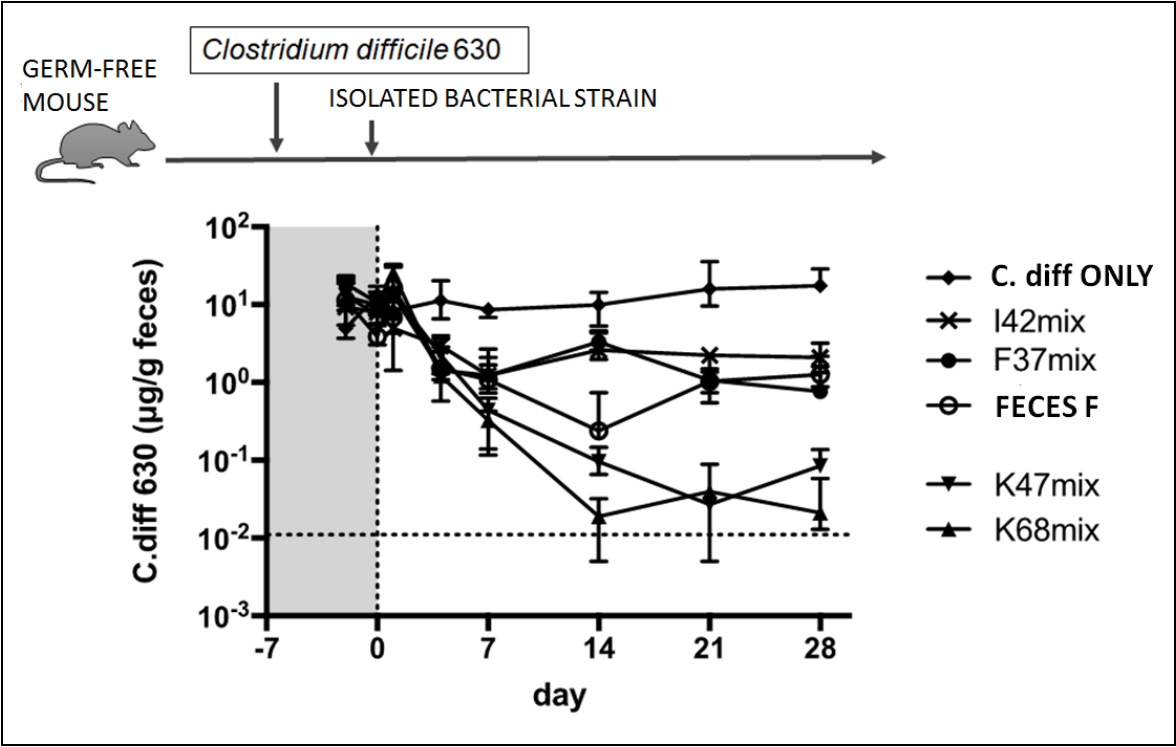


Fig. 26



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<223> K07

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<223> n is a, c, g, or t

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<223> K11

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gga						1383

<210> 13

<211> 1423

<212> DNA

<213> Unknown

<220>

<223> K13

<220>

<221> misc_feature

<222> (4)..(4)

<223> n is a, c, g, or t

<400> 13

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tcagcacgcc	gcggtgaata	cggtcccggg	ccttgtagac	accgcccgtc	acaccacgag	1380
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<210> 14

<211> 1381

<212> DNA

<213> Unknown

<220>

<223> K14

<400> 14

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g						1381

<210> 15

<211> 1391

<212> DNA

<213> Unknown

<220>

<223> K15

<400> 15

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tgctaatacc	gcataagcgc	acagtatcgc	atgatacagt	gtgaaaaact	cgggtggtac	180

aagatggacc	cgcgctctgat	tagctagttg	gtaaggtaac	ggcttaccaa	ggcgacgatc	240
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cgggaggcag	cagtggggaa	tattgcacaa	tgggcgaaag	cctgatgcag	cgacgcccg	360
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<210> 16

<211> 1402

<212> DNA

<213> Unknown

<220>

<223> K16

<400> 16

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ctagaatgg	ctgctaatac	cgcataagac	ctcagtaccg	catggtagag	gggtaaaaac	180
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<210> 17

<211> 1393

<212> DNA

<213> Unknown

<220>

<223> K17

<220>

<221> misc_feature

<222> (942)..(945)

<223> n is a, c, g, or t

<220>

<221> misc_feature

<222> (953)..(956)

<223> n is a, c, g, or t

<220>

<221> misc_feature
 <222> (1067)..(1069)
 <223> n is a, c, g, or t
 <400> 17

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gtaagatgga	cccgcgtctg	attaggtagt	tggtggggta	acggcccacc	aagccgacga	240
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<210> 18
 <211> 1389
 <212> DNA
 <213> Unknown

<220>
 <223> K18
 <400> 18

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gatggcatca	tgagttcaca	tgctccgatg	attaaaggta	ttttccggta	gacgatgggg	180
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accgcgagg						1389

<210> 19
 <211> 1393
 <212> DNA
 <213> Unknown
 <220>
 <223> K19
 <220>
 <221> misc_feature

<222> (1205)..(1206)

<223> n is a, c, g, or t

<400> 19

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<210> 20

<211> 1394

<212> DNA

<213> Unknown

<220>

<223> K20

<400> 20

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gagtgtctaat	accgcatgat	gcagttgggt	cgcatggctc	tgactgcaa	agattttatcg	180
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<210> 21

<211> 1385

<212> DNA

<213> Unknown

<220>

<223> K21

<400> 21

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tccgatggta	tattaaaacc	gcatgggtttt	actattaaag	aatttcggtt	atcgatgggg	180
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cgcaa						1385

<210> 22

<211> 1385

<212> DNA

<213> Unknown

<220>

<223> K22

<400> 22

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tctgagagga	agggtcccca	cattgggaact	gagacacggt	ccaaactcct	acgggaggca	300
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cgcaa						1385

<210> 23

<211> 1387

<212> DNA

<213> Unknown

<220>

<223> K23

<400> 23

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ccataaaaca	gggggtcccg	atgggaatat	ttgttaaaga	ttcatcgctg	atagataggc	180
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ccgcaag						1387

<210> 24

<211> 1384

<212> DNA

<213> Unknown

<220>

<223> K24

<400> 24

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gcgttccatt	agatagtagg	gggggtaacg	gccacactag	tcttcgatgg	ataggggttc	240
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<210> 25

<211> 1396

<212> DNA

<213> Unknown

<220>

<223> K25

<220>

<221> misc_feature

<222> (943)..(944)

<223> n is a, c, g, or t

<220>

<221> misc_feature

<222> (1072)..(1072)

<223> n is a, c, g, or t

<220>

<221> misc_feature

<222> (1290)..(1290)

<223> n is a, c, g, or t

<400> 25

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<210> 26
 <211> 1382
 <212> DNA
 <213> Unknown
 <220>
 <223> K26
 <400> 26

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ga						1382

<210> 27
 <211> 1370
 <212> DNA
 <213> Unknown
 <220>
 <223> K27
 <220>
 <221> misc_feature
 <222> (916)..(917)
 <223> n is a, c, g, or t
 <400> 27

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<210> 28
 <211> 1394
 <212> DNA
 <213> Unknown
 <220>
 <223> K28
 <220>
 <221> misc_feature
 <222> (909)..(911)
 <223> n is a, c, g, or t
 <400> 28

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 <223> n is a, c, g, or t

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 <222> (953)..(954)
 <223> n is a, c, g, or t

<400> 29

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 <220>
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 <400> 30

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 <223> K32
 <400> 32

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 <213> Unknown
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 <212> DNA
 <213> Unknown
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 <223> K34
 <400> 34

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 <223> K35

<220>

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<222> (901)..(901)

<223> n is a, c, g, or t

<400> 35

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<213> Unknown

<220>

<223> K36

<220>

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<223> n is a, c, g, or t

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<212> DNA

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<223> K37

<400> 37

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<211> 1396

<212> DNA

<213> Unknown

<220>

<223> K38

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<211> 1399

<212> DNA

<213> Unknown

<220>

<223> K39

<400> 39

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<212> DNA

<213> Unknown

<220>

<223> K40

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<213> Unknown

<220>

<223> K41

<400> 41

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<213> Unknown

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<211> 1388

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<213> Unknown

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<223> K43

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 <212> DNA
 <213> Unknown
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 <223> K44
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<213> Unknown

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<223> K50

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<212> DNA

<213> Unknown

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<223> K51

<400> 51

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<213> Unknown

<220>

<223> K52

<400> 52

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<213> Unknown

<220>

<223> K53

<400> 53

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<211> 1415

<212> DNA

<213> Unknown

<220>

<223> K54

<400> 54

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<211> 1366

<212> DNA

<213> Unknown

<220>

<223> K55

<400> 55

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<210> 56

<211> 1364

<212> DNA

<213> Unknown

<220>

<223> K56

<220>

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<222> (923)..(924)

<223> n is a, c, g, or t

<220>

<221> misc_feature

<222> (1349)..(1349)

<223> n is a, c, g, or t

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<221> misc_feature

<222> (1351)..(1352)

<223> n is a, c, g, or t

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<210> 57

<211> 1391

<212> DNA

<213> Unknown

<220>

<223> K57

<400> 57

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<210> 58

<211> 1377

<212> DNA

<213> Unknown

<220>

<223> K58

<400> 58

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<210> 59

<211> 1373

<212> DNA

<213> Unknown

<220>

<223> K59

<400> 59

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<210> 60

<211> 1384

<212> DNA

<213> Unknown

<220>

<223> K60

<400> 60

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 <211> 1394
 <212> DNA
 <213> Unknown
 <220>
 <223> K61
 <400> 61

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 <213> Unknown
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 <223> K64
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 <213> Unknown
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<213> Unknown

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<223> K67

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<212> DNA

<213> Unknown

<220>

<223> K68

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<222> (1352)..(1352)

<223> n is a, c, g, or t

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 <223> F02
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<213> Unknown

<220>

<223> F03

<400> 71

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<211> 1515

<212> DNA

<213> Unknown

<220>

<223> F04

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<212> DNA

<213> Unknown

<220>

<223> F05

<400> 73

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<212> DNA

<213> Unknown

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<223> F06

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<211> 1518

<212> DNA

<213> Unknown

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<223> F07

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<212> DNA

<213> Unknown

<220>

<223> F08

<400> 76

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<211> 1515

<212> DNA

<213> Unknown

<220>

<223> F09

<400> 77

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<211> 1518

<212> DNA

<213> Unknown

<220>

<223> F17

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<211> 1522

<212> DNA

<213> Unknown

<220>

<223> F18

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<213> Unknown

<220>

<223> F19

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<212> DNA

<213> Unknown

<220>

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<400> 88

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<213> Unknown

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<223> F21

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<212> DNA

<213> Unknown

<220>

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<211> 1519

<212> DNA

<213> Unknown

<220>

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<211> 1520

<212> DNA

<213> Unknown

<220>

<223> F24

<400> 92

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<212> DNA

<213> Unknown

<220>

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<213> Unknown

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<213> Unknown

<220>

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<213> Unknown

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<400> 96

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<213> Unknown

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<213> Unknown

<220>

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<212> DNA

<213> Unknown

<220>

<223> F31

<400> 99

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 <223> n is a, c, g, or t
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 <222> (923)..(923)
 <223> n is a, c, g, or t
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 <223> I03
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 <213> Unknown
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 <223> I04
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 <211> 1326
 <212> DNA
 <213> Unknown
 <220>
 <223> I05
 <400> 110

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<212> DNA
<213> Unknown
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<223> n is a, c, g, or t
<400> 111

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<211> 1338
<212> DNA
<213> Unknown
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<223> I07
<400> 112

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<210> 113
<211> 1337
<212> DNA
<213> Unknown
<220>
<223> I08
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<221> misc_feature
<222> (105)..(105)
<223> n is a, c, g, or t

<220>

<221> misc_feature

<222> (117)..(119)

<223> n is a, c, g, or t

<400> 113

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<211> 1353

<212> DNA

<213> Unknown

<220>

<223> I09

<400> 114

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<210> 115

<211> 1350

<212> DNA

<213> Unknown

<220>

<223> I10

<400> 115

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tagtaatcgc	gcatacgcca	cggcgcggtg	aatacgttcc	cgggccttgt	acacaccgcc	1320
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<210> 116

<211> 1356

<212> DNA

<213> Unknown

<220>

<223> I11

<400> 116

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ccgtctactc	ttggccagcc	ttctgaaagg	aagattaatc	caggatggga	tcatgagttc	120
acatgtccgc	atgattaaag	gtattttccg	gtagacgatg	gggatgcgtt	ccattagata	180
gtagcggggg	taacggccca	cctagtcac	gatggatagg	ggttctgaga	ggaagggtcc	240
ccacattgga	actgagacac	ggctcaaaact	cctacgggag	gcagcagtg	ggaatattgg	300
tcaatggg	atggcctgaa	ccagccaagt	agcgtgaagg	atgactgcc	tatgggttgt	360
aaacttcttt	tataaaggaa	taaagtccgg	tatgcatacc	cgtttgcatg	tactttatga	420
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ttcgtagta	atcgcgcac	agccacggcg	cgggtgaatac	gttcccgggc	cttgtagaca	1320
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<210> 117

<211> 1353

<212> DNA

<213> Unknown

<220>

<223> I12

<220>

<221> misc_feature

<222> (6)..(6)

<223> n is a, c, g, or t

<220>

<221> misc_feature

<222> (8)..(8)

<223> n is a, c, g, or t

<400> 117

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atgggaatat	ttgttaaaga	ttcatcgctg	atagataggc	atgcgttcca	ttaggcagtt	180
ggcggggtaa	cggccaccca	aaccgacgat	ggataggggt	tctgagagga	aggtcccca	240
cattgggtact	gagacacgga	ccaaactcct	acgggaggca	gcagtgagga	atattgggtca	300
atggccgaga	ggctgaacca	gccaaagtcg	gtgaaggaag	aaggatctat	ggtttgtaaa	360
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gctagtaatc	gcgcatcagc	catggcgcg	tgaatacggt	cccgggcctt	gtacacaccg	1320
cccgtaagc	catgggagcc	gggggtacct	gaa			1353

<210> 118

<211> 824

<212> DNA

<213> Unknown

<220>

<223> I13

<400> 118

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attcatcgct	gatagatagg	catgcgttcc	attaggcagt	tggcggggta	acggcccacc	180
aaaccgacga	tggatagggg	ttctgagagg	aagggtcccc	acattgggtac	tgagacacgg	240
accaaactcc	tacgggaggc	agcagtgagg	aatattgggtc	aatgggcgta	agcctgaacc	300
agccaagtgc	cgtgagggat	gaaggttcta	tggtatcgtaa	acctctttta	taagggaata	360
aagtgcggga	cgtgtcctgt	tttgtatgta	ccttatgaat	aaggatcggc	taactccgtg	420
ccagcagccg	cggtaatacg	gaggatccga	gcgttatccg	gatttatttg	gtttaaagg	480
tgcgtaggcg	gccttttaag	tcagcgggtga	aagtctgtgg	ctcaaccata	gaattgccgt	540
tgaactggg	gggcttgagt	atgtttgagg	caggcggaat	gcgtgggtgta	gcgggtgaaat	600
gcttagatat	cacgcagaac	cccgattgcg	aaggcagcct	gccaagccat	gactgacgct	660
gatgcacgaa	agcgtgggga	tcaaacagga	ttagataccc	tggtagtcca	cgagtaaac	720
gatgatcact	agctgtttgc	gatacagtg	aagcggcaca	gcgaaagcgt	taagtgatcc	780
acctggggag	tacgccggca	acgggtgaaac	tcaaaggaat	tgac		824

<210> 119

<211> 1348

<212> DNA

<213> Unknown

<220>

<223> I14

<400> 119

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cttttatggt	tgaatttcc	gatggttaga	gatgggcatg	cggtgtatta	gctagtgtgt	180
ggggtaacgg	ctcaccaagg	cgacgataca	taggggggact	gagagggttaa	ccccccacac	240
tggtactgag	acacggacca	gactcctacg	ggaggcgagca	gtgaggaata	ttgggtcaatg	300
gacgcaagtc	tgaaccagcc	atgccgcgtg	caggatgacg	gctctatgag	ttgtaaactg	360
cttttgtacg	agggtaaacg	cagatacgtg	tatctgtctg	aaagtatcgt	acgaataagg	420
atcggtctaac	tccgtgccag	cagccgcggg	aatacggagg	attcaagcgt	tatccggatt	480
tattgggttt	aaagggtg	taggcgggtt	gataagttag	aggtgaaatt	tcggggctca	540
accctgaacg	tgcttctaata	actgttgagc	tagagagtag	ttgcggtagg	cggaatgtat	600
ggtgtagcgg	tgaatgctt	agagatcata	cagaacaccg	attgcgaagg	cagcttacca	660
aactatatct	gacgttgagg	cacgaaagcg	tgggggagcaa	acaggattag	ataccctggg	720
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aagcgataag	ttatccacct	ggggagtacg	ttcgcaagaa	tgaaactcaa	aggaattgac	840
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cgcaaccctt	accgttagtt	gcatcaggt	gaagctgggc	actctggcgg	gactgccggt	1080
gtaagccgag	aggaaaggtg	ggatgacgtc	aaatcagcac	ggcccttacg	tccggggcta	1140
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gtaatcgcg	atcagccatg	gcgcggtgaa	tacgttcccc	ggccttgtag	acaccgccc	1320
tcaagccatg	ggagccgggg	gtgcctga				1348

<210> 120
 <211> 1337
 <212> DNA
 <213> Unknown
 <220>
 <223> I15
 <220>
 <221> misc_feature
 <222> (838)..(839)
 <223> n is a, c, g, or t
 <400> 120

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tattaaagaa	tttcgggtcat	tgatggggat	gcgttccatt	aggttgttgg	cggggtaacg	180
gcccaccaag	ccatcgatgg	ataggggttc	tgagaggaag	gtccccaca	ttggaactga	240
gacacggtcc	aaactcctac	gggaggcagc	agtgaggaat	attgggtcaat	ggacgagagt	300
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ctccgtgcc	gcagcccgcg	taatacggag	gatccgagcg	ttatccggat	ttattgggtt	480
taaagggagc	gtaggcggat	gcttaagtca	gttgtgaaag	tttgcggtc	aaccgtaaaa	540
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tgacgctgat	gctcgaaagt	gtgggtatca	aacaggatta	gataccctgg	tagtccacac	720
agtaaacgat	gaatactcgc	tgtttgcgat	atacagtaag	cggccaagcg	aaagcgtaa	780
gtattccacc	tggggagtag	gccggcaacg	gtgaaactca	aaggaattga	cgggggcnng	840
cacaagcgga	ggaacatgtg	gtttaattcg	atgatacgcg	aggaacctta	cccgggctta	900
aattgcaaat	gaatgttctg	gaaacagatc	agccgcaagg	catttgtgaa	ggtgctgcat	960
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tcagttcgga	ttggagtctg	caacccgact	ccatgaagct	ggattcgcta	gtaatcgcg	1260
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aaagccgggg	gtacctg					1337

<210> 121
 <211> 1343
 <212> DNA
 <213> Unknown
 <220>
 <223> I16
 <220>
 <221> misc_feature
 <222> (5)..(5)
 <223> n is a, c, g, or t
 <220>
 <221> misc_feature
 <222> (7)..(7)
 <223> n is a, c, g, or t
 <220>
 <221> misc_feature
 <222> (110)..(113)
 <223> n is a, c, g, or t
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 <221> misc_feature
 <222> (337)..(338)
 <223> n is a, c, g, or t
 <220>
 <221> misc_feature
 <222> (1170)..(1170)
 <223> n is a, c, g, or t

<400> 121

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aactccggaa	tagcctttcg	aaagaaagat	taataccgga	tagcatatcn	nnntcgcatg	120
atatttttat	taaagaattt	cggttatcga	tggggatgcg	ttccattagt	ttgttggcgg	180
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gaactgagac	acggtccaaa	ctcctacggg	aggcagcagt	gaggaatatt	ggtcaatggg	300
cgagagcctg	aaccagccaa	gtagcgtgaa	ggatganngc	tctatgggtc	gtaaaccttct	360
tttatatggg	aataaagttt	tccacgtgtg	gaattttgta	tgtaccatat	gaataaggat	420
cggctaactc	cgtgccagca	gccgcggtaa	tacggaggat	ccgagcgtta	tccggattta	480
ttgggtttta	agggagcgtg	ggtggattgt	taagtcagtt	gtgaaagttt	gcggctcaac	540
cgtaaaattg	cagttgaaac	tggcagtcct	gagtacagta	gagggtggcg	gaattcgtgg	600
tgtagcgggtg	aaatgcttag	atatcacgaa	gaactccgat	tgcgaaggca	gctcactaga	660
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tccacacagt	aaacgatgaa	tactcgtctg	ttgcgatata	cagtaagcgg	ccaagcgaaa	780
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gctgcattgt	tgtcgtcagc	tctgtccgtg	aggtgtcggc	ttaagtcca	taacgagcgc	1020
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acctctctca	gttcggatcg	aagtctgcaa	cccgaattcg	tgaagctgga	ttcgctagta	1260
atcgcgcatc	agccatggcg	cggtgaatac	gttcccgggc	cttgtagaca	ccgcccgtca	1320
agccatgaaa	gccgggggta	cct				1343

<210> 122

<211> 1335

<212> DNA

<213> Unknown

<220>

<223> I17

<400> 122

gggtggcgac	cggcgacagg	gtgcgtaacg	cgtatgcaac	ctaccataa	cagggggata	60
acactgagaa	attggtacta	ataccccata	acatcagaac	cggcatcggg	tttggttgaa	120
aactccgggtg	gttatggatg	ggcatgcgtt	gtattagctg	gttggtgagg	taacggctca	180
ccaaggcaac	gatacatagg	gggactgaga	ggttaacccc	ccacattggt	actgagacac	240
ggaccaaaact	cctacgggag	gcagcagtga	ggaatattgg	tcaatggacg	caagtctgaa	300
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taaacgcttc	tacgtgtagg	agcctgaaag	tatcgtagca	ataaggatcg	gctaactccg	420
tgccagcagc	cgcggtaata	cggaggatcc	aagcgttatc	cggattttatt	gggtttaaag	480
ggtgcgtagg	cggtttgata	agttagaggt	gaaataccgg	tgcttaacac	cggaactgcc	540
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atgccttagag	atcatacaga	acaccgattg	cgaaggcagc	ttaccaaact	atatctgacg	660
ttgaggcacg	aaagcgtggg	gagcaaacag	gattagatac	cctggtagtc	cacgcagtaa	720
acgatgataa	ctcgtctgctg	gcgatacaca	gtcggcggct	aagcgaagc	gataagttat	780
ccacctgggg	agtacgttcg	caagaatgaa	actcaaagga	attgacgggg	gcccgcacaa	840
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ttcggatcgg	aggctgaaac	ccgcctccgt	gaagttggat	tcgctagtaa	tcgcgcatca	1260
gccatggcgc	ggtgaatacg	ttcccgggccc	ttgtacacac	cgcccgtcaa	gccatggaag	1320
ctgggggtgc	ctgaa					1335

<210> 123

<211> 1309

<212> DNA

<213> Unknown

<220>

<223> I18

<400> 123

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ctattagtta	gttggcgggg	taacagccca	ccaagacgat	gataggtagg	ggttctgaga	180
ggaagggtccc	ccacattgga	actgagacac	ggtccaaact	cctacgggag	gcagcagtga	240
ggaatattgg	tcaatggacg	taagtctgaa	ccagccaagt	cgcgtgaggg	aagactgccc	300
tatgggttgt	aaacctcttt	tataagggaa	gaataagttc	tacgtgtaga	atgatgcctg	360

taccttatga	ataagcatcg	gctaactccg	tgccagcagc	cgcggtaata	cggaggatgc	420
gagcggtatc	cggattttatt	gggttttaaag	gggtgcgtagg	cggttttatta	agtttagtggt	480
taaataatttg	agctaaactc	aattgtgcca	ttaatactgg	taaactggag	tacagacgag	540
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gaagttggat	tcgctagtaa	tcgcgcatca	gccatggcgc	ggtgaatacg	ttcccggggc	1260
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<210> 124

<211> 1339

<212> DNA

<213> Unknown

<220>

<223> I19

<220>

<221> misc_feature

<222> (10)..(10)

<223> n is a, c, g, or t

<220>

<221> misc_feature

<222> (124)..(125)

<223> n is a, c, g, or t

<220>

<221> misc_feature

<222> (183)..(183)

<223> n is a, c, g, or t

<220>

<221> misc_feature

<222> (190)..(191)

<223> n is a, c, g, or t

<220>

<221> misc_feature

<222> (911)..(911)

<223> n is a, c, g, or t

<400> 124

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caaagagggg	gacaacagtt	ggaaacgact	gctaataacc	cataagccca	cgggtcggca	120
tcgnncagag	ggaaaaggag	caatccgctt	tgagatggcc	tcgcgtccga	ttagctagtt	180
gngaggtan	ngggccacca	aggcgacgat	cggtagccgg	actgagaggt	tgaacggcca	240
cattgggact	gagacacggc	ccagactcct	acgggaggca	gcagtgggga	atattgcaca	300
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tcctgttggt	gggggaagata	atgacggtag	ccaacaagga	agtgacggct	aactacgtgc	420
cagcagccgc	ggtaaaaacgt	aggtcacaa	cggtgtccgg	aattactggg	tgtaaaggga	480
gcgcaggcgg	gaagacaagt	tggaaagtga	atctatgggc	tcaaccata	aactgctttc	540
aaaactgttt	ttcttgagta	gtgcagaggt	aggcgggaatt	cccgggtgtag	cgggtggaatg	600
cgtagatata	gggaggaaca	ccagtggcga	aggcggccta	ctgggcacca	actgacgctg	660
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ccacctgggg	agtacgaccg	caaggttgaa	actcaaagga	attgacgggg	gcccgcacaa	840
gcagtggagt	atgtggttta	attcgacgca	acgcgaagaa	ccttaccaag	tcttgacatc	900
ccttgacaga	natagaaata	tgttttctct	tcggagcaag	gagacagggt	gtgcatgggt	960
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gatgacgtca	aatcatcatg	ccctttatga	cttgggctac	acacgtacta	caatggcgtt	1140
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ggacccgaag	tcggtagtgc					1339

<210> 125

<211> 1339
 <212> DNA
 <213> Unknown
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 <223> I20
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 <221> misc_feature
 <222> (192)..(192)
 <223> n is a, c, g, or t
 <220>
 <221> misc_feature
 <222> (205)..(206)
 <223> n is a, c, g, or t
 <220>
 <221> misc_feature
 <222> (1337)..(1337)
 <223> n is a, c, g, or t
 <400> 125

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atcgaccaga	gggaaaagga	gtaatccgct	ttgagatggc	ctcgcgtccg	attagctagt	180
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gggacccgaa	gtcggtngt					1339

<210> 126
 <211> 1354
 <212> DNA
 <213> Unknown
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 <223> I21
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 <221> misc_feature
 <222> (184)..(187)
 <223> n is a, c, g, or t
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 <221> misc_feature
 <222> (573)..(574)
 <223> n is a, c, g, or t
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 <221> misc_feature
 <222> (752)..(755)
 <223> n is a, c, g, or t
 <220>
 <221> misc_feature
 <222> (1175)..(1176)
 <223> n is a, c, g, or t
 <400> 126

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cagagctgca	tggctcagtg	tgaaaaactc	cgggtgtata	agatggaccc	gcgttggatt	180
agcnnntgg	tgggtaacg	gcccaccaag	gcgacgatcc	atagccggcc	tgagaggggtg	240

aacggccaca	ttgggactga	gacacggccc	agactcctac	gggaggcagc	agtggggaat	300
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atgtaaaact	ctatcagcag	ggaagatagt	gacggtacct	gactaagaag	ccccggctaa	420
ctacgtgcc	gcagccgcgg	taatacgtag	ggggcaagcg	ttatccggat	ttactgggtg	480
taaagggagc	gtagacgggtg	tggcaagtct	gatgtgaaag	gcatgggctc	aacctgtgga	540
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gtgaaatgcg	tagatattag	gaggaacacc	agtggcgaag	gcggcttact	ggacggtaac	660
tgacgttgag	gctcgaaagc	gtggggagca	aacaggatta	gataccctgg	tagtccacgc	720
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taagtattcc	acctggggag	tacgttcgca	agaatgaaac	tcaaaggaat	tgacggggac	840
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aacgtcccag	ttcggactgt	agtctgcaac	ccgactacac	gaagctggaa	tcgctagtaa	1260
tcgcggatca	gaatgccgcg	gtgaatacgt	tcccgggtct	tgtacacacc	gcccgtcaca	1320
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<210> 127

<211> 1264

<212> DNA

<213> Unknown

<220>

<223> I22

<220>

<221> misc_feature

<222> (144)..(145)

<223> n is a, c, g, or t

<400> 127

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agaccacagc	accgcatggt	gcannngtaa	aaactccggt	ggtagtagat	ggacccgcgt	180
ctgattagct	ggttgggtggg	gtaacggcct	accaaggcga	cgatcagtag	ccggcctgag	240
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gggaatatgg	cacaatgggg	gaaaccctga	tgcagcgacg	ccgcgtgaag	gaagaagtat	360
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gagaggtaag	cgggaattcct	agtgtagcgg	tgaaatgcgt	agatattagg	aggaacacca	540
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aaagccattc	ggtgccgcag	caaacgcaat	aagtagtcca	cctggggagt	acgttcgcaa	720
gaatgaaact	caaaggaatt	gacggggacc	cgcacaagcg	gtggagcatg	tggtttaatt	780
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cgactacatg	aagctggaat	cgctagtaat	cgcgaatcag	aatgtcgcgg	tgaatacgtt	1200
cccgggtctt	gtacacaccg	cccgtcacac	catgggagtc	agtaacgccc	gaagtcagtg	1260
accc						1264

<210> 128

<211> 1340

<212> DNA

<213> Unknown

<220>

<223> I23

<400> 128

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tgatttgatt	atcaaagatt	tatcgctgaa	agatgagctc	gcgtctgatt	agctagttag	180
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ctgtcttcag	ggacgaaaaa	tgacgggtacc	tgaggaggaa	gctccggcta	actacgtgcc	420
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cgtaggcggg	atcgcaagtc	agatgtgaaa	actatgggct	taaccataa	actgcatttg	540
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gattgtaggc	tgcaaccgcg	ctacatgaag	tcggaattgc	tagtaatcgc	agatcagcat	1260
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<210> 129
 <211> 1337
 <212> DNA
 <213> Unknown
 <220>
 <223> I24
 <220>
 <221> misc_feature
 <222> (53)..(54)
 <223> n is a, c, g, or t
 <220>
 <221> misc_feature
 <222> (113)..(115)
 <223> n is a, c, g, or t
 <220>
 <221> misc_feature
 <222> (123)..(125)
 <223> n is a, c, g, or t
 <220>
 <221> misc_feature
 <222> (1155)..(1156)
 <223> n is a, c, g, or t
 <220>
 <221> misc_feature
 <222> (1336)..(1336)
 <223> n is a, c, g, or t
 <400> 129

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gcgcaggcgg	gaagacaagt	tggaaagtga	atctatgggc	tcaaccata	aactgctttc	540
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cgtagatatc	gggaggaaca	ccagtggcga	aggcggccta	ctgggcacca	actgacgctg	660
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gatgacgtca	aatcatcatg	ccctttatga	cttgggctac	acacgtacta	caatggcgtt	1140
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<210> 130
 <211> 1353

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<212> DNA
<213> Unknown
<220>
<223> I25
<220>
<221> misc_feature
<222> (570)..(572)
<223> n is a, c, g, or t
<400> 130
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<210> 131
<211> 1361
<212> DNA
<213> Unknown
<220>
<223> I26
<220>
<221> misc_feature
<222> (9)..(9)
<223> n is a, c, g, or t
<220>
<221> misc_feature
<222> (939)..(940)
<223> n is a, c, g, or t
<220>
<221> misc_feature
<222> (1052)..(1053)
<223> n is a, c, g, or t
<220>
<221> misc_feature
<222> (1357)..(1357)
<223> n is a, c, g, or t
<400> 131
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tgtggtagtc tttggaaact gtgcagctag agtgctggag aggtgaagtgg aattcctagt 600
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gatgactgac gttgaggctc gaaagcgtgg ggagcaaaaca ggattagata ccctggtagt 720
ccacgccgta aacgatgact gctagggtgtc gggtagcaaa gctattcggg gccgcagcta 780

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<210> 132
 <211> 1345
 <212> DNA
 <213> Unknown
 <220>
 <223> I27
 <400> 132

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 <213> Unknown
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 <223> n is a, c, g, or t
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<223> I29

<400> 134

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<211> 1358

<212> DNA

<213> Unknown

<220>

<223> I30

<400> 135

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<211> 1342

<212> DNA

<213> Unknown

<220>

<223> I31

<400> 136

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<212> DNA

<213> Unknown

<220>

<223> I32

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<222> (972)..(972)

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<221> misc_feature

<222> (983)..(983)

<223> n is a, c, g, or t

<220>

<221> misc_feature

<222> (990)..(991)

<223> n is a, c, g, or t

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 <212> DNA
 <213> Unknown
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 <223> I34
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<223> n is a, c, g, or t
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aagcgaagtc gcgagacgga gcggacctca taaacgcact cccagttcag attgcaggct 1260
gcaacccgcc tgcatagaat aggaatcgct agtaatcgcg ggtagcagata ccgcggtgaa 1320
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<210> 141
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<222> (115)..(117)
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<222> (1200)..(1201)
<223> n is a, c, g, or t
<400> 141
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gcatgacgan	atgaagaaag	ggaggcccttc	gggcttttcgc	tggaggagg	gcttgctgtct	180
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<211> 1364

<212> DNA

<213> Unknown

<220>

<223> I37

<220>

<221> misc_feature

<222> (939)..(940)

<223> n is a, c, g, or t

<400> 142

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aagccaatcc	cagaaaaccg	atcgtagtcc	ggattgcagt	ctgcaactcg	actgcatgaa	1260
gtcggaaatcg	ctagtaatcg	cggatcagca	tgccgcgggtg	aatacgttcc	cgggtcttgt	1320
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<210> 143

<211> 1343

<212> DNA

<213> Unknown

<220>

<223> I38

<400> 143

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ttaaaggagc	aatccgctat	gagatggacc	cgcgcgcat	tagctagttg	gtaaggtaat	180

ggcttaccaa	ggcgacgatg	cgtagccgac	ctgagagggt	gatcgccac	attgggactg	240
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cctgatgcag	caacgccgcg	tgagtgatga	cggtcttcgg	attgtaaagc	tctgtctttg	360
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tctagagtgc	aggaggggag	agtggaaattc	ctagtgtagc	ggtgaaatgc	gtagagatta	600
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gaatacgttc	ccgggccttg	tacacaccgc	ccgtcacacc	atgagagttg	gcaataacca	1320
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<210> 144
 <211> 1318
 <212> DNA
 <213> Unknown
 <220>
 <223> I39
 <400> 144

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accagtggcg	aaggcgccct	gctgggcctt	aactgacgct	gaggcacgaa	agcatgggta	660
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 <212> DNA
 <213> Unknown
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 <223> I40
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 <213> Unknown
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 <223> I41
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<210> 147
 <211> 1341
 <212> DNA
 <213> Unknown
 <220>
 <223> I42
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 <221> misc_feature
 <222> (120)..(121)
 <223> n is a, c, g, or t
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ctaggtgtgg	ggtggctgac	ccattccgtg	ccggagttaa	cacaataagt	aatccacctg	780
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<210> 148
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<212> DNA
<213> Artificial Sequence
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<223> 27Forward-mod
<400> 148
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<210> 149
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<212> DNA
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<223> 1492Reverse
<400> 149
ggytaccttg ttacgactt 19

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<210> 150
<211> 20
<212> DNA
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<220>
<223> f01_42H6_Fw1
<400> 150
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<210> 151
<211> 20
<212> DNA
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<223> f01_42H6_Rv1
<400> 151
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<210> 152
<211> 20
<212> DNA
<213> Artificial Sequence
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<223> f02_42I8_Fw1
<400> 152
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<223> f02_42I8_Rv3
<400> 153
gcggttttaa tatactatcg g 21

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<210> 154
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<220>

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<223> f05_43D4_Fw4
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<210> 155
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<223> f05_43D4_Rv2
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<210> 156
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<223> f06_42F4_Fw1
<400> 156
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<210> 157
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<210> 161
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<223> f08_42B5_Rv1
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