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(54) Title: ALPHA (1,2) FUCOSYLTRANSFERASE SYNGENES FOR USE IN THE PRODUCTION OF FUCOSYLATED OLIGOSACCHARIDES

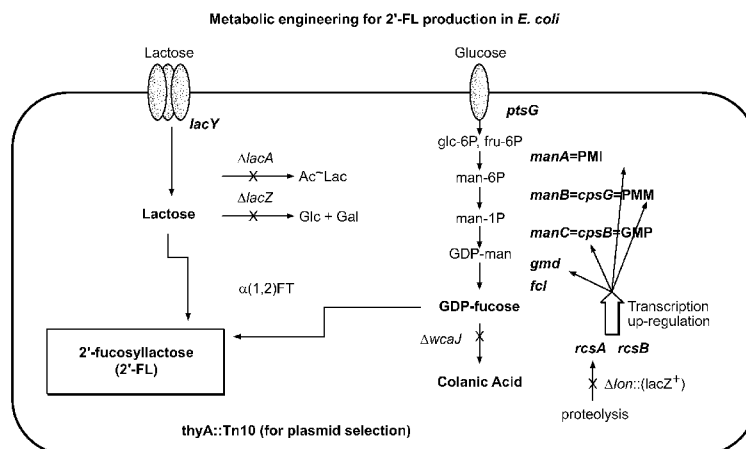


FIG. 2

(57) Abstract: The invention provides compositions and methods for engineering *E. coli* or other host production bacterial strains to produce fucosylated oligosaccharides, and the use thereof in the prevention or treatment of infection.

**ALPHA (1,2) FUCOSYLTRANSFERASE SYNGENES FOR USE IN THE
PRODUCTION OF FUCOSYLATED OLIGOSACCHARIDES**

FIELD OF THE INVENTION

The invention provides compositions and methods for producing purified oligosaccharides, in particular certain fucosylated oligosaccharides that are typically found in human milk.

BACKGROUND OF THE INVENTION

Human milk contains a diverse and abundant set of neutral and acidic oligosaccharides. More than 130 different complex oligosaccharides have been identified in human milk, and their structural diversity and abundance is unique to humans. Although these molecules may not be utilized directly by infants for nutrition, they nevertheless serve critical roles in the establishment of a healthy gut microbiome, in the prevention of disease, and in immune function. Prior to the invention described herein, the ability to produce human milk oligosaccharides (HMOS) inexpensively was problematic. For example, their production through chemical synthesis was limited by stereo-specificity issues, precursor availability, product impurities, and high overall cost. As such, there is a pressing need for new strategies to inexpensively manufacture large quantities of HMOS.

SUMMARY OF THE INVENTION

The invention features an efficient and economical method for producing fucosylated oligosaccharides. Such production of a fucosylated oligosaccharide is accomplished using an isolated nucleic acid comprising a sequence encoding a lactose-utilizing α (1,2) fucosyltransferase gene product (e.g., polypeptide or protein), which is operably linked to one or more heterologous control sequences that direct the production of the recombinant fucosyltransferase gene product in a host production bacterium such as *Escherichia coli* (*E. coli*).

The present disclosure provides novel α (1,2) fucosyltransferases (also referred to herein as α (1,2) FTs) that utilize lactose and catalyzes the transfer of an L-fucose sugar from a GDP-fucose donor substrate to an acceptor substrate in an alpha-1,2-linkage. In a preferred embodiment, the acceptor substrate is an oligosaccharide. The α (1,2) fucosyltransferases identified and described herein are useful for expressing in host bacterium for the production of human milk oligosaccharides (HMOS), such as fucosylated oligosaccharides. Exemplary

fucosylated oligosaccharides produced by the methods described herein include 2'-

fucosyllactose (2'FL), lactodifucotetraose (LDFT), lacto-N-fucopentaose I (LNF I), or lacto-N-difucohexaose I (LDFH I). The "α(1,2) fucosyltransferases" disclosed herein encompasses the amino acid sequences of the α(1,2) fucosyltransferases and the nucleic acid sequences that encode the α(1,2) fucosyltransferases, as well as variants and fragments thereof that exhibit α(1,2) fucosyltransferase activity. Also within the invention is a nucleic acid construct comprising an isolated nucleic acid encoding a lactose-accepting α (1,2) fucosyltransferase enzyme, said nucleic acid being optionally operably linked to one or more heterologous control sequences that direct the production of the enzyme in a host bacteria production strain.

The amino acid sequence of the lactose-accepting α(1,2) fucosyltransferases described herein is at least 15%, at least 20%, at least 25%, at least 30%, at least 35%, at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% identity to *Helicobacter pylori* 26695 alpha-(1,2) fucosyltransferase (futC or SEQ ID NO: 1). Preferably, the lactose-accepting α(1,2) fucosyltransferases described herein is at least 22% identical to *H. pylori* FutC, or SEQ ID NO: 1.

In another aspect, the amino acid sequence of the lactose-accepting α(1,2) fucosyltransferases described herein is at least 15%, at least 20%, at least 25%, at least 30%, at least 35%, at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% identity to *Bacteroides vulgatus* alpha-(1,2) fucosyltransferase (FutN or SEQ ID NO: 3). Preferably, the lactose-accepting α(1,2) fucosyltransferases described herein is at least 25% identical to *B. vulgatus* FutN, or SEQ ID NO: 3.

Alternatively, the exogenous α (1,2) fucosyltransferase preferably comprises at least at least 15%, at least 20%, at least 25%, at least 30%, at least 35%, at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% identity to any one of the novel α (1,2) fucosyltransferases disclosed herein, for example, to the amino acid sequences in Table 1.

Exemplary α(1,2) fucosyltransferases include, but are not limited to, *Prevotella melaninogenica* FutO, *Clostridium bolteae* FutP, *Clostridium bolteae* +13 FutP, *Lachnospiraceae* sp. FutQ, *Methanosphaerula palustris* FutR, *Tannerella* sp. FutS, *Bacteroides caccae* FutU, *Butyrivibrio* FutV, *Prevotella* sp. FutW, *Parabacteroides johnsonii* FutX, *Akkermansia muciniphilia* FutY, *Salmonella enterica* FutZ, *Bacteroides* sp. FutZA.

For example, the $\alpha(1,2)$ fucosyltransferases comprise the amino acid sequences comprising any one of the following: *Prevotella melaninogenica* FutO (SEQ ID NO: 10), *Clostridium bolteae* FutP (SEQ ID NO: 11), *Clostridium bolteae* +13 FutP (SEQ ID NO: 292), *Lachnospiraceae* sp. FutQ (SEQ ID NO: 12), *Methanosphaerula palustris* FutR (SEQ ID NO: 13), *Tannerella* sp. FutS (SEQ ID NO: 14), *Bacteroides caccae* FutU (SEQ ID NO: 15), *Butyrivibrio* FutV (SEQ ID NO: 16), *Prevotella* sp. FutW (SEQ ID NO: 17), *Parabacteroides johnsonii* FutX (SEQ ID NO: 18), *Akkermansia muciniphilia* FutY (SEQ ID NO: 19), *Salmonella enterica* FutZ (SEQ ID NO: 20), and *Bacteroides* sp. FutZA (SEQ ID NO: 21), or a functional variant or fragment thereof. Other exemplary $\alpha(1,2)$ fucosyltransferases include any of the enzymes listed in Table 1, or functional variants or fragments thereof.

The present invention features a method for producing a fucosylated oligosaccharide in a bacterium by providing bacterium that express at least one exogenous lactose-utilizing $\alpha(1,2)$ fucosyltransferase. The amino acid sequence of the exogenous lactose-utilizing $\alpha(1,2)$ fucosyltransferase is preferably at least 22% identical to *H. pylori* FutC or at least 25% identical to *B. vulgatus* FutN. In one aspect, the bacterium also expresses one or more exogenous lactose-utilizing $\alpha(1,3)$ fucosyltransferase enzymes and/or one or more exogenous lactose-utilizing $\alpha(1,4)$ fucosyltransferase enzymes. The combination of fucosyltransferases expressed in the production bacterium is dependent upon the desired fucosylated oligosaccharide product. The method disclosed herein further includes retrieving the fucosylated oligosaccharide from said bacterium or from a culture supernatant of said bacterium.

Examples of suitable $\alpha(1,3)$ fucosyltransferase enzymes include, but are not limited to *Helicobacter pylori* 26695 *futA* gene (GenBank Accession Number HV532291 (GI:365791177), incorporated herein by reference), *H. hepaticus* Hh0072, *H. pylori* 11639 FucT, and *H. pylori* UA948 FucTa (e.g., GenBank Accession Number AF194963 (GI:28436396), incorporated herein by reference)(Rasko, D. A., Wang, G., Palcic, M. M. & Taylor, D. E. J Biol Chem 275, 4988-4994 (2000)). Examples of suitable $\alpha(1,4)$ fucosyltransferase enzymes include, but are not limited to *H. pylori* UA948 FucTa (which has relaxed acceptor specificity and is able to generate both $\alpha(1,3)$ - and $\alpha(1,4)$ -fucosyl linkages). An example of an enzyme possessing only $\alpha(1,4)$ fucosyltransferase activity is given by the FucT III enzyme from *Helicobacter pylori* strain DMS6709 (e.g., GenBank Accession Number AY450598.1 (GI:40646733), incorporated herein by reference) (S. Rabbani, V. Miksa, B. Wipf, B. Ernst, *Glycobiology* **15**, 1076-83 (2005).)

The invention also features a nucleic acid construct or a vector comprising a nucleic acid encoding at least one α (1,2) fucosyltransferase or variant, or fragment thereof, as described herein. The vector can further include one or more regulatory elements, e.g., a heterologous promoter. By “heterologous” is meant that the control sequence and protein-encoding sequence originate from different bacterial strains. The regulatory elements can be operably linked to a gene encoding a protein, a gene construct encoding a fusion protein gene, or a series of genes linked in an operon in order to express the fusion protein. In yet another aspect, the invention comprises an isolated recombinant cell, e.g., a bacterial cell containing an aforementioned nucleic acid molecule or vector. The nucleic acid is optionally integrated into the genome of the host bacterium. In some embodiments, the nucleic acid construct also further comprises one or more α (1,3) fucosyltransferases and/or α (1,4) fucosyltransferases. Alternatively, the α (1,2) fucosyltransferase also exhibits α (1,3) fucosyltransferase and/or α (1,4) fucosyltransferase activity.

The bacterium utilized in the production methods described herein is genetically engineered to increase the efficiency and yield of fucosylated oligosaccharide products. For example, the host production bacterium is characterized as having a reduced level of β -galactosidase activity, a defective colanic acid synthesis pathway, an inactivated ATP-dependent intracellular protease, an inactivated *lacA*, or a combination thereof. In one embodiment, the bacterium is characterized as having a reduced level of β -galactosidase activity, a defective colanic acid synthesis pathway, an inactivated ATP-dependent intracellular protease, and an inactivated *lacA*.

As used herein, an “inactivated” or “inactivation of a” gene, encoded gene product (*i.e.*, polypeptide), or pathway refers to reducing or eliminating the expression (*i.e.*, transcription or translation), protein level (*i.e.*, translation, rate of degradation), or enzymatic activity of the gene, gene product, or pathway. In the instance where a pathway is inactivated, preferably one enzyme or polypeptide in the pathway exhibits reduced or negligible activity. For example, the enzyme in the pathway is altered, deleted or mutated such that the product of the pathway is produced at low levels compared to a wild-type bacterium or an intact pathway. Alternatively, the product of the pathway is not produced. Inactivation of a gene is achieved by deletion or mutation of the gene or regulatory elements of the gene such that the gene is no longer transcribed or translated. Inactivation of a polypeptide can be achieved by deletion or mutation of the gene that encodes the gene product or mutation of the polypeptide to disrupt its activity. Inactivating mutations include

additions, deletions or substitutions of one or more nucleotides or amino acids of a nucleic acid or amino acid sequence that results in the reduction or elimination of the expression or activity of the gene or polypeptide. In other embodiments, inactivation of a polypeptide is achieved through the addition of exogenous sequences (*i.e.*, tags) to the N or C-terminus of the polypeptide such that the activity of the polypeptide is reduced or eliminated (*i.e.*, by steric hindrance).

A host bacterium suitable for the production systems described herein exhibits an enhanced or increased cytoplasmic or intracellular pool of lactose and/or GDP-fucose. For example, the bacterium is *E. coli* and endogenous *E. coli* metabolic pathways and genes are manipulated in ways that result in the generation of increased cytoplasmic concentrations of lactose and/or GDP-fucose, as compared to levels found in wild type *E. coli*. Preferably, the bacterium accumulates an increased intracellular lactose pool and an increased intracellular GDP-fucose pool. For example, the bacteria contain at least 10%, 20%, 50%, or 2X, 5X, 10X or more of the levels of intracellular lactose and/or intracellular GDP-fucose compared to a corresponding wild type bacteria that lacks the genetic modifications described herein.

Increased intracellular concentration of lactose in the host bacterium compared to wild-type bacterium is achieved by manipulation of genes and pathways involved in lactose import, export and catabolism. In particular, described herein are methods of increasing intracellular lactose levels in *E. coli* genetically engineered to produce a human milk oligosaccharide by simultaneous deletion of the endogenous β -galactosidase gene (*lacZ*) and the lactose operon repressor gene (*lacI*). During construction of this deletion, the *lacIq* promoter is placed immediately upstream of (contiguous with) the lactose permease gene, *lacY*, *i.e.*, the sequence of the *lacIq* promoter is directly upstream and adjacent to the start of the sequence encoding the *lacY* gene, such that the *lacY* gene is under transcriptional regulation by the *lacIq* promoter. The modified strain maintains its ability to transport lactose from the culture medium (via LacY), but is deleted for the wild-type chromosomal copy of the *lacZ* (encoding β -galactosidase) gene responsible for lactose catabolism. Thus, an intracellular lactose pool is created when the modified strain is cultured in the presence of exogenous lactose.

Another method for increasing the intracellular concentration of lactose in *E. coli* involves inactivation of the *lacA* gene. A inactivating mutation, null mutation, or deletion of *lacA* prevents the formation of intracellular acetyl-lactose, which not only removes this molecule as a contaminant from subsequent purifications, but also eliminates *E. coli*'s ability

to export excess lactose from its cytoplasm (Danchin A. Cells need safety valves. *Bioessays* 2009, Jul;31(7):769-73.), thus greatly facilitating purposeful manipulations of the *E. coli* intracellular lactose pool.

The invention also provides methods for increasing intracellular levels of GDP-fucose in a bacterium by manipulating the organism's endogenous colanic acid biosynthesis pathway. This increase is achieved through a number of genetic modifications of endogenous *E. coli* genes involved either directly in colanic acid precursor biosynthesis, or in overall control of the colanic acid synthetic regulon. Particularly preferred is inactivation of the genes or encoded polypeptides that act in the colanic acid synthesis pathway after the production of GDP-fucose (the donor substrate) and before the generation of colanic acid. Exemplary colanic acid synthesis genes include, but are not limited to: a *wcaJ* gene, (e.g., GenBank Accession Number (amino acid) BAA15900 (GI:1736749), incorporated herein by reference), a *wcaA* gene (e.g., GenBank Accession Number (amino acid) BAA15912.1 (GI:1736762), incorporated herein by reference), a *wcaC* gene (e.g., GenBank Accession Number (amino acid) BAE76574.1 (GI:85675203), incorporated herein by reference), a *wcaE* gene (e.g., GenBank Accession Number (amino acid) BAE76572.1 (GI:85675201), incorporated herein by reference), a *wcaI* gene (e.g., GenBank Accession Number (amino acid) BAA15906.1 (GI:1736756), incorporated herein by reference), a *wcaL* gene (e.g., GenBank Accession Number (amino acid) BAA15898.1 (GI:1736747), incorporated herein by reference), a *wcaB* gene (e.g., GenBank Accession Number (amino acid) BAA15911.1 (GI:1736761), incorporated herein by reference), a *wcaF* gene (e.g., GenBank Accession Number (amino acid) BAA15910.1 (GI:1736760), incorporated herein by reference), a *wzxE* gene (e.g., GenBank Accession Number (amino acid) BAE77506.1 (GI:85676256), incorporated herein by reference), a *wzxC* gene, (e.g., GenBank Accession Number (amino acid) BAA15899 (GI:1736748), incorporated herein by reference), a *wcaD* gene, (e.g., GenBank Accession Number (amino acid) BAE76573 (GI:85675202), incorporated herein by reference), a *wza* gene (e.g., GenBank Accession Number (amino acid) BAE76576 (GI:85675205), incorporated herein by reference), a *wzb* gene (e.g., GenBank Accession Number (amino acid) BAE76575 (GI:85675204), incorporated herein by reference), and a *wzc* gene (e.g., GenBank Accession Number (amino acid) BAA15913 (GI:1736763), incorporated herein by reference).

Preferably, a host bacterium, such as *E. coli*, is genetically engineered to produce a human milk oligosaccharide by the inactivation of the *wcaJ* gene, which encoding the UDP-glucose lipid carrier transferase. The inactivation of the *wcaJ* gene can be by deletion of the

gene, a null mutation, or inactivating mutation of the *wcaJ* gene, such that the activity of the encoded *wcaJ* is reduced or eliminated compared to wild-type *E. coli*. In a *wcaJ* null background, GDP-fucose accumulates in the *E. coli* cytoplasm.

Over-expression of a positive regulator protein, RcsA (e.g., GenBank Accession Number M58003 (GI:1103316), incorporated herein by reference), in the colanic acid synthesis pathway results in an increase in intracellular GDP-fucose levels. Over-expression of an additional positive regulator of colanic acid biosynthesis, namely RcsB (e.g., GenBank Accession Number E04821 (GI:2173017), incorporated herein by reference), is also utilized, either instead of or in addition to over-expression of RcsA, to increase intracellular GDP-fucose levels.

Alternatively, colanic acid biosynthesis is increased following the introduction of a mutation into the *E. coli lon* gene (e.g., GenBank Accession Number L20572 (GI:304907), incorporated herein by reference). Lon is an adenosine-5'-triphosphate (ATP)-dependant intracellular protease that is responsible for degrading RcsA, mentioned above as a positive transcriptional regulator of colanic acid biosynthesis in *E. coli*. In a *lon* null background, RcsA is stabilized, RcsA levels increase, the genes responsible for GDP-fucose synthesis in *E. coli* are up-regulated, and intracellular GDP-fucose concentrations are enhanced. Mutations in *lon* suitable for use with the methods presented herein include null mutations or insertions that disrupt the expression or function of *lon*.

A functional lactose permease gene is also present in the bacterium. The lactose permease gene is an endogenous lactose permease gene or an exogenous lactose permease gene. For example, the lactose permease gene comprises an *E. coli lacY* gene (e.g., GenBank Accession Number V00295 (GI:41897), incorporated herein by reference). Many bacteria possess the inherent ability to transport lactose from the growth medium into the cell, by utilizing a transport protein that is either a homolog of the *E. coli* lactose permease (e.g., as found in *Bacillus licheniformis*), or a transporter that is a member of the ubiquitous PTS sugar transport family (e.g., as found in *Lactobacillus casei* and *Lactobacillus rhamnosus*). For bacteria lacking an inherent ability to transport extracellular lactose into the cell cytoplasm, this ability is conferred by an exogenous lactose transporter gene (e.g., *E. coli lacY*) provided on recombinant DNA constructs, and supplied either on a plasmid expression vector or as exogenous genes integrated into the host chromosome.

As described herein, in some embodiments, the host bacterium preferably has a reduced level of β -galactosidase activity. In the embodiment in which the bacterium is

characterized by the deletion of the endogenous β -galactosidase gene, an exogenous β -galactosidase gene is introduced to the bacterium. For example, a plasmid expressing an exogenous β -galactosidase gene is introduced to the bacterium, or recombined or integrated into the host genome. For example, the exogenous β -galactosidase gene is inserted into a gene that is inactivated in the host bacterium, such as the *lon* gene.

The exogenous β -galactosidase gene is a functional β -galactosidase gene characterized by a reduced or low level of β -galactosidase activity compared to β -galactosidase activity in wild-type bacteria lacking any genetic manipulation. Exemplary β -galactosidase genes include *E. coli lacZ* and β -galactosidase genes from any of a number of other organisms (e.g., the *lac4* gene of *Kluyveromyces fragilis* (e.g., GenBank Accession Number M84410 (GI:173304), incorporated herein by reference) that catalyzes the hydrolysis of β -galactosides into monosaccharides. The level of β -galactosidase activity in wild-type *E. coli* bacteria is, for example, 6,000 units. Thus, the reduced β -galactosidase activity level encompassed by engineered host bacterium of the present invention includes less than 6,000 units, less than 5,000 units, less than 4,000 units, less than 3,000 units, less than 2,000 units, less than 1,000 units, less than 900 units, less than 800 units, less than 700 units, less than 600 units, less than 500 units, less than 400 units, less than 300 units, less than 200 units, less than 100 units, or less than 50 units. Low, functional levels of β -galactosidase include β -galactosidase activity levels of between 0.05 and 1,000 units, e.g., between 0.05 and 750 units, between 0.05 and 500 units, between 0.05 and 400 units, between 0.05 and 300 units, between 0.05 and 200 units, between 0.05 and 100 units, between 0.05 and 50 units, between 0.05 and 10 units, between 0.05 and 5 units, between 0.05 and 4 units, between 0.05 and 3 units, or between 0.05 and 2 units of β -galactosidase activity. For unit definition and assays for determining β -galactosidase activity, see Miller JH, Laboratory CSH. Experiments in molecular genetics. Cold Spring Harbor Laboratory Cold Spring Harbor, NY; 1972; (incorporated herein by reference). This low level of cytoplasmic β -galactosidase activity is not high enough to significantly diminish the intracellular lactose pool. The low level of β -galactosidase activity is very useful for the facile removal of undesired residual lactose at the end of fermentations.

Optionally, the bacterium has an inactivated *thyA* gene. Preferably, a mutation in a *thyA* gene in the host bacterium allows for the maintenance of plasmids that carry *thyA* as a selectable marker gene. Exemplary alternative selectable markers include antibiotic resistance genes such as BLA (beta-lactamase), or *proBA* genes (to complement a *proAB*

host strain proline auxotrophy) or *purA* (to complement a *purA* host strain adenine auxotrophy).

In one aspect, the *E. coli* bacterium comprises the genotype $\Delta ampC::P_{trp}^B cI$, $\Delta(lacI-lacZ)::FRT$, $P_{lacI} lacY^+$, $\Delta wcaJ::FRT$, $thyA::Tn10$, $\Delta lon:(npt3, lacZ^+)$, $\Delta lacA$, and also comprises any one of the exogenous $\alpha(1,2)$ fucosyltransferases described herein.

The bacterium comprising these characteristics is cultured in the presence of lactose. In some cases, the method further comprises culturing the bacterium in the presence of tryptophan and in the absence of thymidine. The fucosylated oligosaccharide is retrieved from the bacterium (*i.e.*, a cell lysate) or from a culture supernatant of the bacterium.

The invention provides a purified fucosylated oligosaccharide produced by the methods described herein. The fucosylated oligosaccharide is purified for use in therapeutic or nutritional products, or the bacterium is used directly in such products. The fucosylated oligosaccharide produced by the engineered bacterium is 2'-fucosyllactose (2'-FL) or lactodifucotetraose (LDFT). The new $\alpha(1,2)$ -fucosyltransferases are also useful to synthesize HMOS of larger molecular weight bearing $\alpha(1,2)$ fucose moieties, *e.g.*, lacto-N-fucopentaose (LNF I) and lacto-N-difucohexaose (LDFH I). For example, to produce LDFT, the host bacterium is engineered to express an exogenous $\alpha(1,2)$ fucosyltransferase that also possesses $\alpha(1,3)$ fucosyltransferase activity, or an exogenous $\alpha(1,2)$ fucosyltransferase and an exogenous $\alpha(1,3)$ fucosyltransferase. For the production of LNF I and LDFH I, the host bacterium is engineered to express an exogenous $\alpha(1,2)$ fucosyltransferase that also possesses $\alpha(1,3)$ fucosyltransferase activity and/or $\alpha(1,4)$ fucosyltransferase activity, or an exogenous $\alpha(1,2)$ fucosyltransferase, an exogenous $\alpha(1,3)$ fucosyltransferase, and an exogenous $\alpha(1,4)$ fucosyltransferase.

A purified fucosylated oligosaccharide produced by the methods described above is also within the invention. The purified oligosaccharide (2'-FL) obtained at the end of the process is a white/slightly off-white, crystalline, sweet powder. For example, an engineered bacterium, bacterial culture supernatant, or bacterial cell lysate according to the invention comprises 2'-FL, LDFT, LNF I or LDFH I produced by the methods described herein, and does not substantially comprise a other fucosylated oligosaccharides prior to purification of the fucosylated oligosaccharide products from the cell, culture supernatant, or lysate. As a general matter, the fucosylated oligosaccharide produced by the methods contains a negligible amount of 3-FL in a 2'-FL-containing cell, cell lysate or culture, or supernatant, *e.g.*, less than 1% of the level of 2'-FL or 0.5% of the level of 2'-FL. Moreover, the

fucosylated oligosaccharide produced by the methods described herein also have a minimal amount of contaminating lactose, which can often be co-purified with the fucosylated oligosaccharide product, such as 2'-FL. This reduction in contaminating lactose results from the reduced level of β -galactosidase activity present in the engineered host bacterium.

A purified oligosaccharide, e.g., 2'-FL, LDFT, LNF I, or LDFH I, is one that is at least 90%, 95%, 98%, 99%, or 100% (w/w) of the desired oligosaccharide by weight. Purity is assessed by any known method, e.g., thin layer chromatography or other chromatographic techniques known in the art. The invention includes a method of purifying a fucosylated oligosaccharide produced by the genetically engineered bacterium described above, which method comprises separating the desired fucosylated oligosaccharide (e.g., 2'-FL) from contaminants in a bacterial cell lysate or bacterial cell culture supernatant of the bacterium.

The oligosaccharides are purified and used in a number of products for consumption by humans as well as animals, such as companion animals (dogs, cats) as well as livestock (bovine, equine, ovine, caprine, or porcine animals, as well as poultry). For example, a pharmaceutical composition comprises purified 2'-FL and a pharmaceutically-acceptable excipient that is suitable for oral administration. Large quantities of 2'-FL are produced in bacterial hosts, e.g., an *E. coli* bacterium comprising an exogenous α (1,2) fucosyltransferase gene.

A method of producing a pharmaceutical composition comprising a purified human milk oligosaccharide (HMOS) is carried out by culturing the bacterium described above, purifying the HMOS produced by the bacterium, and combining the HMOS with an excipient or carrier to yield a dietary supplement for oral administration. These compositions are useful in methods of preventing or treating enteric and/or respiratory diseases in infants and adults. Accordingly, the compositions are administered to a subject suffering from or at risk of developing such a disease.

The invention also provides methods of identifying an α (1,2) fucosyltransferase gene capable of synthesizing fucosylated oligosaccharides in a host bacterium, *i.e.*, 2'-fucosyllactose (2'-FL) in *E. coli*. The method of identifying novel lactose-utilizing, α (1,2)fucosyltransferase enzyme comprises the following steps:

- 1) performing a computational search of sequence databases to define a broad group of simple sequence homologs of any known, lactose-utilizing α (1,2)fucosyltransferase;
- 2) using the list from step (1), deriving a search profile containing common sequence and/or structural motifs shared by the members of the list;

3) searching sequence databases, using a derived search profile based on the common sequence or structural motif from step (2) as query, and identifying a candidate sequences, wherein a sequence homology to a reference lactose-utilizing $\alpha(1,2)$ fucosyltransferase is a predetermined percentage threshold;

4) compiling a list of candidate organisms, said organisms being characterized as expressing $\alpha(1,2)$ fucosyl- glycans in a naturally-occurring state;

5) selecting candidate sequences that are derived from candidate organisms to generate a list of candidate lactose-utilizing enzymes;

6) expressing the candidate lactose- utilizing enzyme in a host organism; and

7) testing for lactose- utilizing $\alpha(1,2)$ fucosyltransferase activity, wherein detection of the desired fucosylated oligosaccharide product in said organism indicates that the candidate sequence comprises a novel lactose- utilizing $\alpha(1,2)$ fucosyltransferase. In another embodiment, the search profile is generated from a multiple sequence alignment of the amino acid sequences of more than one enzyme with known $\alpha(1,2)$ fucosyltransferase activity. The database search can then be designed to refine and iteratively search for novel $\alpha(1,2)$ fucosyltransferases with significant sequence similarity to the multiple sequence alignment query.

The invention provides a method of treating, preventing, or reducing the risk of infection in a subject comprising administering to said subject a composition comprising a purified recombinant human milk oligosaccharide, wherein the HMOS binds to a pathogen and wherein the subject is infected with or at risk of infection with the pathogen. In one aspect, the infection is caused by a Norwalk-like virus or *Campylobacter jejuni*. The subject is preferably a mammal in need of such treatment. The mammal is, e.g., any mammal, e.g., a human, a primate, a mouse, a rat, a dog, a cat, a cow, a horse, or a pig. In a preferred embodiment, the mammal is a human. For example, the compositions are formulated into animal feed (e.g., pellets, kibble, mash) or animal food supplements for companion animals, e.g., dogs or cats, as well as livestock or animals grown for food consumption, e.g., cattle, sheep, pigs, chickens, and goats. Preferably, the purified HMOS is formulated into a powder (e.g., infant formula powder or adult nutritional supplement powder, each of which is mixed with a liquid such as water or juice prior to consumption) or in the form of tablets, capsules or pastes or is incorporated as a component in dairy products such as milk, cream, cheese, yogurt or kefir, or as a component in any beverage, or combined in a preparation containing live microbial cultures intended to serve as probiotics, or in prebiotic preparations to enhance the growth of beneficial microorganisms either *in vitro* or *in vivo*.

Polynucleotides, polypeptides, and oligosaccharides of the invention are purified and/or isolated. Purified defines a degree of sterility that is safe for administration to a human subject, e.g., lacking infectious or toxic agents. Specifically, as used herein, an "isolated" or "purified" nucleic acid molecule, polynucleotide, polypeptide, protein or oligosaccharide, is substantially free of other cellular material, or culture medium when produced by recombinant techniques, or chemical precursors or other chemicals when chemically synthesized. For example, purified HMOS compositions are at least 60% by weight (dry weight) the compound of interest. Preferably, the preparation is at least 75%, more preferably at least 90%, and most preferably at least 99%, by weight the compound of interest. Purity is measured by any appropriate standard method, for example, by column chromatography, thin layer chromatography, or high-performance liquid chromatography (HPLC) analysis. For example, a "purified protein" refers to a protein that has been separated from other proteins, lipids, and nucleic acids with which it is naturally associated. Preferably, the protein constitutes at least 10, 20, 50, 70, 80, 90, 95, 99-100% by dry weight of the purified preparation.

Similarly, by "substantially pure" is meant an oligosaccharide that has been separated from the components that naturally accompany it. Typically, the oligosaccharide is substantially pure when it is at least 60%, 70%, 80%, 90%, 95%, or even 99%, by weight, free from the proteins and naturally-occurring organic molecules with which it is naturally associated.

By "isolated nucleic acid" is meant a nucleic acid that is free of the genes which, in the naturally-occurring genome of the organism from which the DNA of the invention is derived, flank the gene. The term covers, for example: (a) a DNA which is part of a naturally occurring genomic DNA molecule, but is not flanked by both of the nucleic acid sequences that flank that part of the molecule in the genome of the organism in which it naturally occurs; (b) a nucleic acid incorporated into a vector or into the genomic DNA of a prokaryote or eukaryote in a manner, such that the resulting molecule is not identical to any naturally occurring vector or genomic DNA; (c) a separate molecule such as a cDNA, a genomic fragment, a fragment produced by polymerase chain reaction (PCR), or a restriction fragment; and (d) a recombinant nucleotide sequence that is part of a hybrid gene, i.e., a gene encoding a fusion protein. Isolated nucleic acid molecules according to the present invention further include molecules produced synthetically, as well as any nucleic acids that have been altered chemically and/or that have modified backbones.

A "heterologous promoter" is a promoter which is different from the promoter to which a gene or nucleic acid sequence is operably linked in nature.

The term "overexpress" or "overexpression" refers to a situation in which more factor is expressed by a genetically-altered cell than would be, under the same conditions, by a wild type cell. Similarly, if an unaltered cell does not express a factor that it is genetically altered to produce, the term "express" (as distinguished from "overexpress") is used indicating the wild type cell did not express the factor at all prior to genetic manipulation.

The terms "treating" and "treatment" as used herein refer to the administration of an agent or formulation to a clinically symptomatic individual afflicted with an adverse condition, disorder, or disease, so as to effect a reduction in severity and/or frequency of symptoms, eliminate the symptoms and/or their underlying cause, and/or facilitate improvement or remediation of damage. The terms "preventing" and "prevention" refer to the administration of an agent or composition to a clinically asymptomatic individual who is susceptible to a particular adverse condition, disorder, or disease, and thus relates to the prevention of the occurrence of symptoms and/or their underlying cause.

By the terms "effective amount" and "therapeutically effective amount" of a formulation or formulation component is meant a nontoxic but sufficient amount of the formulation or component to provide the desired effect.

The transitional term "comprising," which is synonymous with "including," "containing," or "characterized by," is inclusive or open-ended and does not exclude additional, unrecited elements or method steps. By contrast, the transitional phrase "consisting of" excludes any element, step, or ingredient not specified in the claim. The transitional phrase "consisting essentially of" limits the scope of a claim to the specified materials or steps "and those that do not materially affect the basic and novel characteristic(s)" of the claimed invention.

The host organism used to express the lactose-accepting fucosyltransferase gene is typically the enterobacterium *Escherichia coli* K12 (*E. coli*). *E. coli* K-12 is not considered a human or animal pathogen nor is it toxicogenic. *E. coli* K-12 is a standard production strain of bacteria and is noted for its safety due to its poor ability to colonize the colon and establish infections (see, e.g., epa.gov/oppt/biotech/pubs/fra/fra004.htm). However, a variety of bacterial species may be used in the oligosaccharide biosynthesis methods, e.g., *Erwinia herbicola* (*Pantoea agglomerans*), *Citrobacter freundii*, *Pantoea citrea*, *Pectobacterium carotovorum*, or *Xanthomonas campestris*. Bacteria of the genus *Bacillus* may also be used, including *Bacillus subtilis*, *Bacillus licheniformis*, *Bacillus coagulans*, *Bacillus thermophilus*,

Bacillus laterosporus, *Bacillus megaterium*, *Bacillus mycoides*, *Bacillus pumilus*, *Bacillus lentus*, *Bacillus cereus*, and *Bacillus circulans*. Similarly, bacteria of the genera *Lactobacillus* and *Lactococcus* may be modified using the methods of this invention, including but not limited to *Lactobacillus acidophilus*, *Lactobacillus salivarius*, *Lactobacillus plantarum*, *Lactobacillus helveticus*, *Lactobacillus delbrueckii*, *Lactobacillus rhamnosus*, *Lactobacillus bulgaricus*, *Lactobacillus crispatus*, *Lactobacillus gasseri*, *Lactobacillus casei*, *Lactobacillus reuteri*, *Lactobacillus jensenii*, and *Lactococcus lactis*. *Streptococcus thermophiles* and *Propionibacterium freudenreichii* are also suitable bacterial species for the invention described herein. Also included as part of this invention are strains, modified as described here, from the genera *Enterococcus* (e.g., *Enterococcus faecium* and *Enterococcus thermophiles*), *Bifidobacterium* (e.g., *Bifidobacterium longum*, *Bifidobacterium infantis*, and *Bifidobacterium bifidum*), *Sporolactobacillus* spp., *Micromonospora* spp., *Micrococcus* spp., *Rhodococcus* spp., and *Pseudomonas* (e.g., *Pseudomonas fluorescens* and *Pseudomonas aeruginosa*). Bacteria comprising the characteristics described herein are cultured in the presence of lactose, and a fucosylated oligosaccharide is retrieved, either from the bacterium itself or from a culture supernatant of the bacterium. The fucosylated oligosaccharide is purified for use in therapeutic or nutritional products, or the bacteria are used directly in such products. A suitable production host bacterial strain is one that is not the same bacterial strain as the source bacterial strain from which the fucosyltransferase-encoding nucleic acid sequence was identified.

Other features and advantages of the invention will be apparent from the following description of the preferred embodiments thereof, and from the claims. Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. Although methods and materials similar or equivalent to those described herein can be used in the practice or testing of the present invention, suitable methods and materials are described below. All published foreign patents and patent applications cited herein are incorporated herein by reference. Genbank and NCBI submissions indicated by accession number cited herein are incorporated herein by reference. All other published references, documents, manuscripts and scientific literature cited herein are incorporated herein by reference. In the case of conflict, the present specification, including definitions, will control. In addition, the materials, methods, and examples are illustrative only and not intended to be limiting.

BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1 is a schematic illustration showing the synthetic pathway of the major neutral fucosyl-oligosaccharides found in human milk.

FIG. 2 is a schematic demonstrating metabolic pathways and the changes introduced into them to engineer 2'-fucosyllactose (2'-FL) synthesis in *Escherichia coli* (*E. coli*). Specifically, the lactose synthesis pathway and the GDP-fucose synthesis pathway are illustrated. In the GDP-fucose synthesis pathway: *manA* = phosphomannose isomerase (PMI), *manB* = phosphomannomutase (PMM), *manC* = mannose-1-phosphate guanylyltransferase (GMP), *gmd* = GDP-mannose-4,6-dehydratase, *fcl* = GDP-fucose synthase (GFS), and $\Delta wcaJ$ = mutated UDP-glucose lipid carrier transferase.

FIG. 3A and FIG. 3B show the sequence identity and a multiple sequence alignment of 4 previously known lactose-utilizing $\alpha(1,2)$ -fucosyltransferase protein sequences. FIG. 3A is a table showing the sequence identity between the 4 known lactose-utilizing $\alpha(1,2)$ -fucosyltransferases: *H. pylori* futC (SEQ ID NO: 1), *H. mustelae* FutL (SEQ ID NO: 2), *Bacteroides vulgatus* futN (SEQ ID NO: 3), and *E. coli* 0126 wbgL (SEQ ID NO: 4). FIG. 3B shows multiple sequence alignment of the 4 known $\alpha(1,2)$ -fucosyltransferases. The ovals highlight regions of particularly high sequence conservation between the four enzymes in the alignment.

FIG. 4 shows the sequence alignment of the 12 identified $\alpha(1,2)$ -fucosyltransferase syngenes identified, along with the 4 previously known lactose-utilizing $\alpha(1,2)$ -fucosyltransferase protein sequences. The 4 known lactose-utilizing $\alpha(1,2)$ -fucosyltransferases are boxed and include *H. pylori* futC (SEQ ID NO: 1), *H. mustelae* FutL (SEQ ID NO: 2), *Bacteroides vulgatus* futN (SEQ ID NO: 3), and *E. coli* 0126 wbgL (SEQ ID NO: 4). The 12 identified $\alpha(1,2)$ -fucosyltransferase are as follows: *Prevotella melaninogenica* FutO (SEQ ID NO: 10), *Clostridium bolteae* +13 FutP (SEQ ID NO: 292), *Lachnospiraceae* sp. FutQ (SEQ ID NO: 12), *Methanosphaerula palustris* FutR (SEQ ID NO: 13), *Tannerella* sp. FutS (SEQ ID NO: 14), *Bacteroides caccae* FutU (SEQ ID NO: 15), *Butyrivibrio* FutV (SEQ ID NO: 16), *Prevotella* sp. FutW (SEQ ID NO: 17), *Parabacteroides johnsonii* FutX (SEQ ID NO: 18), *Akkermansia muciniphilia* FutY (SEQ ID NO: 19), *Salmonella enterica* FutZ (SEQ ID NO: 20), *Bacteroides* sp. FutZA (SEQ ID NO: 21). The sequence for *Clostridium bolteae* FutP (without the 13 additional amino acids in the N-terminus) (SEQ ID NO: 11) is also shown in the alignment.

FIG. 5A and FIG. 5B are two pictures of gels showing the construction of the syngenes for each of the 12 novel $\alpha(1,2)$ -fucosyltransferases. FIG. 5A shows post-Gibson assembly PCR. FIG. 5B shows gel-purified RI/XhoI syngene fragments.

FIG. 6A and FIG. 6B are two photographs showing thin layer chromatograms of fucosylated oligosaccharide products produced in *E. coli* cultures using the 12 novel $\alpha(1,2)$ -fucosyltransferase syngenes. FIG. 6A shows fucosylated oligosaccharide products from 2 μ l of culture supernatant. FIG. 6B shows fucosylated oligosaccharide products from 0.2 OD₆₀₀ cell equivalents of whole cell heat extracts.

FIG. 7 is a graph showing the growth curve of the host bacterium expressing plasmids containing the $\alpha(1,2)$ fucosyltransferase genes *WbgL*, *FutN*, *FutO*, *FutQ*, and *FutX* after tryptophan induction in the presence of lactose in the culture medium (i.e. lac + trp).

FIG. 8 is a photograph of a SDS-PAGE gel showing the proteins produced from host bacterium expressing $\alpha(1,2)$ fucosyltransferase genes *WbgL*, *FutN*, *FutO*, *FutQ*, and *FutX* after induction.

FIG. 9A and FIG. 9B are two photographs of thin layer chromatograms showing the production of fucosylated oligosaccharide products from in *E. coli* cultures expressing select $\alpha(1,2)$ -fucosyltransferase syngenes *WbgL*, *FutN*, *FutO*, *FutQ*, and *FutX* at 7 hours or 24 hours after induction. FIG. 9A shows fucosylated oligosaccharide products from 2 μ l of culture supernatant. FIG. 9B shows fucosylated oligosaccharide products from 0.2 OD₆₀₀ cell equivalents of whole cell heat extracts.

FIG. 10A and FIG. 10B are two photographs of thin layer chromatograms showing the fucosylated oligosaccharide products after two different 1.5L fermentation runs from *E. coli* expressing *FutN*: FIG. 10A) 36B and FIG. 10B) 37A. The culture yield for run 36B was 33g/L while the yield for run 37A was 36.3 g/L.

FIG. 11 is a plasmid map of pG217 carrying the *B. vulgatus FutN* gene.

FIG. 12 is a schematic diagram showing the insertion of the *LacIq* promoter, the functional *lacY* gene, and the deletion of *lacA*.

FIG. 13 is a schematic diagram showing the deletion of the endogenous *wcaJ* gene using FRT recombination.

FIG. 14 is a schematic diagram of the *E. coli W3110* chromosome, showing the insertion of a DNA fragment carrying kanamycin resistance gene (derived from transposon Tn5) and wild-type *lacZ* into the *lon* gene.

DETAILED DESCRIPTION OF THE INVENTION

While some studies suggest that human milk glycans could be used as antimicrobial anti-adhesion agents, the difficulty and expense of producing adequate quantities of these agents of a quality suitable for human consumption has limited their full-scale testing and perceived utility. What has been needed is a suitable method for producing the appropriate glycans in sufficient quantities at reasonable cost. Prior to the invention described herein, there were attempts to use several distinct synthetic approaches for glycan synthesis. Some chemical approaches can synthesize oligosaccharides (Flowers, H. M. *Methods Enzymol* 50, 93-121 (1978); Seeberger, P. H. *Chem Commun (Camb)* 1115-1121 (2003)), but reactants for these methods are expensive and potentially toxic (Koeller, K. M. & Wong, C. H. *Chem Rev* 100, 4465-4494 (2000)). Enzymes expressed from engineered organisms (Albermann, C., Piepersberg, W. & Wehmeier, U. F. *Carbohydr Res* 334, 97-103 (2001); Bettler, E., Samain, E., Chazalet, V., Bosso, C., et al. *Glycoconj J* 16, 205-212 (1999); Johnson, K. F. *Glycoconj J* 16, 141-146 (1999); Palcic, M. M. *Curr Opin Biotechnol* 10, 616-624 (1999); Wymer, N. & Toone, E. J. *Curr Opin Chem Biol* 4, 110-119 (2000)) provide a precise and efficient synthesis (Palcic, M. M. *Curr Opin Biotechnol* 10, 616-624 (1999)); Crout, D. H. & Vic, G. *Curr Opin Chem Biol* 2, 98-111 (1998)), but the high cost of the reactants, especially the sugar nucleotides, limits their utility for low-cost, large-scale production. Microbes have been genetically engineered to express the glycosyltransferases needed to synthesize oligosaccharides from the bacteria's innate pool of nucleotide sugars (Endo, T., Koizumi, S., Tabata, K., Kakita, S. & Ozaki, A. *Carbohydr Res* 330, 439-443 (2001); Endo, T., Koizumi, S., Tabata, K. & Ozaki, A. *Appl Microbiol Biotechnol* 53, 257-261 (2000); Endo, T. & Koizumi, S. *Curr Opin Struct Biol* 10, 536-541 (2000); Endo, T., Koizumi, S., Tabata, K., Kakita, S. & Ozaki, A. *Carbohydr Res* 316, 179-183 (1999); Koizumi, S., Endo, T., Tabata, K. & Ozaki, A. *Nat Biotechnol* 16, 847-850 (1998)). However, prior to the invention described herein, there was a growing need to identify and characterize additional glycosyltransferases that are useful for the synthesis of HMOS in metabolically engineered bacterial hosts.

Human Milk Glycans

Human milk contains a diverse and abundant set of neutral and acidic oligosaccharides (Kunz, C., Rudloff, S., Baier, W., Klein, N., and Strobel, S. (2000). *Annu Rev Nutr* 20, 699-722; Bode, L. (2006). *J Nutr* 136, 2127-130). More than 130 different complex oligosaccharides have been identified in human milk, and their structural diversity and abundance is unique to humans. Although these molecules may not be utilized directly

by infants for nutrition, they nevertheless serve critical roles in the establishment of a healthy gut microbiome (Marcobal, A., Barboza, M., Froehlich, J. W., Block, D. E., et al. *J Agric Food Chem* 58, 5334-5340 (2010)), in the prevention of disease (Newburg, D. S., Ruiz-Palacios, G. M. & Morrow, A. L. *Annu Rev Nutr* 25, 37-58 (2005)), and in immune function (Newburg, D. S. & Walker, W. A. *Pediatr Res* 61, 2-8 (2007)). Despite millions of years of exposure to human milk oligosaccharides (HMOS), pathogens have yet to develop ways to circumvent the ability of HMOS to prevent adhesion to target cells and to inhibit infection. The ability to utilize HMOS as pathogen adherence inhibitors promises to address the current crisis of burgeoning antibiotic resistance. Human milk oligosaccharides produced by biosynthesis represent the lead compounds of a novel class of therapeutics against some of the most intractable scourges of society.

One alternative strategy for efficient, industrial-scale synthesis of HMOS is the metabolic engineering of bacteria. This approach involves the construction of microbial strains overexpressing heterologous glycosyltransferases, membrane transporters for the import of precursor sugars into the bacterial cytosol, and possessing enhanced pools of regenerating nucleotide sugars for use as biosynthetic precursors (Dumon, C., Samain, E., and Priem, B. (2004). *Biotechnol Prog* 20, 412-19; Ruffing, A., and Chen, R.R. (2006). *Microb Cell Fact* 5, 25). A key aspect of this approach is the heterologous glycosyltransferase selected for overexpression in the microbial host. The choice of glycosyltransferase can significantly affect the final yield of the desired synthesized oligosaccharide, given that enzymes can vary greatly in terms of kinetics, substrate specificity, affinity for donor and acceptor molecules, stability and solubility. A few glycosyltransferases derived from different bacterial species have been identified and characterized in terms of their ability to catalyze the biosynthesis of HMOS in *E. coli* host strains (Dumon, C., Bosso, C., Utile, J.P., Heyraud, A., and Samain, E. (2006). *Chembiochem* 7, 359-365; Dumon, C., Samain, E., and Priem, B. (2004). *Biotechnol Prog* 20, 412-19; Li, M., Liu, X.W., Shao, J., Shen, J., Jia, Q., Yi, W., Song, J.K., Woodward, R., Chow, C.S., and Wang, P.G. (2008). *Biochemistry* 47, 378-387). The identification of additional glycosyltransferases with faster kinetics, greater affinity for nucleotide sugar donors and/or acceptor molecules, or greater stability within the bacterial host significantly improves the yields of therapeutically useful HMOS. Prior to the invention described herein, chemical syntheses of HMOS were possible, but were limited by stereo-specificity issues, precursor availability, product impurities, and high overall cost (Flowers, H. M. *Methods Enzymol* 50, 93-121 (1978); Seeberger, P. H. *Chem Commun (Camb)* 1115-1121 (2003);

Koeller, K. M. & Wong, C. H. Chem Rev 100, 4465-4494 (2000)). The invention overcomes the shortcomings of these previous attempts by providing new strategies to inexpensively manufacture large quantities of human milk oligosaccharides (HMOS) for use as dietary supplements. Advantages include efficient expression of the enzyme, improved stability and/or solubility of the fucosylated oligosaccharide product (2'-FL, LDFT, LNF I, and LDFH I) and reduced toxicity to the host organism. The present invention features novel $\alpha(1,2)$ FTs suitable for expression in production strains for increased efficacy and yield of fucosylated HMOS compared to $\alpha(1,2)$ FTs currently utilized in the field.

As described in detail below, *E. coli* (or other bacteria) is engineered to produce selected fucosylated oligosaccharides (*i.e.*, 2'-FL, LDFT, LDHF I, or LNF I) in commercially viable levels. For example, yields are >5 grams/liter in a bacterial fermentation process. In other embodiments, the yields are greater than 10 grams/liter, greater than 15 grams/liter, greater than 20 grams/liter, greater than 25 grams/liter, greater than 30 grams/liter, greater than 35 grams/liter, greater than 40 grams/liter, greater than 45 grams/liter, greater than 50 grams/liter, greater than 55 grams/liter, greater than 60 grams/liter, greater than 65 grams/liter, greater than 70 grams/liter, or greater than 75 grams/liter of fucosylated oligosaccharide products, such as 2'-FL, LDFT, LDHF I, and LNF I.

Role of Human milk glycans in infectious disease

Human milk glycans, which comprise both unbound oligosaccharides and their glycoconjugates, play a significant role in the protection and development of the infant gastrointestinal (GI) tract. Neutral fucosylated oligosaccharides, including 2'-fucosyllactose (2'-FL), protect infants against several important pathogens. Milk oligosaccharides found in various mammals differ greatly, and the composition in humans is unique (Hamosh M., 2001 *Pediatr Clin North Am*, 48:69-86; Newburg D.S., 2001 *Adv Exp Med Biol*, 501:3-10). Moreover, glycan levels in human milk change throughout lactation and also vary widely among individuals (Morrow A.L. et al., 2004 *J Pediatr*, 145:297-303; Chaturvedi P et al., 2001 *Glycobiology*, 11:365-372). Approximately 200 distinct human milk oligosaccharides have been identified and combinations of simple epitopes are responsible for this diversity (Newburg D.S., 1999 *Curr Med Chem*, 6:117-127; Ninonuevo M. et al., 2006 *J Agric Food Chem*, 54:7471-7480).

Human milk oligosaccharides are composed of 5 monosaccharides: D-glucose (Glc), D-galactose (Gal), N-acetylglucosamine (GlcNAc), L-fucose (Fuc), and sialic acid (N-acetylneuraminic acid, Neu5Ac, NANA). Human milk oligosaccharides are usually divided into

two groups according to their chemical structures: neutral compounds containing Glc, Gal, GlcNAc, and Fuc, linked to a lactose (Gal β 1-4Glc) core, and acidic compounds including the same sugars, and often the same core structures, plus NANA (Charlwood J. et al., 1999 Anal Biochem, 273:261-277; Martín-Sosa et al., 2003 J Dairy Sci, 86:52-59; Parkkinen J. and Finne J., 1987 Methods Enzymol, 138:289-300; Shen Z. et al., 2001 J Chromatogr A, 921:315-321).

Approximately 70-80% of oligosaccharides in human milk are fucosylated, and their synthetic pathways are believed to proceed as shown in FIG. 1. A smaller proportion of the oligosaccharides are sialylated or both fucosylated and sialylated, but their synthetic pathways are not fully defined. Understanding of the acidic (sialylated) oligosaccharides is limited in part by the ability to measure these compounds. Sensitive and reproducible methods for the analysis of both neutral and acidic oligosaccharides have been designed. Human milk oligosaccharides as a class survive transit through the intestine of infants very efficiently, being essentially indigestible (Chaturvedi, P., Warren, C. D., Buescher, C. R., Pickering, L. K. & Newburg, D. S. Adv Exp Med Biol 501, 315-323 (2001)).

Human milk glycans inhibit binding of enteropathogens to their receptors

Human milk glycans have structural homology to cell receptors for enteropathogens and function as receptor decoys. For example, pathogenic strains of *Campylobacter* bind specifically to glycans containing H-2, i.e., 2'-fucosyl-N-acetylglucosamine or 2'-fucosyllactose (2'FL); *Campylobacter* binding and infectivity are inhibited by 2'-FL and other glycans containing this H-2 epitope. Similarly, some diarrheagenic *E. coli* pathogens are strongly inhibited *in vivo* by human milk oligosaccharides containing 2-linked fucose moieties. Several major strains of human caliciviruses, especially the noroviruses, also bind to 2-linked fucosylated glycans, and this binding is inhibited by human milk 2-linked fucosylated glycans. Consumption of human milk that has high levels of these 2-linked fucosyloligosaccharides was associated with lower risk of norovirus, *Campylobacter*, ST of *E. coli*-associated diarrhea, and moderate-to-severe diarrhea of all causes in a Mexican cohort of breastfeeding children (Newburg D.S. et al., 2004 Glycobiology, 14:253-263; Newburg D.S. et al., 1998 Lancet, 351:1160-1164). Several pathogens utilize sialylated glycans as their host receptors, such as influenza (Couceiro, J. N., Paulson, J. C. & Baum, L. G. Virus Res 29, 155-165 (1993)), parainfluenza (Amonsén, M., Smith, D. F., Cummings, R. D. & Air, G. M. J Virol 81, 8341-8345 (2007)), and rotaviruses (Kuhlenschmidt, T. B., Hanafin, W. P., Gelberg, H. B. & Kuhlenschmidt, M. S. Adv Exp Med Biol 473, 309-317 (1999)). The

sialyl-Lewis X epitope is used by *Helicobacter pylori* (Mahdavi, J., Sondén, B., Hurtig, M., Olfat, F. O., et al. Science 297, 573-578 (2002)), *Pseudomonas aeruginosa* (Scharfman, A., Delmotte, P., Beau, J., Lamblin, G., et al. Glycoconj J 17, 735-740 (2000)), and some strains of noroviruses (Rydell, G. E., Nilsson, J., Rodriguez-Diaz, J., Ruvoën-Clouet, N., et al. Glycobiology 19, 309-320 (2009)).

Identification of novel $\alpha(1,2)$ fucosyltransferases

The present invention provides novel $\alpha(1,2)$ fucosyltransferase enzymes. The present invention also provides nucleic acid constructs (*i.e.*, a plasmid or vector) carrying the nucleic acid sequence of a novel $\alpha(1,2)$ fucosyltransferases for the expression of the novel $\alpha(1,2)$ fucosyltransferases in host bacterium. The present invention also provides methods for producing fucosylated oligosaccharides by expressing the novel $\alpha(1,2)$ fucosyltransferases in suitable host production bacterium, as further described herein.

Not all $\alpha(1,2)$ fucosyltransferases can utilize lactose as an acceptor substrate. An acceptor substrate includes, for example, a carbohydrate, an oligosaccharide, a protein or glycoprotein, a lipid or glycolipid, *e.g.*, N-acetylglucosamine, N-acetyllactosamine, galactose, fucose, sialic acid, glucose, lactose, or any combination thereof. A preferred alpha (1,2) fucosyltransferase of the present invention utilizes GDP-fucose as a donor, and lactose is the acceptor for that donor.

A method of identifying novel $\alpha(1,2)$ fucosyltransferase enzymes capable of utilizing lactose as an acceptor was previously carried out (as described in PCT/US2013/051777, hereby incorporated by reference in its entirety) using the following steps: 1) performing a computational search of sequence databases to define a broad group of simple sequence homologs of any known, lactose-utilizing $\alpha(1,2)$ fucosyltransferase; 2) using the list of homologs from step 1 to derive a search profile containing common sequence and/or structural motifs shared by the members of the broad group, *e.g.* by using computer programs such as MEME (Multiple Em for Motif Elicitation available at <http://meme.sdsc.edu/meme/cgi-bin/meme.cgi>) or PSI-BLAST (Position-Specific Iterated BLAST available at ncbi.nlm.nih.gov/blast with additional information at cnx.org/content/m11040/latest/); 3) searching sequence databases (*e.g.*, using computer programs such as PSI-BLAST, or MAST (Motif Alignment Search Tool available at <http://meme.sdsc.edu/meme/cgi-bin/mast.cgi>);, using this derived search profile as query, and identifying “candidate sequences” whose simple sequence homology to the original lactose-accepting $\alpha(1,2)$ fucosyltransferase is 40% or less; 4) scanning the scientific literature and

developing a list of “candidate organisms” known to express $\alpha(1,2)$ fucosyl-glycans; 5) selecting only those “candidate sequences” that are derived from “candidate organisms” to generate a list of “candidate lactose-utilizing enzymes”; and 6) expressing each “candidate lactose-utilizing enzyme” and testing for lactose-utilizing $\alpha(1,2)$ fucosyltransferase activity.

The MEME suite of sequence analysis tools (meme.sdsc.edu/meme/cgi-bin/meme.cgi) can also be used as an alternative to PSI-BLAST. Sequence motifs are discovered using the program “MEME”. These motifs can then be used to search sequence databases using the program “MAST”. The BLAST and PSI-BLAST search algorithms are other well known alternatives.

To identify additional novel $\alpha(1,2)$ fucosyltransferases, a multiple sequence alignment query was generated using four previously identified lactose-utilizing $\alpha(1,2)$ fucosyltransferase protein sequences: *H. pylori* futC (SEQ ID NO: 1), *H. mustelae* FutL (SEQ ID NO: 2), *Bacteroides vulgatus* futN (SEQ ID NO: 3), and *E. coli* 0126 wbgL (SEQ ID NO: 4). These sequence alignment and percentage of sequence identity is shown in FIG. 3. An iterative PSI-BLAST was performed, using the FASTA-formatted multiple sequence alignment as the query, and the NCBI PSI-BLAST program run on a local copy of NCBI BLAST+ version 2.2.29. An initial position-specific scoring matrix file (.pssm) was generated by PSI-BLAST, which the program then used to adjust the score of iterative homology search runs. The process is iterated to generate an even larger group of candidates, and the results of each run were used to further refine the matrix.

This PSI-BLAST search resulted in an initial 2515 hits. There were 787 hits with greater than 22% sequence identity to FutC. 396 hits were of greater than 275 amino acids in length. Additional analysis of the hits was performed, including sorting by percentage identity to FutC, comparing the sequences by BLAST to existing $\alpha(1,2)$ fucosyltransferase inventory (of known $\alpha(1,2)$ fucosyltransferases), and manual annotation of hit sequences to identify those originating from bacteria that naturally exist in the gastrointestinal tract. An annotated list of the novel $\alpha(1,2)$ fucosyltransferases identified by this screen are listed in Table 1. Table 1 provides the bacterial species from which the candidate enzyme is found, the GenBank Accession Number, GI Identification Number, amino acid sequence, and % sequence identity to FutC.

Of the identified hits, 12 novel $\alpha(1,2)$ fucosyltransferases were further analyzed for their functional capacity: *Prevotella melaninogenica* FutO, *Clostridium bolteae* FutP, *Clostridium bolteae* +13 FutP, *Lachnospiraceae* sp. FutQ, *Methanosphaerula palustries*

FutR, *Tannerella* sp. FutS, *Bacteroides caccae* FutU, *Butyrivibrio* FutV, *Prevotella* sp.

FutW, *Parabacteroides johnsonii* FutX, *Akkermansia muciniphila* FutY, *Salmonella enterica*

FutZ, and *Bacteroides* sp. FutZA. For *Clostridium bolteae* FutP, the annotation named the wrong initiation methionine codon. Thus, the present invention includes FutP with an

additional 13 amino acids at the N-terminus of the annotated FutP (derived in-frame from the natural upstream DNA sequence), which is designated herein as *Clostridium bolteae* +13

FutP. The sequence identity between the 12 novel $\alpha(1,2)$ fucosyltransferases identified and the 4 previously identified $\alpha(1,2)$ fucosyltransferases is shown in Table 2 below.

Table 2. Sequence Identity

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
<i>H. pylori</i> futC	1	70.10	23.87	20.82	27.68	27.36	23.56	23.28	23.62	25.75	23.72	24.05	22.29	24.19	22.92	22.29
<i>H. mustelae</i> futL	2	70.10	23.87	19.88	26.38	28.21	24.30	23.38	24.62	25.31	25.31	24.47	23.56	25.15	23.55	23.26
<i>Bacteroides vulgatus</i> futN	3	23.99	23.87	25.16	32.05	28.71	28.94	25.79	37.46	32.27	26.13	61.27	71.63	27.67	25.15	84.75
<i>E. coli</i> O126 wbqL	4	20.82	19.88	25.16	24.25	22.73	22.32	26.04	25.45	24.77	21.49	23.29	26.71	24.83	21.45	25.16
<i>Prevotella melaninogenica</i> FutO WP_003814512.1	5	27.68	26.38	32.05	24.25	36.96	31.63	35.74	35.16	55.74	30.28	30.03	32.80	30.09	26.28	31.83
<i>Clostridium bolteae</i> + 13 FutP WP_002570768.1	6	27.36	28.21	28.71	22.73	36.96	37.87	35.10	33.77	36.91	35.74	29.58	31.39	27.67	26.33	29.13
<i>Lachnospiraceae</i> sp. FutQ WP_009251343.1	7	23.56	24.30	28.94	22.32	31.63	37.87	29.87	29.17	32.90	51.02	28.53	30.00	27.69	24.00	27.74
<i>Methanospaerula palustris</i> FutR WP_002467213.1	8	23.28	23.38	25.79	26.04	35.74	35.10	29.87	28.71	38.24	31.41	25.39	28.08	30.65	23.93	25.55
<i>Tannerella</i> sp. FutS WP_021929367.1	9	23.62	24.62	37.46	25.45	35.16	33.77	29.17	28.71	34.41	30.03	35.71	36.27	26.48	21.75	36.60
<i>Bacteroides caccae</i> FutU WP_005675707.1	10	25.75	25.31	32.27	24.77	55.74	36.91	32.90	38.24	34.41	31.21	29.94	33.33	29.28	24.46	33.01
<i>Butyrivibrio</i> FutV WP_022772718.1	11	23.72	25.31	26.11	21.49	30.28	35.74	51.02	31.41	30.03	31.21	27.62	26.20	26.46	22.15	26.52
<i>Prevotella</i> sp. FutW WP_022481266.1	12	24.05	24.47	61.27	23.29	30.03	29.58	28.53	25.39	35.71	29.94	27.62	57.60	25.79	22.15	59.01
<i>Parabacteroides johnsonii</i> FutX WP_008155883.1	13	22.29	23.56	71.63	26.71	32.80	31.39	30.00	28.08	36.27	33.33	26.20	57.60	28.71	24.00	74.02
<i>Akkermansia muciniphila</i> FutY WP_001877555	14	24.19	25.15	27.67	24.63	30.09	27.67	27.69	30.65	26.48	29.28	26.46	25.79	28.71	21.45	28.08
<i>Salmonella enterica</i> FutZ WP_023214330	15	22.92	23.55	25.15	21.45	26.28	26.53	24.00	23.93	21.75	24.46	22.15	22.15	24.00	21.45	24.62
<i>Bacteroides</i> sp. FutZA WP_022161880.1	16	22.29	23.26	84.75	25.16	31.83	29.13	27.74	25.55	36.60	33.01	26.52	59.01	74.02	28.08	24.62

Based on the amino acid sequences of the identified $\alpha(1,2)$ fucosyltransferases (*i.e.*, in Table 1), syngenes can be readily designed and constructed by the skilled artisan using standard methods known in the art. For example, the syngenes include a ribosomal binding site, are codon-optimized for expression in a host bacterial production strain (*i.e.*, *E. coli*), and have common 6-cutter restriction sites or sites recognized by endogenous restriction enzymes present in the host strain (*i.e.*, EcoK restriction sites) removed to ease cloning and expression in the *E. coli* host strain. In a preferred embodiment, the syngenes are constructed with the following configuration: EcoRI site – T7g10 RBS – $\alpha(1,2)$ FT syngene – XhoI site. The nucleic acid sequences of sample syngenes for the 12 identified $\alpha(1,2)$ fucosyltransferases are shown in Table 3. (the initiating methionine ATG codon is bolded)

Table 3. Nucleic acid sequences of 12 novel $\alpha(1,2)$ fucosyltransferase syngenes

Bacteria/ Gene name	Sequence	SEQ ID NO:
FutO	CAGTCAGTCAGAATTCAGAAGGAGATATACAT ATG AAAATCGTCAAAATCCTGGGCGGTCTGGGCAATCAGATGTTCCAGTATGCTCTGTACCTGAGCCTGCAAGAAAGTTTCCAAAAA GAACGTGTGGCCCTGGACCTGTCCTCCTTCCACGGCTATCACCTGCATAATGGCTTTGAG CTGGAGAACATTTTCTCCGTTACCGCTCAGAAAGCATCCGCCGCAGATATCATGCGTATT GCTTATTACTACCCGAACATCTGCTGTGGCGCATTGGCAAACGTTTTCTGCCGCGTCGT AAAGGTATGTGCCTGGAATCTAGCTCCCTGCGTTTCGATGAAAGCGTTCTGCGTCAGGAA GGTAACCGTTATTTTGACGGTTACTGGCAAGACGAACGCTACTTCGCAGCCTATCGTGAA	276

	AAAGTGCCTGAAGGCTTTCACCTTCTCCTGCATTCAAACGCGCAGAAAACCTGAGCCTGCTG GAAAAACCTGGACGAAAACAGCATTTGCTCTGCATGTTTCGTCGCGGTGATTACGTAGGTAAT AACCTGTACCAAGGCATCTGTGACCTGGACTACTACCGTACCGCTATCGAGAAAATGTGT GCACACGTTACTCCGTCTCTGTTTTGTATCTTTTCCAACGACATCACGTGGTGCCAGCAG CACCTGCAACCGTACCTGAAGGCCCTGTGGTGTACGTTACTTGGAACACCGGTGTTGAA TCCACCGCGATATGCAGCTGATGTCCTGCTGCGCACATAACATCATCGCGAATAGCTCC TTCTCTTGGTGGGGTGCTTGGCTGAATCAGAACCGTGAAAAAGTTGTTATCGCCCCGAAA AAATGGCTGAACATGGAAGAATGTCACTTCACGCTGCCGGCAAGCTGGATCAAAATTTAG CTCGAGTGAAGTACTG	
FutP	CAGTCAGTCAGAATTCAAGAAGGAGATATACATATGGTGATTATCAAAATGATGGGTGGT CTGGGCAACCAGATGTTCCAGTACGCACTGTACAAAGCATTTCGAGCAGAAGCACATCGAT GTGTATGCAGACCTGGCATGGTACAAAAACAAATCCGTGAAATTTGAACTGTACAACCTC GGCATTAAAAATCAACGTAGCATCCGAGAAAGACATCAACCGTCTGAGCGATTGCCAGGCG GACTTTGTTTTCCCGCATCCGCCGTAAAAATCTTTGGTAAAAAAAAGAGCTTCGTATCTGAA AAAAATGACTCCTGCTATGAAAACGACATCCTGCGTATGGACAACGTTTATCTGAGCGGT TATTGGCAGACCGAAAAATACTTCTCTAACACGCGTGAGAAGCTGCTGGAGGATTATTCC TTCGCTCTGGTAACTCTCAGGTGTCCGAATGGGAAGACTCCATTTCGCAACAAAAACAGC GTTAGCATCCATATCCGTCGTGGTGATTATCTACAGGGCGAAGCTGTATGGTGGTATTTGC ACCTCTCTGTACTACGCCGAAGCAATCGAGTACATTAAAAATGCGTGTTCCGAACGCAAAA TTCTTCGTTTTCTCTGATGACGTTGAATGGGTAAACAGCAAGAAGACTTCAAAGGCTTC GTAATCGTTGATCGCAACGAGTATTCTAGCGCTCTGTCCGATATGTACCTGATGTCCCTG TGCAAGCATAACATTATTGCTAACTCCTCTTTCAGCTGGTGGGCGAGCTTGGCTGAACCGT AACGAAGAAAAAATTGTAATCGCGCCGCGCGTGGCTGAACGGCAAGTGCACCCCGAT ATCTGGTGTAAAAAATGGATTTCGTATCTAGCTCGAGTGAAGTACTG	277
FutQ	CAGTCAGTCAGAATTCAAGAAGGAGATATACATATGGTGATCGTACAGCTGAGCGGCGGT CTGGGCAACCAGATGTTTCGAATACGCGCTGTACCTGAGCCTGAAAGCAAAGGCAAAGAA GTGAAAATTGACGATGTTACGTGTTACGAGGGCCCTGGCACCCGTCCGCGTCAACTGGAT GTTTTTGGTATCACGTACGATCGCGCTCTCGTGAGGAGCTGACTGAGATGACGGACGCG AGCATGGATGCGCTGTCTCGTGTTCGTGCGAACTGACCGGTCCGCGCACTAAAGCGTAC CGCGAACGCGACATCAACTTCGATCCACTGGTTATGAAAAAGACCCGGCACTGCTGGAA GGCTGTTTTCCAGTCTGACAAATACTTTCGTGATTGCGAAGGCCGCGTGCGCGAAGCGTAT CGTTTCCGCGGCATTGAATCCGGCGCGTTCGCGTCCGGAAGACTATCTGCGCCTGGAA AAGCAGATCGAAGATTGTCAGTCCGTATCCGTACACATCCGTGCGTGGCGACTACCTGGAC GAATCTCATGGTGGTCTGTACACCGGCATTGTAAGTGGAGGCGTACTATAAAGAGGCTTTT GCTCGCATGGAACGTCTGGTTCGCGGCGCACGTTTCTTCTGTCTCTAACGATCCAGAA TGGACTCGTGAGCACTTTGAGAGCAAGAACTGCGTTCTGGTTGAAGGTAGCACCGAAGAC ACGGGTACATGGACCTGTACCTGATGAGCCGCTGCCGCCACAATATTATTGCCAACTCT TCTTTCAGCTGGTGGGCGCTTGGCTGAATGAGAACCCTGAGAAAAAGTCATCGCACCG GCTAAATGGCTGAACGGTCTGTGAGTGCCGTGATATCTATACCGAACGCATGATTCTGTCTG TAGCTCGAGTGAAGTACTG	278
FutR	CAGTCAGTCAGAATTCAAGAAGGAGATATACATATGATCATTGTTTCGTCTGAAAGGCGGT CTGGGCAACCAACTGTCTCAGTATGCACTGGGCCGTAAAGTCGCGCATCTGCACAATACC GAACTGAAACTGGACACCACCTGGTTCCACCATATCTCCTCCGACACTCCACGTACCTAC CGTCTGAACAATTATAACATCATCGGCACTATTGCATCCGCAAAGGAAATCCAGCTGATC GAACGTGGTTCGCGCGCAAGGCCGTGGCTACCTGCTGTCTAAAAATTTCTGATCTGCTGACT CCGATGTACCGTCTGATACCTACGTCCGTGAACGTATGCATACCTTCGATAAAGCTATCCTG ACCGTTCCGGACAACGTGTACCTGGATGGTTACTGGCAGACCGAAAAGTACTTCAAAGAC ATCGAAGAAATCCTGCGCCGTGAGGTTACGCTGAAAGATGAACCGGATAGCATCAACCTG GAAATGGCTGAACGATTTACGGCTTGCCACAGCGTTTCCCTGCACGTGCGTGGTGGCGAC TACGTTTTCCAACCCGACCACCTCAACAATTCACGGCTGTTGCTCCATTGACTACTACAAC CGCGCTATCTCTCTGATTGAAGAAAAAGTGGATGACCCGTCTTTCTTTATTTTTCTGAC GATCTGCCGTGGGCTAAAGAAAACCTGGACATCCCTGGCGAAAAAACCTTCGTTGCGCAT AACGGCCCCGAAAAAGAGTATTGCGATCTGTGGCTGATGTCTCTGTGCCAGCACCATATC ATCGCAAACTCTTCTTTCAGCTGGTGGGGTGCTGGCTGGGTCAAGACGCCGAAAAGATG GTGATCGCGCCGCTGCTGGGCCCTGTCCGAGAGCTTTGACACTTCTGACATCATTCCG GACTCTTGGATTACTATCTAGCTCGAGTGAAGTACTG	279
FutS	CAGTCAGTCAGAATTCAAGAAGGAGATATACATATGGTACGCATTGTGGAAATCATCGGC GGTCTGGGTAACCAGATGTTCCAGTACGCATTCTCCCTGTACCTGAAAAACAAATCTCAC ATCTGGGACCGTCTGTATGTGGACATCGAGGCGATGAAACCTACGATCGTCACTATGGT CTGGAACCTGGAGAAAGTTTTCAATCTGAGCCTGTGTCCAATCTCTAACCGTCTGCACCGC AACCTGCAAAAACGCTCCTTCGCAAAACACTTTGTAAAGAGCCTGTACGAGCACTCTGAA	280

	TGCGAGTTTCGACGAACCGGTGTACCGTGGCCTGCGTCCTTATCGCTATTATCGCGGCTAC TGGCAAAACGAAGTTACTTCGTTGATATTGAACCGATGATCCGTGAGGCTTTTCAGTTT AACGTTAACATCCTGAGCAAAAAGACTAAAGCGATCGCATCCAAATGCGCCGTGAACGT TCCGTATCTATCCATGTTTCGCCGTGGTGATTACGAAAACCTGCCGGAAGCGAAAGCGATG CATGGCGGTATTTGTTCTCTGGACTATTACCACAAAGCGATCGACTTCATCCGCCAGCGT CTGGATAATAACATCTGTTTCTATCTGTTCTCCGACGATATCAATTGGGTAGAAGAAAAC CTGCAACTGGAAAACCGTTGCATCATCGACTGGAACCAGGGCGAAGATAGCTGGCAGGAC ATGTACCTGATGAGCTGCTGCCGCCACCACATTATCGCAAACAGCTCTTTCTCCTGGTGG GCGGATGGCTGAATCCAAACAAGAACAAAATCGTACTGACCCCGAACAATGGTTCAAC CATACTGACGCAGTGGGTATCGTCCCAAAGTCCTGGATTAAAATTCCTGTGTTTTAGCTC GAGTGACTGACTG	
FutU	CAGTCAGTCAGAATTCAGAAGGAGATATACATATGAAAATCGTTAAAATCCTGGGCGGC CTGGGTAACCAGATGTTTCAGTACGCCCTGTTCTGTCTCTGAAAGAACGCTTCCCGCAT GAACAGGTGATGATTGACACCAGCTGCTTCCGCAATTACCCACTGCACAACGGTTTCGAA GTGGATCGTATCTTCGCCCAGAAAGCACCAGGTTGCCTCTTGGCGTAACATCCTGAAGGTT GCC'TACCCGTACCCGAAC'TACCGTTTCTGGAAAATCGGTAAATACATCCTGCCTAAACGT AAAACCATGTGTGTAGAGCGTAAAACTTCAGCTTTGACGCCCGAGTCCTGACCCGTAAA GGCGATTGCTACTATGATGGCTACTGGCAGCATGAGGAATATTTCTGTGATATGAAAGAA ACGATTTGGGAGGCTTTCTCCTTCCCTGAGCCGGTTGATGGTCGTAACAAGGAGATCGGT GCCCTGCTACAGGCATCTGATAGCGCTTCCCTGCACGTTTCGTCGCGGTGACTACGTGAAC CACCAC'TGTTTCGTGGTATTTGTGACCTGGACTATTATAAACGTGCCATCCACTACATG GAAGAACGCGTCAACCCACAGCTGTA'CTGCGTTTTCAGCAACGATATGGCCTGGTGCGAG TCCACCTGCGTGCACTGCTGCCAGGCAAAGAAGTAGTTTATGTTGACTGGAACAAGGGT GCGGAATCTTACGTTGATATGCGTCTGATGAGCCTGTGCCGTCACAACATCATCGCTAAC TCTTCTTTTACGCTGGTGGGGCGCATGGCTGAACCGTAACCCGAGAAAGTGGTGGTAGCG CCGGAACGTTGGATGAACAGCCCGATTGAAGACCCAGTGAGCGACAAATGGATTAAACTG TAGCTCGAGTGACTGACTG	281
FutV	CAGTCAGTCAGAATTCAGAAGGAGATATACATATGATCATCATCCAGCTGAAAGGTGGC CTGGGCAACCAAATGTTCCAGTACGCGCTGTACAAAATCCCTGAAAAACGTGGTAAAGAA GTTAAATTTGATGACAAAAC'TGGCTTCGTGAACGACAAAAC'TGCGTATCCCGGTACTGTCC CGTTGGGGTGT'TGAGTACGATCGTGCAACCCGACGAAGAGATTATTAACCTGACCGACTCC AAAATGGACCTGTTCTCTCGCATCCGCCGTAAACTGACTGGCCGCAAAACGTTCCGTATC GACGAAGAATCCGGTAAATTC AACCCGGAATCCTGAAAAAGAGAACGCTTATCTGGTG GGTTATTGGCAGTGCGACAAGTACTTCGACGACAAAGATGTGGTTTCGCGAAATTCGTGAA GCGTTTCGAGAAAAAACCGCAGGAGCTGATGACCGACGCCAGCTCTTGGTCTACTCTACAG CAGATTGAATGCTGCGAGTCCGTATCCCTGACGTACGTCTGATTACGTGGACGAG GAACATATTCATATCCATAACATCTGTACGGAAAAATACTATAAAAAACGCCATTGATCGT GTGCGTAAACAGTACCCGAGCGCAGTGTTCCTTATCTTACCCGATGATAAAGAATGGTG CGCGACCACTTTAAAGGTCCGAAC'TTCATCGTAGTCGAACTGGAAGAAGGCGACGGTACC GACATCGCTGAAATGACTCTGATGTCCCGCTGTAAACATCACATCATCGCTAATTCTAGC TTTAGCTGGTGGGCGGCGTGGCTGAACGACTCCCGGAAAAAATCGTGATCGCTCCTCAG AAATGGATTAAACAACCGGACATGGACGATATTTACACCGAGCGTATGACTAAAAATCGCA CTGTAGCTCGAGTGACTGACTG	282
FutW	CAGTCAGTCAGAATTCAGAAGGAGATATACATATGCGCCTGGTTAAAATGATCGGCGGT CTGGGTAATCAGATGTTTCATCTACGCGTTT'TACCTACAGATGCGTAAGCGTTTCTCCAAC GTTTCGTATCGACCTGACCGATATGATGCAC'TACAACGTACACTATGGCTACGAACTGCAC AAAGTTTTTCGGTCTGCCGCGCACCGAGTTC'TGTATGAACCAGCCTCTGAAAAAGGTTCTG GAGTTCCTGTTCTTCCGTACCATTGTTGAACGTAAACAGCACGGTTCGTATGGAGCCGTAT ACTTGCCAGTATGTTTGGCCGCTGGTTTAC'TTTAAGGGCTTCTATCAGTCCGAACGTTAC TTCTCCGAAGTTAAGGACGAAGTTCGTGAGTGTTCACCTTCAATCCGGCACTGGCGAAT CGTTCTTCCCAACAGATGATGGAACAGATCCAGAATGATCCTCAGGCTGTCTCTATCCAC ATCCGTCTGTGGCGACTATCTGAATCCGAAGCACTACGACACTATCGGTTGTATCTGTCTCAG CTGCCGTATTACAAGCACGCCGTTTCCGAAATTA AAAAGTACGTTTCTAACCCCTCACTTT TACGTTTTTCTCCGAAGACCTGGATTGGGTCAAAGCAAACCTGCCGCTGGAAAAACGCACAG TACATCGATTGGAACAAAGGCGCAGATAGCTGGCAGGATATGATGCTGATGAGCTGTTGC AAACACCACATTATCTGTAACCTCACCTTTAGCTGGTGGGCGGCGTGGCTGAACCCATCT GTGCAAAAAACCGTGATCATGCCGGAACAGTGGACGTCTCGTCAAGATTCCGTGGACTTT GTGGCTAGCTGTGGCCGTTGGGTCCGTGTTAAAACGGAGTAGCTCGAGTGACTGACTG	283
FutX	CAGTCAGTCAGAATTCAGAAGGAGATATACATATGCGTCTGATCAAGATGATCGGCGGC CTGGGTAACCAGATGTTTATCTACGCGTTTCTACCTGAAAATGAAACACCAATTACCCGGAT ACGAACATCGATCTGTCTGACATGGTTTATTATAAAGTTTCAACCGGTTATGAGATGAAC	284

	CGTATCTTTGACCTGAGCCAGACTGAATTTTGCATCAACCGTACCCTGAAAAAATCCTG GAGTTCCTGTTCTTCAAAAAATCTACGAACGTCGCCAGGACCCGTCTACTCTGTATCCA TACGAAAAACGTTATTTTGGCCGCTGCTGTACTTTAAAGGTTTCTACCAGTCTGAACGC TTCTTCTTCGATATCAAAGACGACGTTTCGTAAAGCCTTCTCTTTTAACCTGAACATCGCT AACCCGGAAGCCTGGAAGTGTGAAACAGATCGAAGTTGACGACCAAGCTGTTTCTATC CACATCCGCCGTGGTGACTACCTGCTGCCGCGTCACTGGGCAAACACGGGTTCGCTGTGC CAGCTGCCGTATTACAAGAACGCGATCGCGGAAATGGAGAACCGTATTACTGGCCCGAGC TACTACGTGTTCTCTGATGATATCTCTTGGGTTAAAGAAAACATCCCGCTGAAGAAAGCG GTCTACGTGACGTGGAACAAGGGCGAAGACAGCTGGCAGGATATGATGCTGATGAGCCAC TGTCGTCAACCATATCTGTAATTCTACGTTCTCCTGGTGGGGTGCTTGGCTGAACCCA CGTAAAGAGAAAAATCGTCATCGCGCCGTGTGCTGGTTCCAGCATAAAGAAACCCCGGAC ATGTACCCGAAAGAATGGATCAAAGTACCGATTAACTAGCTCGAGTGACTGACTG	
FutZ	CAGTCAGTCAGAATTCAAGAAGGAGATATACATATGTTATTCTTGCCTGTCTGGTGGCCTG GGTAACCAAATGTTTCAATACGCAGCAGCGTATATCCTGAAGCAGTATTTTCAGTCTACC ACTCTGGTCTTGGATGATAGCTATTACTATTCCAGCCGAAACGTGATACCGTTTCGTAGC CTGGAACCTGAATCAGTTCAACATCTCTTATGATCGTTTTCAGCTTCGCGGATGAAAAAGAG AAGATCAAACCTGCTGCGCAAATTCAAACGTAACCCGTTCCCTAAACAGATTTCCGAGATC CTGTCTATTGCGCTGTTTCGGCAAATACGCGCTGTCCGACCGTGCATTTTACACCTTCGAA ACTATCAAAAACATCGACAAAGCGTGCTGTCTCCTTTTACCAGGACGCCGATCTGCTG AATAAATATAAGCAGCTGATCCTGCCGCTGTTTGAACCTGCGCGATGACCTGCTGGATATC TGCAAGAACCTGGAACGTATTCCTGATCCAACGCAGCAACAATACCACTGCACTGCAT ATCCGCCGTGGCGACTACGTGACCAACCAGCACGCCGCGAAATACACGGCGTGCTGGAC ATCAGCTACTATAACCACGCAATGGAATACGTGGAACGTGAACGCGGCAAACAGAACCTTC ATTATCTTCAGCGATGATGTACGTTGGGCACAGAAAGCGTTTCTGGAGAACGATAATTGC TACGTGATTAACAACCTCCGACTACGATTTCTCTGCGATCGATATGATCTGATGTCTCTG TGCAAAAACAACATCATCGCAAATTCACCTACTCCTGGTGGGGTGCGTGGCTGAACAAA TACGAGGACAAACTGGTTATCTCTCCGAAACAATGGTTTCTGGGTAACAACGAAACCTCT CTGCGTAACGCGTCTTGGATCACCTGTAGCTCGAGTGACTGACTG	285
FutZA	CAGTCAGTCAGAATTCAAGAAGGAGATATACATATGCGTCTGATCAAGATGACCGGTGGC CTGGGTAACAGATGTTTCATCTACGCGTTTATCTGCGTATGAAAAACGTTATCCGAAA GTTTCGTATTGATCTGTCTGATATGGTTTATTATCACGTTTACCACGGCTATGAAATGCAC CGTGTGTTTCAATCTGCCGCACACCGAATTTTGCATCAACCAGCCGCTGAAAAAAGTGATC GAGTTCTCTGTTTTTCAAAAAGATTTACGAACGTAAACAGGACCTAATCTCTGCGTGCA TTCGAGAAGAAGTATCTGTGGCCGCTGCTGTACTTCAAAGGTTTCTATCAATCTGAGCGC TTCTTTGCTGACATCAAAGACGAGGTTTCGTAAAGCATTACCTTTGACTCTTCTAAAGTG AACGCTCGCTCTGCCGAACCTGCTGCGTGCCTGGATGCCGATGCTAACGCGGTTAGCCTG CACATTCGTGCGGGTACTATCTACAGCCGCAGCATTGGGCTACCACTGGTTCTGTCTGC CAGCTGCCGTACTACCAGAACGCGATCGCTGAAATGAACCGTCGCGTTGCTGCCCCGAGC TACTACGTTTTTCAGCGATGACATCGCGTGGGTGAAGGAAAACATCCCTCTACAGAACGCA GTGTACATCGACTGGAATAAAGGCGAAGAAAGCTGGCAGGATATGATGCTGATGAGCCAC TGCCGCCACACATTATCTGTAACAGCACCTTCTCTTGGTGGGGCGCGTGGCTGGACCCG CACGAGGACAAAATTGTAATCGTTCCGAATCGTTGGTTCCAGCATTGCGAAACTCCTAAC ATCTATCCGCGAGGCTGGGTGAAAGTTGCGATTAAATTAGCTCGAGTGACTGACTG	286

In any of the methods described herein, the $\alpha(1,2)$ fucosyltransferase genes or gene products may be variants or functional fragments thereof. A variant of any of genes or gene products disclosed herein may have 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, or 99% sequence identity to the nucleic acid or amino acid sequences described herein.

Variants as disclosed herein also include homolog, orthologs, or paralogs of the genes or gene products described herein that retain the same biological function as the genes or gene products specified herein. These variants can be used interchangeably with the genes recited in these methods. Such variants may demonstrate a percentage of homology or

identity, for example, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, or 99% identity conserved domains important for biological function, preferably in a functional domain, e.g. catalytic domain.

The term "% identity," in the context of two or more nucleic acid or polypeptide sequences, refer to two or more sequences or subsequences that are the same or have a specified percentage of amino acid residues or nucleotides that are the same, when compared and aligned for maximum correspondence, as measured using one of the following sequence comparison algorithms or by visual inspection. For example, % identity is relative to the entire length of the coding regions of the sequences being compared, or the length of a particular fragment or functional domain thereof.

For sequence comparison, typically one sequence acts as a reference sequence, to which test sequences are compared. When using a sequence comparison algorithm, test and reference sequences are input into a computer, subsequence coordinates are designated, if necessary, and sequence algorithm program parameters are designated. The sequence comparison algorithm then calculates the percent sequence identity for the test sequence(s) relative to the reference sequence, based on the designated program parameters.

Percent identity is determined using search algorithms such as BLAST and PSI-BLAST (Altschul et al., 1990, J Mol Biol 215:3, 403-410; Altschul et al., 1997, Nucleic Acids Res 25:17, 3389-402). For the PSI-BLAST search, the following exemplary parameters are employed: (1) Expect threshold was 10; (2) Gap cost was Existence:11 and Extension:1; (3) The Matrix employed was BLOSUM62; (4) The filter for low complexity regions was "on".

Changes can be introduced by mutation into the nucleic acid sequence or amino acid sequence of any of the genes or gene products described herein, leading to changes in the amino acid sequence of the encoded protein or enzyme, without altering the functional ability of the protein or enzyme. For example, nucleotide substitutions leading to amino acid substitutions at "non-essential" amino acid residues can be made in the sequence of any of sequences expressly disclosed herein. A "non-essential" amino acid residue is a residue at a position in the sequence that can be altered from the wild-type sequence of the polypeptide without altering the biological activity, whereas an "essential" amino acid residue is a residue at a position that is required for biological activity. For example, amino acid residues that are conserved among members of a family of proteins are not likely to be amenable to mutation. Other amino acid residues, however, (e.g., those that are poorly conserved among members of the protein family) may not be as essential for activity and thus are more likely to be

amenable to alteration. Thus, another aspect of the invention pertains to nucleic acid molecules encoding the proteins or enzymes disclosed herein that contain changes in amino acid residues relative to the amino acid sequences disclosed herein that are not essential for activity (*i.e.*, fucosyltransferase activity).

An isolated nucleic acid molecule encoding a protein essentially retaining the functional capability compared to any of the genes described herein can be created by introducing one or more nucleotide substitutions, additions or deletions into the corresponding nucleotide sequence, such that one or more amino acid substitutions, additions or deletions are introduced into the encoded protein.

Mutations can be introduced into a nucleic acid sequence by standard techniques such that the encoded amino acid sequence is altered, such as site-directed mutagenesis and PCR-mediated mutagenesis. Preferably, conservative amino acid substitutions are made at one or more predicted non-essential amino acid residues. A "conservative amino acid substitution" is one in which the amino acid residue is replaced with an amino acid residue having a similar side chain. Families of amino acid residues having similar side chains have been defined in the art. Certain amino acids have side chains with more than one classifiable characteristic. These families include amino acids with basic side chains (*e.g.*, lysine, arginine, histidine), acidic side chains (*e.g.*, aspartic acid, glutamic acid), uncharged polar side chains (*e.g.*, glycine, asparagine, glutamine, serine, threonine, tyrosine, tryptophan, cysteine), nonpolar side chains (*e.g.*, alanine, valine, leucine, isoleucine, proline, phenylalanine, methionine, tyrosine, tryptophan), beta-branched side chains (*e.g.*, threonine, valine, isoleucine) and aromatic side chains (*e.g.*, tyrosine, phenylalanine, tryptophan, histidine). Thus, a predicted nonessential amino acid residue in a given polypeptide is replaced with another amino acid residue from the same side chain family. Alternatively, in another embodiment, mutations can be introduced randomly along all or part of a given coding sequence, such as by saturation mutagenesis, and the resultant mutants can be screened for given polypeptide biological activity to identify mutants that retain activity. Conversely, the invention also provides for variants with mutations that enhance or increase the endogenous biological activity. Following mutagenesis of the nucleic acid sequence, the encoded protein can be expressed by any recombinant technology known in the art and the activity of the protein can be determined. An increase, decrease, or elimination of a given biological activity of the variants disclosed herein can be readily measured by the ordinary person skilled in the art, *i.e.*, by measuring the capability for mediating oligosaccharide modification, synthesis, or degradation (via detection of the products).

The present invention also provides for functional fragments of the genes or gene products described herein. A fragment, in the case of these sequences and all others provided herein, is defined as a part of the whole that is less than the whole. Moreover, a fragment ranges in size from a single nucleotide or amino acid within a polynucleotide or polypeptide sequence to one fewer nucleotide or amino acid than the entire polynucleotide or polypeptide sequence. Finally, a fragment is defined as any portion of a complete polynucleotide or polypeptide sequence that is intermediate between the extremes defined above.

For example, fragments of any of the proteins or enzymes disclosed herein or encoded by any of the genes disclosed herein can be 10 to 20 amino acids, 10 to 30 amino acids, 10 to 40 amino acids, 10 to 50 amino acids, 10 to 60 amino acids, 10 to 70 amino acids, 10 to 80 amino acids, 10 to 90 amino acids, 10 to 100 amino acids, 50 to 100 amino acids, 75 to 125 amino acids, 100 to 150 amino acids, 150 to 200 amino acids, 200 to 250 amino acids, 250 to 300 amino acids, 300 to 350 amino acids, 350 to 400 amino acids, 400 to 450 amino acids, or 450 to 500 amino acids. The fragments encompassed in the present invention comprise fragments that retain functional fragments. As such, the fragments preferably retain the catalytic domains that are required or are important for functional activity. Fragments can be determined or generated by using the sequence information herein, and the fragments can be tested for functional activity using standard methods known in the art. For example, the encoded protein can be expressed by any recombinant technology known in the art and the activity of the protein can be determined. The biological function of said fragment can be measured by measuring ability to synthesize or modify a substrate oligosaccharide, or conversely, to catabolize an oligosaccharide substrate.

Within the context of the invention, “functionally equivalent”, as used herein, refers to a gene or the resulting encoded protein variant or fragment thereof capable of exhibiting a substantially similar activity as the wild-type fucosyltransferase. Specifically, the fucosyltransferase activity refers to the ability to transfer a fucose sugar to an acceptor substrate via an α -(1,2)-linkage. As used herein, “substantially similar activity” refers to an activity level within 5%, 10%, 20%, 30%, 40%, or 50% of the wild-type fucosyltransferase.

To test for lactose-utilizing fucosyltransferase activity, the production of fucosylated oligosaccharides (*i.e.*, 2'-FL) is evaluated in a host organism that expresses the candidate enzyme (or syngene) and which contains both cytoplasmic GDP-fucose and lactose pools. The production of fucosylated oligosaccharides indicates that the candidate enzyme-encoding sequence functions as a lactose-utilizing α -(1,2)fucosyltransferase.

Engineering of *E. coli* to produce human milk oligosaccharide 2'-FL

Described herein is a gene screening approach, which was used to validate the novel α (1,2) fucosyltransferases (α (1,2) FTs) for the synthesis of fucosyl-linked oligosaccharides in metabolically engineered *E. coli*. Of particular interest are α (1,2) FTs that are capable of the synthesis of the HMOS 2'-fucosyllactose (2'-FL). 2'-FL is the most abundant fucosylated oligosaccharide present in human milk, and this oligosaccharide provides protection to newborn infants against infectious diarrhea caused by bacterial pathogens such as *Campylobacter jejuni* (Ruiz-Palacios, G.M., et al. (2003). *J Biol Chem* 278, 14112-120; Morrow, A.L. et al. (2004). *J Pediatr* 145, 297-303; Newburg, D.S. et al. (2004). *Glycobiology* 14, 253-263). Other α (1,2) FTs of interest are those capable of synthesis of HMOS lactodifucotetraose (LDFT), laco-N-fucopentaose I (LNFI), or lacto-N-difucohexaose I (LDFH I).

The synthetic pathway of fucosyl oligosaccharides of human milk is illustrated in FIG. 1. Structurally, 2'-FL consists of a fucose molecule α 1,2 linked to the galactose portion of lactose (Fuc α 1-2Gal β 1-4Glc). An α (1,2) FT from *H. pylori* strain 26695 termed FutC has been utilized to catalyze the synthesis of 2'-FL in metabolically engineered *E. coli* (Drouillard, S. et al. (2006). *Angew Chem Int Ed Engl* 45, 1778-780).

Candidate α (1,2) FTs (*i.e.*, syngenes) were cloned by standard molecular biological techniques into an expression plasmid. This plasmid utilizes the strong leftwards promoter of bacteriophage λ (termed P_L) to direct expression of the candidate genes (Sanger, F. et al. (1982). *J Mol Biol* 162, 729-773). The promoter is controllable, *e.g.*, a *trp-cl* construct is stably integrated into the *E. coli* host's genome (at the *ampC* locus), and control is implemented by adding tryptophan to the growth media. Gradual induction of protein expression is accomplished using a temperature sensitive *cl* repressor. Another similar control strategy (temperature independent expression system) has been described (Mieschendahl et al., 1986, *Bio/Technology* 4:802-808). The plasmid also carries the *E. coli* *rcsA* gene to up-regulate GDP-fucose synthesis, a critical precursor for the synthesis of fucosyl-linked oligosaccharides. In addition, the plasmid carries a β -lactamase (*bla*) gene for maintaining the plasmid in host strains by ampicillin selection (for convenience in the laboratory) and a native *thyA* (thymidylate synthase) gene as an alternative means of selection in *thyA*⁻ hosts. Alternative selectable markers include the *proBA* genes to complement proline auxotrophy (Stein et al., (1984), *J Bacteriol* 158:2, 696-700 (1984) or *purA* to complement adenine auxotrophy (S. A. Wolfe, J. M. Smith, *J Biol Chem* **263**, 19147-53

(1988)). To act as plasmid selectable markers each of these genes are first inactivated in the host cell chromosome, then wild type copies of the genes are provided on the plasmid.

Alternatively a drug resistance gene may be used on the plasmid, e.g. beta-lactamase (this gene is already on the expression plasmid described above, thereby permitting selection with ampicillin). Ampicillin selection is well known in the art and described in standard manuals such as Maniatis et al., (1982) Molecular cloning, a laboratory manual. Cold Spring Harbor Laboratory, Cold Spring, NY.

The nucleic acid sequence of such an expression plasmid, pEC2-(T7)FutX-rcsA-thyA (pG401) is provided below. The underlined sequence represents the FutX syngene, which can be readily replaced with any of the novel $\alpha(1,2)$ FTs described herein using standard recombinant DNA techniques.

TCGCGCGTTTCGGTGATGACGGTGAAAACCTCTGACACATGCAGCTCCCGGAGACGGTCACAGCTTGT
CTGTAAGCGGATGCCGGGAGCAGACAAGCCCGTCAGGGCGCGTCAGCGGGTGTGGCGGGTGTGGGG
CTGGCTTAACCTATGCGGCATCAGAGCAGATTGTACTGAGAGTGCACCATATATGCGGTGTGAAATACC
GCACAGATGCGTAAGGAGAAAATACCGCATCAGGCGCCTCCTCAACCTGTATATTCGTAAACACGCC
CAATGGGAGCTGTCTCAGGTTTGTTCCTGATTGGTTACGGCGCGTTTCGCATCATTTGTTGAGTTTTTC
CGCCAGCCCGACGCGCAGTTTACCGGTGCCTGGGTGCAGTACATCAGCATGGGGCAAATTCCTTCCAT
CCCGATGATTGTGCGGGTGTGATCATGATGGTCTGGGCATATCGTCGCAGCCACAGCAACACGTTT
CCTGAGGAACCATGAAACAGTATTTAGAACTGATGCAAAAAGTGCTCGACGAAGGCACACAGAAAAAC
GACCGTACCGGAACCGGAACGCTTTCATTTTTTGGTCATCAGATGCGTTTTAACCTGCAAGATGGATT
CCCGCTGGTGACAATAACGTTGCCACCTGCGTTCCATCATCCATGAACTGCTGTGGTTTCTGCAGG
GCGACACTAACATTGCTTATCTACACGAAAACAATGTCACCATCTGGGACGAATGGGCCGATGAAAAC
GGCGACCTCGGGCCAGTGTATGGTAAACAGTGGCGCGCCTGGCCAACGCCAGATGGTCGTATATTGA
CCAGATCACTACGGTACTGAACCAGCTGAAAACGACCCGGATTTCGCGCCGCATTATTGTTTCAGCGT
GGAACGTAGGCGAACTGGATAAAATGGCGCTGGCACCGTGCCATGCATTCTTCCAGTTCTATGTGGCA
GACGGCAAACTCTCTTGCCAGCTTTATCAGCGCTCCTGTGACGTCTTCCTCGGCCTGCCGTTCAACAT
TGCCAGCTACGCGTTATTGGTGCATATGATGGCGCAGCAGTGCATCTGGAAGTGGGTGATTTTGTCT
GGACCGGTGGCGACACGCATCTGTACAGCAACCATATGGATCAAACCTCATCTGCAATTAAGCCGCGAA
CCGCGTCCGCTGCCGAAGTTGATTATCAAACGTAAACCCGAATCCATCTTCGACTACCGTTTCGAAGA
CTTTGAGATTGAAGGCTACGATCCGCATCCGGGCATTAAAGCGCCGGTGGCTATCTAATTACGAAACA
TCCTGCCAGAGCCGACGCCAGTGTGCGTCGGTTTTTTTACCCTCCGTTAAATTCTTCGAGACGCCTTC
CCGAAGGCGCCATTTCGCCATTTCAGGCTGCGCAACTGTTGGGAAGGGCGATCGGTGCGGGCCTCTTCGC
TATTACGCCAGCTGGCGAAAGGGGGATGTGCTGCAAGGCGATTAAAGTTGGGTAAACGCCAGGGTTTTCC
CAGTCACGACGTTGTAAACGACGGCCAGTGCCAAGCTTTCTTTAATGAAGCAGGGCATCAGGACGGT
ATCTTTGTGGAGAAAGCAGAGTAATCTTATTCAGCCTGACTGGTGGGAAACCACCAGTCAGAATGTGT
TAGCGCATGTTGACAAAAATACCATTAGTCACATTATCCGTCAGTCGGACGACATGGTAGATAACCTG
TTTATTATGCGTTTTGATCTTACGTTTAAATATTACCTTTATGCGATGAAACGGTCTTGGCTTTGATAT
TCATTTGGTCAGAGATTTGAATGGTTCCTGACCTGCCATCCACATTTCGCAACATACTCGATTTCGGTT
CGGCTCAATGATAACGTCGGCATATTTAAAAACGAGGTTATCGTTGTCTCTTTTTTTCAGAATATCGCC
AAGGATATCGTCGAGAGATTCCGGTTTTAATCGATTTAGAACTGATCAATAAATTTTTTCTGACCAATA
GATATTCATCAAAATGAACATTGGCAATTGCCATAAAAACGATAAATAACGTATTGGGATGTTGATTA
ATGATGAGCTTGATACGCTGACTGTTAGAAGCATCGTGGATGAAACAGTCCTCATTAATAAACACCAC
TGAAGGGCGCTGTGAATCACAAGCTATGGCAAGGTCATCAACGGTTTCAATGTGTTGATTTCTCTTT
TTTTAACCCCTCTACTCAACAGATAACCCGGTTAAACCTAGTCGGGTGTAACCTACATAAATCCATAATA
ATCGTTGACATGGCATACCTCACTCAATGCGTAACGATAAATCCCCTTACCTGAATATTTTCATCATG
ACTAAACGGAACAACATGGGTACCTAATGCGCCACTCTCGCGATTTTTTCAGGCGGACTTACTATCCC
GTAAAGTGTTGTATAATTTGCCTGGAATTGTCTTAAAGTAAAGTAAATGTTGCGATATGTGAGTGAGC
TTAAACAAATATTTTCGCTGCAGGAGTATCCTGGAAGATGTTCTGTAGAAGCTTACTGCTCACAAGAAA

AAAGGCACGTCATCTGACGTGCCTTTTTTATTTGTACTACCCTGTACGATTACTGCAGCTCGAGCTAG
TTAATCGGTACTTTGATCCATTCTTTCCGGGTACATGTCCGGGGTTTCTTTATGCTGGAACCAGCGACA
CGGCGCGATGACGATTTTCTCTTTACGTGGGTTCAGCCAAGCACCCACCAGGAGAACGTAGAATTAC
AGATAATGTGGTGACGACAGTGGCTCATCAGCATCATATCCTGCCAGCTGTCTTCGCCCTTGTTCAC
GTCACGTAGACCGCTTTCTTCAGCGGGATGTTTTCTTTAACCCAAGAGATATCATCAGAGAACACGTA
GTAGCTCGGGCCAGTAATACGGTTCTCCATTTCCGCGATCGCGTTCTTGTAATACGGCAGCTGGCACA
CGGAACCCGTGTTTTGCCAGTGACGCGGCAGCAGGTAGTACCACGGCGGATGTGGATAGAAACAGCT
TGGTCGTCAACTTCGATCTGTTTCAGCAGTTCCAGGCTTTCCGGGTAGCGATGTTTCAAGTTAAAGA
GAAGGCTTTACGAACGTCTCTTTGATATCGAAGAAGAAGCGTTTCAAGTGGTAGAAACCTTTAAAGT
ACAGCAGCGGCCAAAATAACGTTTTTCGTATGGATACAGAGTAGACGGGTCTGGCGACGTTCTGTAG
ATTTTTTTGAAGAACAGGAACCTCCAGGATTTTTTTCAGGGTACGGTTGATGCAAAATTCAGTCTGGCT
CAGGTCAAAGATACGGTTCATCTCATAACCGTTGTGAACTTTATAATGAACCATGTACAGACAGATCGA
TGTTCTGTATCCGGGTAAATGGTGTTCATTTTCAGGTAGAACCGGTAGATAAACATCTGGTTACCCAGG
CCGCCGATCATCTTGATCAGACGCATATGTATATCTCCTTCTTGAATTCTAAAAATTGATTGAATGTA
TGCAAAATAAATGCATACACCATAGGTGTGGTTTAAATTTGATGCCCTTTTTTCAGGGCTGGAATGTGTAA
GAGCGGGGTATTTATGCTGTTGTTTTTTTGTACTCGGGAAGGGCTTTACCTCTTCCGCATAAACGC
TTCCATCAGCGTTTATAGTTAAAAAATCTTTCGGAACCTGGTTTTTGCCTTACCCCAACCAACAGGGG
ATTTGCTGCTTTCCATTGAGCCTGTTTCTCTGCGCGACGTTCCGCGCGGCGTGTGTTGTGCATCCATCT
GGATTCTCCTGTCTAGTTAGCTTTGGTGGTGTGTGGCAGTTGTAGTCCTGAACGAAAACCCCCGCGAT
TGGCACATTGGCAGCTAATCCGGAATCGCACTTACGGCCAATGCTTCGTTTCGTATCACACACCCCAA
AGCCTTCTGCTTTGAATGCTGCCCTTCTCAGGGCTTAATTTTTAAGAGCGTCACCTTCATGGTGGTC
AGTGCGTCTGCTGATGTGCTCAGTATCACCGCCAGTGGTATTTATGTCAACACCGCCAGAGATAATT
TATCACCGCAGATGGTTATCTGTATGTTTTTTATATGAATTTATTTTTTGCAGGGGGGCATTGTTTGG
TAGGTGAGAGATCAATTCTGCATTAATGAATCGGCCAACGCGCGGGGAGAGGCGGTTTGGCGTATTGGG
CGCTCTTCCGCTTCCCTCGCTCACTGACTCGCTGCGCTCGTTCGCTCGGCTGCGGCGAGCGGTATCAGC
TCACTCAAAGGCGGTAATACGGTTATCCACAGAATCAGGGGATAACGCAGGAAAGAACATGTGAGCAA
AAGGCCAGCAAAAGGCCAGGAACCGTAAAAAGGCCGCTTGCTGGCGTTTTTCCATAGGCTCCGCCCC
CCTGACGAGCATCACAAAATCGACGCTCAAGTCAGAGGTGGCGAAACCCGACAGGACTATAAAGATA
CCAGGCGTTTTCCCCCTGGAAGCTCCCTCGTGCGCTCTCCTGTTCCGACCCTGCCGCTTACCGGATACC
TGTCCGCTTTCTCCCTTCGGGAAGCGTGGCGCTTCTCATAGCTCACGCTGTAGGTATCTCAGTTCTG
GTGTAGGTGTTTCGCTCCAAGCTGGGCTGTGTGCACGAACCCCCCGTTTACGCCGACCGCTGCGCCTT
ATCCGGTAACTATCGTCTTGAGTCCAACCCGGTAAGACACGACTTATCGCCACTGGCAGCAGCCACTG
GTAACAGGATTAGCAGAGCGAGGTATGTAGGCGGTGCTACAGAGTTCTTGAAGTGGTGGCCTAACTAC
GGCTACACTAGAAGAACAGTATTTGGTATCTGCGCTCTGCTGAAGCCAGTTACCTTCGGAAAAAGAGT
TGGTAGCTCTTGATCCGGCAAAACAAACACCGCTGGTAGCGGTGGTTTTTTTTGTTTGCAGCAGCAGA
TTACGCGCAGAAAAAAGGATCTCAAGAAGATCCTTTGATCTTTTCTACGGGGTCTGACGCTCAGTGG
AACGAAAACTCAGGTTAAGGGATTTTGGTTCATGAGATTATCAAAAAGGATCTTCACCTAGATCCTTTT
AAATTAATAATGAAGTTTTAAATCAATCTAAAGTATATATGAGTAACTTGGTCTGACAGTTACCAAT
GCTTAATCAGTGAGGCACCTATCTCAGCGATCTGTCTATTTTCGTTTCATCCATAGTTGCCTGACTCCCC
GTCGTGTAGATAACTACGATACGGGAGGGCTTACCATCTGGCCCCAGTGCTGCAATGATACCGCGAGA
CCCACGCTCACCAGCTCCAGATTTATCAGCAATAAACAGCCAGCCGGAAGGGCCGAGCGCAGAAGTG
GTCCTGCAACTTTATCCGCTCCATCCAGTCTATTAATTGTTGCGGGAAGCTAGAGTAAGTAGTTTCG
CCAGTTAATAGTTTTCGCAACGTTGTTGCCATTGCTACAGGCATCGTGGTGTACGCTCGTCTGTTGG
TATGGCTTCATTAGCTCCGGTTCCCAACGATCAAGGCGAGTTACATGATCCCCCATGTTGTGCAAAA
AAGCGTTAGCTCCTTCGGTCTCCGATCGTTGTGAGAAGTAAGTTGGCCGAGTGTTTACTCATG
GTTATGGCAGCACTGCATAATTCTCTTACTGTATGCCATCCGTAAGATGCTTTTCTGTGACTGGTGA
GTACTCAACCAAGTCATTCTGAGAATAGTGTATGCGGCGACCGAGTTGCTCTTGCCCGGCGTCAATAC
GGGATAATACCGCGCCACATAGCAGAACTTTAAAGTGCTCATCATTGGAAAACGTTCTTCGGGGCGA
AACTCTCAAGGATCTTACCGCTGTTGAGATCCAGTTTCGATGTAACCCACTCGTGACCCCACTGATC
TTCAGCATCTTTTACTTTTACCAGCGTTTCTGGGTGAGCAAAAACAGGAAGGCAAAATGCCGCAAAA
AGGGAATAAGGGCGACACGGAATGTTGAATACTCATACTCTTCTTTTCAATATTATTGAAGCAT
TATCAGGGTTATTGTCTCATGAGCGGATACATATTTGAATGTATTTAGAAAAATAAACAAATAGGGGT
TCCGCGCACATTTCCCCGAAAAGTGCCACCTGACGTCTAAGAAACCATTTATCATGACATTAACCT
ATAAAAATAGGCGTATCACGAGGCCCTTTCGTC (SEQ ID NO: 287)

The expression constructs were transformed into a host strain useful for the production of 2'-FL. Biosynthesis of 2'-FL requires the generation of an enhanced cellular pool of both lactose and GDP-fucose (FIG. 2). The wild-type *Eschericia coli* K12 prototrophic strain W3110 was selected as the parent background to test the ability of the candidates to catalyze 2'-FL production (Bachmann, B.J. (1972). Bacteriol Rev 36, 525-557). The particular W3110 derivative employed was one that previously had been modified by the introduction (at the *ampC* locus) of a tryptophan-inducible P_{trpB} *cI*⁺ repressor cassette, generating an *E.coli* strain known as GI724 (LaVallie, E.R. et al. (2000). Methods Enzymol 326, 322-340). Other features of GI724 include *lacIq* and *lacPL8* promoter mutations. *E.coli* strain GI724 affords economical production of recombinant proteins from the phage λ P_L promoter following induction with low levels of exogenous tryptophan (LaVallie, E.R. et al. (1993). Biotechnology (N Y) 11, 187-193; Mieschendahl, et al. (1986). Bio/Technology 4, 802-08). Additional genetic alterations were made to this strain to promote the biosynthesis of 2'-FL. This was achieved in strain GI724 through several manipulations of the chromosome using λ Red recombineering (Court, D.L. et al. (2002). Annu Rev Genet 36, 361-388) and generalized P1 phage transduction.

First, the ability of the *E. coli* host strain to accumulate intracellular lactose was engineered by simultaneous deletion of the endogenous β -galactosidase gene (*lacZ*) and the lactose operon repressor gene (*lacI*). During construction of this deletion, the *lacIq* promoter was placed immediately upstream of the lactose permease gene, *lacY*. The modified strain maintains its ability to transport lactose from the culture medium (via LacY), but is deleted for the wild-type copy of the *lacZ* (β -galactosidase) gene responsible for lactose catabolism. Therefore, an intracellular lactose pool is created when the modified strain is cultured in the presence of exogenous lactose. A schematic of the P_{lacIq} *lacY*⁺ chromosomal construct is shown in FIG. 12.

Genomic DNA sequence of the P_{lacIq} *lacY*⁺ chromosomal construct is set forth below (SEQ ID NO: 288):

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CACCATCGAATGGCGCAAAACCTTTTCGCGGTATGGCATGATAGCGCCCGGAAGAGAGTCAAGTGTAGGCTGGAGC
TGCTTCGAAGTTCCTATACCTTTCTAGAGAATAGGAACCTTCGGAATAGGAACCTAAGGAGGAT
ATTCATATGTACTATTTAAAAACACAACTTTTGGATGTTTCGGTATTCTTTTCTTTTACTTTTTTATCATG
GGAGCCTACTTCCCGTTTTTCCCGATTGGCTACATGACATCAACCATATCAGCAAAAGTGATACGGGTATTATT
TTTGCCGCTATTTCTCTGTTCTCGCTATTATCCAACCGCTGTTTGGTCTGCTTTCTGACAACTCGGGCTGCGC
AAATACCTGCTGTGGATTATTACCGGCATGTTAGTGATGTTTGGCGCGTTCTTTATTTTATCTTCGGGCCACTG
TTACAATACAACATTTTAGTAGGATCGATTGTTGGTGGTATTATCTAGGCTTTTGTTTTAAACGCCGGTGCGCCA
GCAGTAGAGGCATTTATTGAGAAAGTCAGCCGTCGCAGTAATTTTGAATTTGGTTCGCGCGCGGATGTTTGGCTGT
GTTGGCTGGGCGCTGTGTGCCTCGATTGTCGGCATCATGTTTACCATCAATAATCAGTTTGTCTGGCTGGGC
TCTGGCTGTGCACTCATCCTCGCGTTTTACTCTTTTTCGCCAAAACGGATGCGCCCTCTTCTGCCACGGTTGCC
AATGCGGTAGGTGCCAACCATTTCGGCATTTAGCCTTAAGCTGGCACTGGAACCTGTTTCTAGACAGCCAAAACCTGTGG
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TTTTTGTCACTGTATGTTATTGGCGTTTCTTGCACCTACGATGTTTTTGACCAACAGTTTGCTAATTTCTTTACT
 TCGTTCTTTTGCTACCGGTGAACAGGGTACGCGGGTATTTGGCTACGTAAACGACAATGGGCGAATTACTTAACGCC
 TCGATTATGTTCTTTGCGCCACTGATCATTAAATCGCATCGGTGGGAAAAACGCCCTGCTGCTGGCTGGCACTATT
 ATGTCTGTACGTATTATTGGCTCATCGTTCCGCACCTCAGCGCTGGAAGTGTTATTCTGAAAAACGCTGCATATG
 TTTGAAGTACCGTTCTGCTGGTGGGCTGCTTTAAATATATTACCAGCCAGTTTGAAGTGGCTTTTTCAGCGACG
 ATTTATCTGGTCTGTTTCTGCTTCTTTAAGCAACTGGCGATGATTTTTATGTCTGTACTGGCGGGCAATATGTAT
 GAAAGCATCGGTTTCCAGGGCGCTTATCTGGTGTGGGTCTGGTGGCGCTGGGCTTCACCTTAATTTCCGTGTTT
 ACGCTTAGCGGGCCCCGGCCGCTTTCCCTGCTGCGTCGTCAGGTGAATGAAGTCGCTTAAGCAATCAATGTCGGA
 TGCGGCGCGAGCGCCTTATCCGACCAACATATCATACGAGGTGATCGCATTGTAAATTATAAAAAATTGCCTGAT
 ACGTCTGCGCTTATCAGGCCTACAAGTTCAGCGATCTACATTAGCCGCATCCGGCATGAACAAAGCGCAGGAACAA
 GCGTCGCA

Second, the ability of the host *E. coli* strain to synthesize colanic acid, an extracellular capsular polysaccharide, was eliminated by the deletion of the *wcaJ* gene, encoding the UDP-glucose lipid carrier transferase (Stevenson, G. et al. (1996). J Bacteriol 178, 4885-893). In a *wcaJ* null background GDP-fucose accumulates in the *E. coli* cytoplasm (Dumon, C. et al. (2001). Glycoconj J 18, 465-474). A schematic of the chromosomal deletion of *wcaJ* is shown in FIG. 13.

The sequence of the chromosomal region of *E. coli* bearing the $\Delta wcaJ::FRT$ mutation is set forth below (SEQ ID NO: 289):

GTTTCGGTTATATCAATGTCAAAAACCTCACGCCGCTCAAGCTGGTGATCAACTCCGGGAACGGCGCAGCGGGTCC
 GGTGGTGGACGCCATTGAAGCCCGCTTTAAAGCCCTCGGCGCGCCCGTGAATTAATCAAAAGTGCAACACGCC
 GGACGGCAATTTCCCAACGGTATTCTTAACCCACTACTGCCGGAATGCCGCGACGACCCGCAATGCGGTCTAT
 CAAACACGGCGCGGATATGGGCATTGCTTTTGATGGCGATTTTGACCGCTGTTTCTGTTGACGAAAAAGGGCA
 GTTTATTGAGGGCTACTACATTGTGCGCCTGTTGGCAGAAGCATTCTCGAAAAAATCCCGCGCGAAGATCAT
 CCACGATCCACGTCTCTCTGGAACACCGTTGATGTGGTGACTGCCGAGGTGGCAGCGTAATGTGCAAAAC
 CGGACACGCCTTTATTAAAGAACGTATGCGCAAGGAAGACGCCATCTATGGTGGCGAAATGAGCGCCCACCATTA
 CTTCCGTGATTTTCGTTACTGCGACAGCGGCATGATCCCGTGGCTGCTGGTCCCGAAGTGGTGTGCTGAAAGA
 TAAACGCTGGGCGAACTGGTACGCGACCGGATGGCGGCGTTTCCGGCAAGCGGTGAGATCAACAGCAAACTGGC
 GCAACCCGTTGAGGCGATTAACCGCGTGGAACAGCATTTTAGCCGTGAGGCGCTGGCGGTGGATCGCACCGATGG
 CATCAGCATGACCTTTGCCGACTGGCGCTTTAACCTGCGCACCTCCAATACCGAACCAGGTGGTGCCTGAATGT
 GGAATCGCGCGGTGATGTGCCGCTGATGGAAGCGCGAAGCGCAACTCTGCTGACGTTGCTGAACGAGTAATGTG
 GATCTTCCCTTACCCCACTGCGGGTAAGGGGCTAATAACAGGAACAACGATGATTCCGGGGATCCGTGCACTGC
 AGTTCGAAGTTCTATTCTCTAGAAAAGTATAGGAACCTCGAAGCAGCTCCAGCCTACAGTTAACAAAAGCGGCATA
 TTGATATGAGCTTACGTGAAAAAACCATCAGCGGCGCAAGTGGTTCGGCGATTGCCACGTTGCGCCTGCTGG
 TCGGGCTGGTGCAGATGACCGTGCTGGCGCGGATTATCGACAACCACAGTTTCGGCCTGCTTACCGTGCTGCTGG
 TGATTATCGCGCTGGCAGATACGCTTTCTGACTTCGGTATCGCTAACTCGATTATTTCAGCGAAAAGAAATCAGTC
 ACCTTGAATCACCACGTTGTACTGGCTGAACGTCGGGCTGGGGATCGTGGTGTGCGGTGTTTTTGTGGA
 GTGATCTCATCGCGACGTGCTGAATAACCCGACCTGGCACCGTTGATTAAACATTATCGCTGGCGTTTGTGG
 TAATCCCCACGGGCAACAGTTCCGCGCGTTGATGCAAAAAGAGCTGGAGTTCAACAAAATCGGCATGATCGAAA
 CCAGCGCGGTGCTGGCGGGCTTCACTTGTACGGTGGTTAGCGCCCATTTCTGGCCGCTGGCGATGACCGCGATCC
 TCGGTTATCTGGTCAATAGTGCAGGTGAGAACGCTGCTGTTTGGCTACTTTGGCCGCAAAATTTATCGCCCCGGTC
 TGCATTTCTCGCTGGCGTCCGTGGCACCGAATTACGCTTTGGTGCCTGGCTGACGGCGGACAGCATCATCAACT
 ATCTCAATACCAACCTTCAACGCTCGTGCTGGCGCGTATTCTCGGCGCGGCGTGGCAGGGGGATACAACCTGG
 CGTACAACGTGGCCGTTGTGCCACCGATGAAGCTGAACCAATCATCACCCGCGTGTGTTTCCGGCATTTCGCCA
 AAATTCAGGACGATACCGAAAAGCTGCGTGTTAACTTCTACAAGCTGCTGTGCGTAGTGGGGATTATCAACTTTC
 CGGCGCTGCTCGGGCTAATGGTGGTGTGCAATAACTTTGTACCGCTGGTCTTTGGTGAGAAGTGAACAGCATTA
 TTCCGGTGTGCAATTGCTGTGTGTGGTGGTCTGCTGCGCTCCG

Third, the magnitude of the cytoplasmic GDP-fucose pool was enhanced by the introduction of a null mutation into the *lon* gene. Lon is an ATP-dependant intracellular

protease that is responsible for degrading RcsA, which is a positive transcriptional regulator of colanic acid biosynthesis in *E. coli* (Gottesman, S. & Stout, V. Mol Microbiol 5, 1599-1606 (1991)). In a *lon* null background, RcsA is stabilized, RcsA levels increase, the genes responsible for GDP-fucose synthesis in *E. coli* are up-regulated, and intracellular GDP-fucose concentrations are enhanced. The *lon* gene was almost entirely deleted and replaced by an inserted functional, wild-type, but promoter-less *E. coli lacZ*⁺ gene ($\Delta lon::(kan, lacZ^+)$). λ Red recombineering was used to perform the construction. A schematic of the *kan, lacZ*⁺ insertion into the *lon* locus is shown in FIG. 14.

Genomic DNA sequence surrounding the *lacZ*⁺ insertion into the *lon* region in the *E. coli* strain is set forth below (SEQ ID NO: 290):

```
GTGGATGGAAGAGGTGGAAGAAAGTGGTTATGGAGGAGTGGGTAATTGATGGTGAAAGGAAAGGGTTGGTGATTTA
TGGGAAGGGGGAAGGGGAAGAGGGATGTGGTGAATAATTAAGGATTGGGATAGAATTAGTTAAGGAAAAAGGGGG
GATTTTATGTGGGGTTTAAATTTTTGGTGTATTGTGGGGTTGAATGTGGGGGAAAGATGGGGATATAGTGAGGTA
GATGTTAATAGATGGGGTGAAGGAGAGTGGTGTGATCTGATTAGGTGGGGGAAATTAAGTAAGAGAGAGGTGTA
TGATTGGGGGATGGGTGGAGGTGGAGTTGGAAGTTGCTATTGTGTAGAAAAGTATAGGAAGTTGAGAGGGGTTTT
GAAGGTGAGGTTGGGGGAAGGAGTGAGGGGGGAAGGGGTGCTAAAGGAAGGGGAAGAGGTAGAAAGGGAGTGGGG
AGAAAGGGTGGTGAGGGGGGATGAATGTGAGGTAGTGGGCTATGTGGAGAAGGGGAAAGGGAAGGGGAAAGAGAA
AGGAGGTAGGTTGGAGTGGGGTTAGATGGGGATAGGTAGAGTGGGGGTTTTATGGAGAGGAAGGGGAAGGGGAAT
TGGGAGGTGGGGGGGGTGTGGTAAGGTTGGGAAGGGGTGGAAGTAAGTGGATGGGTTTGTGGGGGGAAGGA
TGTGATGGGGGAGGGGATGAAGATGTGATGAAGAGAGAGGATGAGGATGCTTTGGGATGATTGAAGAAGATGGAT
TGGAGGGAGGTTGTGGGGGGGTTGGGTGGAGAGGGTATTGGGGTATGAGTGGGGAGAAGAGAGAATGGGGTGGT
GTGATGGGGGGGTGTGGGGGTGTGAGGGGAGGGGGGGGGGGTGTGTTTTGTGAAGAGGGAGGTGTGGGGTGGGG
TGAATGAAGTGGAGGAGGAGGGAGGGGGGGTATGGTGGGTGGGAGGAGGGGGGTTGGTTGGGGAGGTGTGGTGG
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TTGGTGGTGGGAGAAAGTATGGATGTGATGGGTGATGGAATGGGGGGGGTGGATAGGGTTGATGGGGGTAGGTGGG
GATTGGAGGAGGAAGGGAAAGATGGGATGGAGGGAGGAGGTAGTGGGATGGAAGGGGGTGTGTTGGATGAGGATG
ATGTGGAGGAAGAGGATGAGGGGGTGGGGGAGGGGAAGTGTGGGGAGGGTGAAGGGGGGATGGGGGAGGGGGA
GGATGTGGTGGTGAGGGATGGGGATGGGTGGTTGGGGAATATGATGGTGGAAAATGGGGGGTTTTGTGGATTGAT
GGAGTGTGGGGGGTGGGTGTGGGGGAGGGGTATGAGGAGATAGGGTTGGGTAGGGGTGATATTGGTGAAGAGGT
TGGGGGGGAATGGGGTGGGGGTTGGTGGTGGTTTAGGGTATGGGGGGTGGGGATTGGGAGGGGATGGGGTTGTA
TGGGGTTGTTGAGGAGTTGTTGTAATAAGGGGATGTTGAAGTTGGTATTGGGAAGTTGGTATTGTGTAGAAAAGTA
TAGGAAGTTGGAAGGAGGTGGAGGGTAGATAAAGGGGGGGGTTATTTTTGAGAGGAGAGGAAGTGGTAATGGTAG
GGAGGGGGGGTGGAGTGAATTTGGGGGATAGTGAGGGGGTGGAGGAGTGGTGGGGAGGAATGGGGATATGGAAG
GGGTGGATATTGAGGGATGTGGGTGTTGGGGGTGGAGGAGATGGGGATGGGTGGTTTGGATGAGTTGGTGTGTA
GTGTAGGGGGTGTGTTGAAGTGAAGTGGGGGGGGAGTGGTGTGGGGGATAATTGAATTTGGGGGGTGGGGGAG
GGGAGAGGGTTTTGGGTGGGGAAGAGGTAGGGGGTATAGATGTTGAGAATGGGAGATGGGAGGGGTGAAAAGAGG
GGGGAGTAAGGGGGTGGGGATAGTTTTGTTGGGGGGTAAATGGGAGGGAGTTTAGGGGGTGTGGTAGGTGGGGGA
GGTGGGAGTTGAGGGGAATGGGGGGGGATGGGGTGTATGGGTGGGGAGTTGAAGATGAAGGTAATGGGGATTT
GAGGAGTAGGATGAATGGGGTAGGTTTTGGGGGTGATAAATAAGGTTTTGGGGTGTGGTGGGAGGGGTGAGGGG
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GTGAGTGGGGGTTAAATTTGGGAAGGGTTATTAGGGAGGTGGATGGAAGAAATGGATTTGGGTGGTGGTGGATGGG
GGATGGGGTGGGAGGGGGGGGGGAGGGTGAGAGTGAAGTTTTGGGGGAGAGGGGAGTGGTGGGAGGGGGTGTGTT
GGGGGGGTGTGAGGATGGGGTGGGGTTGGGTGGAGTAGGGGTAGTGTGAGGGAGAGTTGGGGGGGGGTGTGGG
GGTGGGGTAGTTGAGGGAGTTGAATGAAGTGTATTAGGTTGTGGAGGGAGATGGAGAGGGAGTTGAGGGGTGGGA
GGGGGTAGGATGGAGGGGGAGGATGGAGTGGAGGAGCTGCTTATGGGTATGAGGGAAGAGGTATTGGGTGGTGA
GTTGGATGGTTTTGGGGGATAAAGGGAAGTGGAAAAAGTGGTGGTGGTGTGTTTTGGTTGGGTGAGGGGTGGATGGG
GGGTGGGGTGGGGAAGAGGAGAGGGTTGATAGAGAACTGGGGATGGTTGGGGGTATGGGGAAGATGAGGGGGGT
AAGGGGAGGAGGGGTGGGGTTTTGATGATATTTAATGAGGAGCTGATGGAGGGAGTGGGAGAGGAAGGGGGGGT
GTAAAGGGGATAGTGAGGAAGGGGTGGGAGTATTTAGGGAAGGGGGAAGAGTGTAGGGATGGGGTGGGGGT
ATTGGGAAGGATGAGGGGGGGGTGTGTGGAGGTAGGGAAGGGATTTTTTGTGATGGAGGATTTGGGGAGAGGGG
GGAAGGGGTGGTGTGATGGAGGGGGGGTAGATGGGGGAAATAATATGGGTGGGGGTGGTGTGGGGTGGGGGGG
```

GTTGATAGTGGAGGGGGGGGAAGGATGGAGAGATTTGATGGAGGGATAGAGGGGGTGGTGATTAGGGGGGTGGG
 GTGATTGATTGGGGAGGGAGGAGATGATGAGAGTGGGGTGATTAGGATGGGGGTGGAGGATTGGGGTTAGGGGTT
 GGGTGATGGGGGGTAGGGAGGGGGGATGATGGGTGAGAGGATTGATTGGGAGGATGGGGTGGGTTTGAATATTGG
 GTTGATGGAGGAGATAGAGGGGGTAGGGGTGGGAGAGGGTGTAGGAGAGGGGATGGTTGGGATTAATGGGAAGAGG
 GGAGGGGGTTAAAGTTGTTGTGGTTGATGAGGAGGATATGGTGGAGGATGGTGTGGTGATGGATGAGGTGAGGAT
 GGAGAGGATGATGGTGGTGAGGGTTAAGGGGTGGAATGAGGAAGGGGTGGGGTTGAGGAGGAGGAGGAGGATTTT
 GAATGGGGAGGTGGGGGAAAGGAGATGGGAGGGTTGTGGTTGAATGAGGGTGGGGTGGGGGTGTGGAGTTGAA
 GGAGGGGAGGATAGAGATTGGGGATTGGGGGGTGGAGAGTTTGGGGTTTTGGAGGTTGAGAGGTAGTGTGAGGG
 GATGGGGATAAGGAGGAGGGTGATGGATAATTTGAGGGGGGAAAGGGGGGTGGGGTGGGGAGGTGGGTTTGGAG
 GGTGGGATAAAGAAAGTGTTAGGGGTAGGTAGTGAGGGAAGTGGGGGAGATGTGAAGTTGAGGGTGGAGTAGAG
 GGGGGGTGAAATGATGATTAAAGGGAGTGGGAAGATGGAAATGGGTGATTTGTGTAGTGGGTTTATGGAGGAAGG
 AGAGGTGAGGGAATAAGGGGTGATGGGGGAGATATGGTGATGTTGGAGATAAGTGGGGTGGTGGAGGGGAGGA
 GGATGAGGGGGAGGGGGTTTTGTGGGGGGGTAAAAATGGGGTGAGGTGAAATTGAGAGGGGAAAGGAGTGTGGT
 GGGGGTAAGGGAGGAGGGGGGGTTGGAGGAGAGATGAAAGGGGGAGTTAAGGGGATGAAAAATAATTGGGGTGT
 GGGGTTGGTGTAGGGAGGTTTGTGATGAAGATTAAATGTGAGGGAGTAAGAAGGGGTGGGATTGTGGGTGGGAAGAA
 AGGGGGGATTGAGGGTAATGGGATAGGTGAGGTGGTGTAGATGGGGGGATGGTAAGGGTGGATGTGGGAGTTTG
 AGGGGAGGAGGAGATATGGGGGTGAGGAAGATGGGAGGGAGGGAGGTTTGGGGGAGGGGTGTGGTGGGGGAAA
 GGAGGGAAAGGGGGATTGGGGATTGAGGGTGGGGAAGTGTGGGAAGGGGGATGGGTGGGGGGGTGTGGGTATT
 AGGGGAGGTGGGGAAAGGGGGATGTGGTGAAGGGGATTAAAGTTGGGTAAAGGGGAGGGTTTTGGGAGTGAGGAGG
 TTGTAAAAGGAGGGGGAGTGAATGGGTAATGATGGTGATAGTGGTTGGTGAGGTTGTGAGTGGAATAAGTGGA
 GGTGGGGGAAAATGGAGTAATAAAAAGAGGGGTGGGAGGGTAATTGGGGGTGGGAGGGTTTTTTTGTGTGGGTGA
 AGTTAGATGGGGGATGGGGGTGGGGTTATTAAGGGGTGTTGTAAGGGGATGGGTGGGGTGATATAAGTGGTGGG
 GGTTGGTAGGTTGAAGGATTGAAGTGGGATATAAATTATAAAGAGGAAGAGAAGAGTGAATAAATGTGAATTGAT
 GGAGAAGATTGGTGGAGGGGTGATATGTGTAAGGTGGGGGTGGGGGTGGGTGTAGATGGTATTATTGGTTGGGT
 AAGTGAATGTGTGAAAGAAGG

Fourth, a *thyA* (thymidylate synthase) mutation was introduced into the strain by P1 transduction. In the absence of exogenous thymidine, *thyA* strains are unable to make DNA and die. The defect can be complemented in *trans* by supplying a wild-type *thyA* gene on a multicopy plasmid (Belfort, M., Maley, G.F., and Maley, F. (1983). *Proc Natl Acad Sci U S A* 80, 1858-861). This complementation was used here as a means of plasmid maintenance.

An additional modification that is useful for increasing the cytoplasmic pool of free lactose (and hence the final yield of 2'-FL) is the incorporation of a *lacA* mutation. LacA is a lactose acetyltransferase that is only active when high levels of lactose accumulate in the *E. coli* cytoplasm. High intracellular osmolarity (e.g., caused by a high intracellular lactose pool) can inhibit bacterial growth, and *E. coli* has evolved a mechanism for protecting itself from high intra cellular osmolarity caused by lactose by "tagging" excess intracellular lactose with an acetyl group using LacA, and then actively expelling the acetyl-lactose from the cell (Danchin, A. *Bioessays* 31, 769-773 (2009)). Production of acetyl-lactose in *E. coli* engineered to produce 2'-FL or other human milk oligosaccharides is therefore undesirable: it reduces overall yield. Moreover, acetyl-lactose is a side product that complicates oligosaccharide purification schemes. The incorporation of a *lacA* mutation resolves these problems. Sub-optimal production of fucosylated oligosaccharides occurs in strains lacking either or both of the mutations in the colanic acid pathway and the lon protease. Diversion of lactose into a side product (acetyl-lactose) occurs in strains that do not contain the *lacA*

mutation. A schematic of the *lacA* deletion and corresponding genomic sequence is provided above (SEQ ID NO: 288).

The strain used to test the different $\alpha(1,2)$ FT candidates incorporates all the above genetic modifications and has the following genotype:

$\Delta amp^C::P_{trp}^B cI$, $\Delta(lacI-lacZ)::FRT$, $P_{lacI} lacY^+$, $\Delta wcaJ::FRT$, $thyA::Tn10$, $\Delta lon:(npt3, lacZ^+)$, $\Delta lacA$

The *E. coli* strains harboring the different $\alpha(1,2)$ FT candidate expression plasmids were analyzed. Strains were grown in selective media (lacking thymidine) to early exponential phase. Lactose was then added to a final concentration of 0.5%, and tryptophan (200 μ M) was added to induce expression of each candidate $\alpha(1,2)$ FT from the P_L promoter. At the end of the induction period (~24 h) equivalent OD 600 units of each strain and the culture supernatant was harvested. Lysates were prepared and analyzed for the presence of 2'-FL by thin layer chromatography (TLC).

A map of plasmid pG217 is shown in FIG. 11, which carries the *B. vulgatus* FutN.

The sequence of plasmid pG217 is set forth below (SEQ ID NO: 291):

TCTAGAATTCTAAAAATTGATTGAATGTATGCAAATAAATGCATACACCATAGGTGTGGTTTAATTTGATGCCCT
TTTTTCAGGGCTGGAATGTGTAAGAGCGGGTTATTTATGCTGTTGTTTTTTTGTACTCGGGAAGGGCTTTACCT
CTTCCGCATAAACGCTTCCATCAGCGTTTATAGTTAAAAAATCTTTCGGAACCTGGTTTTGCGCTTACCCCAACC
AACAGGGGATTTGCTGCTTTCCATTGAGCCTGTTTCTCTGCGCGACGTTGCGGGCGCGTGTGTTGTCATCCATC
TGGATTCTCCTGTCTAGTTAGCTTTGGTGGTGTGTGGCAGTTGTAGTCCTGAACGAAAACCCCGCGATTGGCAC
ATTGGCAGCTAATCCGGAATCGCACTTACGGCCAATGCTTCGTTTCGTATCACACACCCCAAGCCTTCTGCTTT
GAATGCTGCCCTTCTTCAGGGCTTAATTTTTAAGAGCGTCACCTTCATGGTGGTCAGTGCCTCTGCTGATGTGC
TCAGTATCACCGCCAGTGGTATTTATGTCAACACCGCCAGAGATAATTTATCACCGCAGATGGTTATCTGTATGT
TTTTTATATGAATTTATTTTTGTCAGGGGGGCGATTGTTTGGTAGGTGAGAGATCAATTCTGCATTAATGAATCGG
CCAACGCGCGGGGAGAGGCGGTTTTCGCTATTGGGCGCTCTTCCGCTTCCCTCGCTCACTGACTCGCTGCGCTCGGT
CGTTCCGCTGCGGCGAGCGGTATCAGCTCACTCAAAGGCGGTAATACGGTTATCCACAGAATCAGGGGATAACGC
AGGAAAGAACATGTGAGCAAAAGGCCAGCAAAAGGCCAGGAACCGTAAAAAGGCCGCGTTGCTGGCGTTTTTCCA
TAGGCTCCGCCCCCTGACGAGCATCAAAAAATCGACGCTCAAGTCAGAGGTGGCGAAAACCCGACAGGACTATA
AAGATACCAGGCGTTTCCCCCTGGAAGCTCCCTCGTGCCTCTCTGTTCCGACCTGCGCTTACCGGATACCT
GTCCGCTTTCTCCCTTCGGGAAGCGTGGCGCTTTCTCATAGCTCACGCTGTAGGTATCTCAGTTCCGTGTAGGT
CGTTCCGCTCCAAGCTGGGCTGTGTGCACGAACCCCCCGTTGAGCCGACCGCTGCGCTTATCCGGTAACATATCG
TCTTGAGTCCAACCCGGTAAGACACGACTTATCGCCACTGGCAGCAGCCACTGGTAACAGGATTAGCAGAGCGAG
GTATGTAGGCGGTGTACAGAGTTCTTGAAGTGGTGGCTAATACGGCTACACTAGAAGGACAGTATTTGGTAT
CTGCGCTCTGCTGAAGCCAGTTACCTTCGGAAGAGTTGGTAGCTCTTGATCCGGCAAAACAAACCCGCTGG
TAGCGGTGGTTTTTTTTGTTTGAAGCAGCAGATTACGCGCAGAAAAAAGGATCTCAAGAAGATCCTTTGATCTT
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CTTACCTAGATCCTTTTAAATTAAAAATGAAGTTTTAAATCAATCTAAAGTATATATGAGTAAACTTGGTCTGA
CAGTTACCAATGCTTAATCAGTGAGGCACCTATCTCAGCGATCTGTCTATTTCTGTTTCATCCATAGTTGCCTGACT
CCCCGTGCTGTAGATAACTACGATACGGGAGGGCTTACCATCTGGCCCCAGTGTGCAATGATACCGCGAGACCC
ACGCTCACCGGCTCCAGATTTATCAGCAATAAACAGCCAGCCGGAAGGGCCGAGCGCAGAAGTGGTCTGCAAC
TTTATCCGCTCCATCCAGTCTATTAATTGTTGCCGGAAGCTAGAGTAAGTAGTTCCGCGAGTTAATAGTTTGCG
CAACGTTGTTGCCATTGCTACAGGCATCGTGGTGTACGCTCGTCTGTTGGTATGGCTTCATTCAGCTCCGGTTC
CCAACGATCAAGGCGAGTTACATGATCCCCATGTTGTGCAAAAAAGCGTTAGCTCCTTCGGTCTCCGATCGT
TGTCAGAAGTAAGTTGGCCGCGAGTGTATCACTCATGGTTATGGCAGCACTGCATAATTCTCTTACTGTCTATGCC
ATCCGTAAGATGCTTTTCTGTGACTGGTGAGTACTCAACCAAGTCATTCTGAGAATAGTGTATGCGGCGACCGAG
TTGCTCTTGCCCGCGTCAATACGGGATAATACCGCGCCACATAGCAGAACTTTAAAAGTGCTCATCATTTGGAAA
ACGTTCTTCGGGGCGAAAACCTCTCAAGGATCTTACCGCTGTTGAGATCCAGTTCGATGTAACCCACTCGTGCACC
CAACTGATCTTCAGCATCTTTTACTTTCAACAGCGTTTCTGGGTGAGCAAAAACAGGAAGGCAAAATGCCGCAAA
AAAGGAATAAGGGCGACACGGAAATGTTGAATACTCATACTCTTCTTTTTCAATATTATTGAAGCATTTATCA
GGGTTATTGTCTCATGAGCGGATACATATTTGAATGTATTTAGAAAAATAAACAAATAGGGGTTCCGCGCACATT

TCCCCGAAAAGTGCCACCTGACGTCTAAGAAACCATTATTATCATGACATTAACCTATAAAAATAGGCGTATCAC
 GAGGCCCTTTTCGTCTCGCGCTTTTCGGTGATGACGGTGAAAACCTCTGACACATGCAGCTCCCGGAGACGGTCAC
 AGCTTGTCTGTAAGCGGATGCCGGGAGCAGACAAGCCCGTCAGGGCGCGTCAGCGGGTGTGGCGGGTGTCCGGG
 CTGGCTTAACATATGCGGCATCAGAGCAGATTGTACTGAGAGTGACCATATATGCGGTGTGAAATACCGCACAGA
 TCGCTAAGGAGAAAAATACCGCATCAGGCGCCTCCTCAACCTGTATATTCTGTAACACGCCCCAATGGGAGCTGTC
 TCAGGTTTGTTCCTGATTGGTTACGGCGCGTTTCGCATCATTGTTGAGTTTTTCCGCCAGCCGACGCGCAGTTT
 ACCGGTGCCTGGGTGCAGTACATCAGCATGGGGCAAATTCTTTCCATCCCCGATGATTGTGCGGGGTGTGATCATG
 ATGGTCTGGGCATATCGTCGCAGCCACAGCAACACGTTTCTTGAGGAACCATGAAACAGTATTTAGAAGTATGATG
 CAAAAAGTGCTCGACGAAGGCACACAGAAAAACGACCGTACCGGAACCGGAACGCTTTCCATTTTTGGTCATCAG
 ATGCGTTTTTAACCTGCAAGATGGATTCCCGCTGGTGACAATAACGTTGCCACCTGCGTTCCATCATCCATGAA
 CTGCTGTGGTTTTCTGCAGGGCGACACTAACATTGCTTATCTACAGAAAACAATGTACCATCTGGGACGAATGG
 GCGGATGAAAACGGCGACCTCGGGCCAGTGTATGGTAACAGTGGCGCGCCTGGCCAACGCCAGATGGTGTGTCAT
 ATTGACCAGATCACTACGGTACTGAACCAGCTGAAAAACGACCCGATTTCGCGCCGATTATTTGTTTTCAGCGTGG
 AACGTAGGCGAATGGATAAAATGGCGCTGGCACCCTGCCATGCATTCTTCCAGTTCTATGTGGCAGACGGCAAA
 CTCTCTTGCCAGCTTTATCAGCGCTCCTGTGACGTCTTCTCGGCCTGCGGTTCAACATTGGCAGCTACGCGTTA
 TTGGTGCATATGATGGCGCAGCAGTGCATCTGGAAGTGGGTGATTTTGTCTGGACCGGTGGCGACACGCATCTG
 TACAGCAACCATATGGATCAAATCATCTGCAATTAAGCCGCGAACCAGCGTCCGCTGCCGAAGTTGATTATCAAA
 CGTAAACCCGAATCCATCTTCGACTACCGTTTCGAAGACTTTGAGATTGAAGGTACGATCCGCATCCGGGCATT
 AAAGCGCCGGTGGCTATCTAATTACGAAACATCTTGCCAGAGCCGACCGAGTGTGCGTGGTTTTTTTACCCTC
 CGTTAAATCTTCGAGACGCCCTTCCCGAAGGCGCATTCGCCATTTCAGGCTGCGCAACTGTTGGGAAGGGCGATC
 GGTGCGGGCCTCTTCGCTATTACGCCAGCTGGCGAAAGGGGGATGTGCTGCAAGGCGATTAAGTTGGGTAAACGCC
 AGGGTTTTTCCAGTCACGACGTTGTAAAACGACGGCCAGTGCCAAGCTTCTTTAATGAAGCAGGGCATCAGGAC
 GGTATCTTTGTGGAGAAAGCAGAGTAATCTTATTCAGCCTGACTGGTGGGAAACCACAGTCAGAATGTGTTAGC
 GCATGTTGACAAAAATACCATTAGTCACATTATCCGTCACTCGGACGACATGGTAGATAACCTGTTTATTATGCG
 TTTTGATCTTACGTTTAAATATTACCTTTATGCGATGAAACGGTCTTGCGCTTTGATATTCAATTTGGTCAGAGATT
 GAATGGTTCCCTGACCTGCCATCCACATTCGCAACATACTCGATTTCGGTTCGGCTCAATGATAACGTCGGCATAT
 TTA AAAACGAGGTTATCGTTGTCTCTTTTTTTCAGAAATATCGCCAAGGATATCGTCGAGAGATTCCGGTTTAAATCG
 ATTTAGAAGTATCAATAAAATTTTTTCTGACCAATAGATATTATCAAAAATGAACATTGGCAATTGCCATAAAAA
 CGATAAATAACGTATTGGGATGTTGATTAAATGATGAGCTTGATACGCTGACTGTTAGAAGCATCGTGGATGAAAC
 AGTCCTCATTAATAAACACCACTGAAGGGCGCTGTGAATCACAAGCTATGGCAAGGTCATCAACGGTTTCAATGT
 CGTTGATTTCTCTTTTTTTTAAACCCCTCTACTCAACAGATACCCGGTTAAACCTAGTCGGGTGTAACATATAAT
 CCATAATAATCGTTGACATGGCATACCCCTCACTCAATGCGTAACGATAATTCCCCTTACCTGAATATTTTCATCAT
 GACTAAACGGAACAACATGGGTACCTAATGCGCCACTCTCGGATTTTTTCAGGCGGACTTACTATCCCGTAAAG
 TGTTGTATAATTTGCCTGGAATTGTCTTAAAGTAAAGTAAATGTTGCGATATGTGAGTGAGCTTAAAAACAATAT
 TTCGCTGCAGGAGTATCCTGGAAGATGTTTCGTAGAAGCTTACTGCTCACAAGAAAAAGGCACGTATCTGACGT
 GCCTTTTTTTATTTGTACTACCCTGTACGATTACTGCAGCTCGAGTTAGGATACCGGCACCTTTGATCCAACAGTC
 GGGTAGATATCCGGTGCTTCGGAGTGCTGGAACCAACGGCTCGGCACAATAACAGTCTTATCCATATTAGGGTTC
 AGCCAGGCACCCACCAAGAAAACGTGCTGTTACAAATGATGTGATGTTTGAATGAGACATCAGCATCATATCC
 TGCCAGGAGTCTTCATCAGTGTTCAGTCAATATAAACCGCATTCTGCAGTGGCAGATTTTCTTTAACCCACGCG
 ATATCGTCGGAGAAGATATAGTAAGATGGGCTAGCAACACGACGGGACATTTCCGCGATAGCATTCTGGTAATAC
 GGCAGCTGGCACACGGAACCGGTAGTAGCCAGTGTTCGGCTGCAGATAGTCACCACGACGAATGTGCAGGGAA
 ACCGCGTTTTTCATCTTTGTCCAGGATTTCCAGCATGTTTCAGGCTGCGGGAATTTGCTTTGTTCTTATCAAAGGTG
 AAGGATTCACGCACTTCGTCTTTGATATCAGCGAAGAAACGCTCGCTCTGATAGAAACCTTTAAAGTACAGCAGC
 GGCCAGAAATACTTCTTCTCGAACGCACGCAGAGAGTTTCGGCGCCTGCTTGCCTTCGTAGATTTTTTTTAAAAAAC
 AGGAATTCGATAACTTTTTTTCAGCGGTTGGTTGATGCAGAATTCGGTGTGCGGCAGGTTGAACACGCGGTGCATT
 TCGTAACCGTAATGGACTTTGTAATGCATCATGTGCTCAGGTCGATACGGACCTTCGGGTAATACTTTTTTCATA
 CGCAGATAGAAAGCATAGATAAACATCTGGTTGCCAGACCGCCAGTCACTTTGATCAGACGCATTATATCTCCT
 TCTTG

Fucosylated oligosaccharides produced by metabolically engineered *E. coli* cells are purified from culture broth post-fermentation. An exemplary procedure comprises five steps. (1) Clarification: Fermentation broth is harvested and cells removed by sedimentation in a preparative centrifuge at 6000 x g for 30 min. Each bioreactor run yields about 5-7 L of partially clarified supernatant. (2) Product capture on coarse carbon : A column packed with coarse carbon (Calgon 12x40 TR) of ~1000 ml volume (dimension 5 cm diameter x 60 cm

length) is equilibrated with 1 column volume (CV) of water and loaded with clarified culture supernatant at a flow rate of 40 ml/min. This column has a total capacity of about 120 g of sugar. Following loading and sugar capture, the column is washed with 1.5 CV of water, then eluted with 2.5 CV of 50% ethanol or 25% isopropanol (lower concentrations of ethanol at this step (25-30%) may be sufficient for product elution.) This solvent elution step releases about 95% of the total bound sugars on the column and a small portion of the color bodies. In this first step capture of the maximal amount of sugar is the primary objective. Resolution of contaminants is not an objective. (3) Evaporation: A volume of 2.5 L of ethanol or isopropanol eluate from the capture column is rotary-evaporated at 56 C° and a sugar syrup in water is generated. Alternative methods that could be used for this step include lyophilization or spray-drying. (4) Flash chromatography on fine carbon and ion exchange media: A column (GE Healthcare HiScale50/40, 5x40cm, max pressure 20 bar) connected to a Biotage Isolera One FLASH Chromatography System is packed with 750 ml of a Darco Activated Carbon G60 (100-mesh): Celite 535 (coarse) 1:1 mixture (both column packings were obtained from Sigma). The column is equilibrated with 5 CV of water and loaded with sugar from step 3 (10-50 g, depending on the ratio of 2'-FL to contaminating lactose), using either a celite loading cartridge or direct injection. The column is connected to an evaporative light scattering (ELSD) detector to detect peaks of eluting sugars during the chromatography. A four-step gradient of isopropanol, ethanol or methanol is run in order to separate 2'-FL from monosaccharides (if present), lactose and color bodies. Fractions corresponding to sugar peaks are collected automatically in 120-ml bottles, pooled and directed to step 5. In certain purification runs from longer-than-normal fermentations, passage of the 2'-FL-containing fraction through anion-exchange and cation exchange columns can remove excess protein/DNA/caramel body contaminants. Resins tested successfully for this purpose are Dowex 22.

The gene screening approach described herein was successfully utilized to identify new $\alpha(1,2)$ FTs for the efficient biosynthesis of 2'-FL in metabolically engineered *E. coli* host strains. The results of the screen are summarized in Table 1.

Production Host Strains

E. coli K-12 is a well-studied bacterium which has been the subject of extensive research in microbial physiology and genetics and commercially exploited for a variety of industrial uses. The natural habitat of the parent species, *E. coli*, is the large bowel of mammals. *E. coli* K-12 has a history of safe use, and its derivatives are used in a large

number of industrial applications, including the production of chemicals and drugs for human administration and consumption. *E. coli* K-12 was originally isolated from a convalescent diphtheria patient in 1922. Because it lacks virulence characteristics, grows readily on common laboratory media, and has been used extensively for microbial physiology and genetics research, it has become the standard bacteriological strain used in microbiological research, teaching, and production of products for industry and medicine. *E. coli* K-12 is now considered an enfeebled organism as a result of being maintained in the laboratory environment for over 70 years. As a result, K-12 strains are unable to colonize the intestines of humans and other animals under normal conditions. Additional information on this well known strain is available at <http://epa.gov/oppt/biotech/pubs/fra/fra004.htm>. In addition to *E. coli* K12, other bacterial strains are used as production host strains, e.g., a variety of bacterial species may be used in the oligosaccharide biosynthesis methods, e.g., *Erwinia herbicola* (*Pantoea agglomerans*), *Citrobacter freundii*, *Pantoea citrea*, *Pectobacterium carotovorum*, or *Xanthomonas campestris*. Bacteria of the genus *Bacillus* may also be used, including *Bacillus subtilis*, *Bacillus licheniformis*, *Bacillus coagulans*, *Bacillus thermophilus*, *Bacillus laterosporus*, *Bacillus megaterium*, *Bacillus mycoides*, *Bacillus pumilus*, *Bacillus lentus*, *Bacillus cereus*, and *Bacillus circulans*. Similarly, bacteria of the genera *Lactobacillus* and *Lactococcus* may be modified using the methods of this invention, including but not limited to *Lactobacillus acidophilus*, *Lactobacillus salivarius*, *Lactobacillus plantarum*, *Lactobacillus helveticus*, *Lactobacillus delbrueckii*, *Lactobacillus rhamnosus*, *Lactobacillus bulgaricus*, *Lactobacillus crispatus*, *Lactobacillus gasseri*, *Lactobacillus casei*, *Lactobacillus reuteri*, *Lactobacillus jensenii*, and *Lactococcus lactis*. *Streptococcus thermophiles* and *Propionibacterium freudenreichii* are also suitable bacterial species for the invention described herein. Also included as part of this invention are strains, modified as described here, from the genera *Enterococcus* (e.g., *Enterococcus faecium* and *Enterococcus thermophiles*), *Bifidobacterium* (e.g., *Bifidobacterium longum*, *Bifidobacterium infantis*, and *Bifidobacterium bifidum*), *Sporolactobacillus* spp., *Micromonospora* spp., *Micrococcus* spp., *Rhodococcus* spp., and *Pseudomonas* (e.g., *Pseudomonas fluorescens* and *Pseudomonas aeruginosa*).

Suitable host strains are amenable to genetic manipulation, e.g., they maintain expression constructs, accumulate precursors of the desired end product, e.g., they maintain pools of lactose and GDP-fucose, and accumulate endproduct, e.g., 2'-FL. Such strains grow well on defined minimal media that contains simple salts and generally a single carbon source. The strains engineered as described above to produce the desired fucosylated

oligosaccharide(s) are grown in a minimal media. An exemplary minimal medium used in a bioreactor, minimal "FERM" medium, is detailed below.

Ferm (10 liters): Minimal medium comprising:

40g $(\text{NH}_4)_2\text{HPO}_4$

100g KH_2PO_4

10g $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$

40g NaOH

1X Trace elements:

1.3g NTA (nitrilotriacetic acid)

0.5g $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$

0.09g $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$

0.09g $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$

0.01g $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$

0.01g $\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$

0.02g H_3BO_3

0.01g $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$ (pH 6.8)

Water to 10 liters

DF204 antifoam (0.1ml/L)

150 g glycerol (initial batch growth), followed by fed batch mode with a 90% glycerol-1% MgSO_4 -1X trace elements feed, at various rates for various times.

A suitable production host strain is one that is not the same bacterial strain as the source bacterial strain from which the fucosyltransferase-encoding nucleic acid sequence was identified.

Bacteria comprising the characteristics described herein are cultured in the presence of lactose, and a fucosylated oligosaccharide is retrieved, either from the bacterium itself or from a culture supernatant of the bacterium. The fucosylated oligosaccharide is purified for use in therapeutic or nutritional products, or the bacteria are used directly in such products.

EXAMPLES

Example 1: Identification of novel $\alpha(1,2)$ fucosyltransferases

To identify additional novel $\alpha(1,2)$ fucosyltransferases, a multiple sequence alignment query was generated using the alignment algorithm of the CLCbio Main Workbench package, version 6.9 (CLCbio, 10 Rogers Street #101, Cambridge, Massachusetts 02142, USA) using four previously identified lactose-utilizing $\alpha(1,2)$ fucosyltransferase protein sequences: *H. pylori* futC (SEQ ID NO: 1), *H. mustelae* FutL (SEQ ID NO: 2), *Bacteroides vulgatus* futN (SEQ ID NO: 3), and *E. coli* 0126 wbgL (SEQ ID NO: 4). This sequence alignment and percentages of sequence identity between the four previously identified lactose-utilizing $\alpha(1,2)$ fucosyltransferase protein sequences is shown in FIG. 3. An iterative PSI-BLAST was performed, using the FASTA-formatted multiple sequence alignment as the query, and the NCBI PSI-BLAST program run on a local copy of NCBI BLAST+ version 2.2.29. An initial position-specific scoring matrix file (.pssm) was generated by PSI-BLAST, which was then used to adjust the score of iterative homology search runs. The process is iterated to generate an even larger group of candidates, and the results of each run were used to further refine the matrix.

A portion of the initial position-specific scoring matrix file used is shown below:

Last position-specific scoring matrix computed																				
	A	R	N	D	C	Q	E	G	H	I	L	K	M	F	P	S	T	W	Y	V
1 M	-1	-1	-2	-3	-2	0	-2	-3	-2	1	2	-1	6	0	-3	-2	-1	-2	-1	1
2 A	2	-2	0	4	-2	-1	1	-1	-1	-2	-3	-1	-2	-3	-1	1	-1	-3	-3	-1
3 F	-2	-3	-3	-4	-3	-3	-3	-3	-1	0	0	-3	0	7	-4	-3	-2	1	3	-1
4 K	0	3	0	-1	-2	1	0	-1	-1	-3	-3	3	-2	-3	-1	2	0	-3	-2	-2
5 V	-1	-3	-3	-4	-1	-3	-3	-4	-3	4	2	-3	1	0	-3	-2	-1	-3	-1	3
6 V	-1	-3	-3	-3	-1	-3	-3	-4	-3	4	1	-3	1	-1	-3	-2	0	-3	-1	3
7 Q	-1	4	0	-1	-3	4	1	-2	0	-3	-2	3	-1	-3	-2	0	-1	-3	-2	-3
8 I	-1	-3	-3	-4	-1	-2	-3	-4	-3	3	2	-3	1	0	-3	-2	-1	-3	-1	3
9 C	-1	-1	0	-1	5	3	0	-2	4	-2	-2	0	-1	-2	-2	0	2	-2	-1	-1
10 G	0	-3	-1	-1	-3	-2	-2	6	-2	-4	-4	-2	-3	-3	-2	0	-2	-3	-3	-3
11 G	0	-3	-1	-1	-3	-2	-2	6	-2	-4	-4	-2	-3	-3	-2	0	-2	-3	-3	-3
12 L	-2	-2	-4	-4	-1	-2	-3	-4	-3	2	4	-3	2	0	-3	-3	-1	-2	-1	1
13 G	0	-3	-1	-1	-3	-2	-2	6	-2	-4	-4	-2	-3	-3	-2	0	-2	-3	-3	-3
14 N	-2	-1	6	1	-3	0	0	-1	1	-4	-4	0	-2	-3	-2	1	0	-4	-2	-3
15 Q	-1	1	0	0	-3	6	2	-2	0	-3	-2	1	-1	-3	-1	0	-1	-2	-2	-2
16 M	-1	-2	-3	-4	-2	-1	-2	-3	-2	1	3	-2	5	0	-3	-2	-1	-2	-1	1
17 F	-2	-3	-3	-4	-3	-3	-4	-3	-1	0	0	-3	0	7	-4	-3	-2	1	3	-1
18 Q	-1	0	-1	-1	-3	5	1	-2	0	1	-1	1	0	-2	-2	-1	-1	-2	-2	0
19 Y	-2	-2	-3	-3	-3	-2	-3	-3	1	-1	-1	-2	-1	5	-3	-2	-2	2	6	-1
20 A	4	-1	-1	-1	-1	-1	-1	0	-2	-2	-2	-1	-1	-2	-1	2	0	-3	-2	-1
21 F	-2	-3	-3	-4	-3	-3	-4	-3	-1	0	0	-3	0	7	-4	-3	-2	1	3	-1
22 A	3	-2	-1	-2	-1	-1	-1	4	-2	-2	-2	-1	-2	-3	-1	1	-1	-3	-2	-1
23 K	-1	0	-1	-2	-3	0	-1	-3	1	-2	-2	3	-1	2	-2	-1	-1	1	6	-2
24 S	2	-1	-1	-2	-1	-1	-1	-1	-2	-1	1	-1	0	-1	-1	3	0	-3	-2	0
25 L	-2	3	-2	-3	-2	-1	-2	-3	-2	1	3	0	1	0	-3	-2	-1	-2	-1	0
26 Q	0	0	0	-1	-2	4	1	-2	-1	-1	0	0	3	-2	-2	2	0	-2	-2	-1
27 K	-1	2	0	-1	-3	1	0	-2	-1	-2	-2	4	-1	-3	-1	0	2	-3	-2	-2
28 H	-1	0	0	-2	-3	0	0	-2	6	1	-1	2	-1	-1	-2	-1	-1	-3	0	0
29 S	-1	-1	3	-1	-2	-1	-1	-2	0	-1	1	-1	0	1	-2	1	0	0	4	-1
30 N	-1	-1	4	0	-3	-1	-1	3	0	-3	-3	-1	-2	0	-3	0	-1	-1	4	-3
31 T	-1	-2	-1	-2	-2	-1	-2	-2	-2	1	-1	-1	-1	-2	5	0	3	-3	-2	0
32 P	-1	0	-2	-1	-3	0	-1	-2	-2	-3	-3	2	-2	-4	7	-1	-1	-4	-3	-3
33 V	-1	-3	-3	-4	-1	-2	-3	-4	-3	2	2	-3	1	-1	-3	-2	0	-3	-1	4
34 L	-2	3	-2	-3	-2	-1	-2	-3	0	0	2	0	1	1	-3	-2	-1	0	4	-1
35 L	-2	-3	-4	-4	-2	-3	-3	-4	-3	3	3	-3	1	3	-3	-3	-1	-1	1	1
36 D	-2	-2	1	6	-4	0	1	-2	-1	-4	-4	-1	-3	-4	-2	0	-1	-5	-3	-4

The command line of PSI-BLAST that was used is as follows:

```
psiblast-db<LOCAL NR database name> -max_target_seqs 2500-in_msa<MSA file in FAST
format> -out <results output file> -outfmt "7sskingdoms sscinames scomnames sseqid stitle
eval length pident" -out_pssm<PSSM file output> -out_ascii_pssm<PSSM (ascii) output>
-num_iterations 6 -num_threads 8
```

This PSI-BLAST search resulted in an initial 2515 hits. There were 787 hits with greater than 22% sequence identity to FutC. 396 hits were of greater than 275 amino acids in length. Additional analysis of the hits was performed, including sorting by percentage identity to FutC, comparing the sequences by BLAST to an existing $\alpha(1,2)$ fucosyltransferase inventory (of known $\alpha(1,2)$ fucosyltransferases, to eliminate known lactose-utilizing enzymes and duplicate hits), and manual annotation of hits to identify those originating from bacteria that naturally exist in the gastrointestinal tract. An annotated list of the novel $\alpha(1,2)$ fucosyltransferases identified by this screen are listed in Table 1. Table 1 provides the bacterial species from which the enzyme is found, the GenBank Accession Number, GI Identification Number, amino acid sequence, and % sequence identity to FutC.

Multiple sequence alignment of the 4 known $\alpha(1,2)$ FTs used for the PSI-BLAST query and 12 newly identified $\alpha(1,2)$ FTs is shown in FIG. 4.

Example 2: Validation of novel $\alpha(1,2)$ FTs

To test for lactose-utilizing fucosyltransferase activity, the production of fucosylated oligosaccharides (*i.e.*, 2'-FL) is evaluated in a host organism that expresses the candidate enzyme (*i.e.*, syngene) and which contains both cytoplasmic GDP-fucose and lactose pools. The production of fucosylated oligosaccharides indicates that the candidate enzyme-encoding sequence functions as a lactose-utilizing $\alpha(1,2)$ fucosyltransferase. Of the identified hits, 12 novel $\alpha(1,2)$ fucosyltransferases were further analyzed for their functional capacity to produce 2'-fucosyllactose: *Prevotella melaninogenica* FutO, *Clostridium bolteae* FutP, *Clostridium bolteae* +13 FutP, *Lachnospiraceae sp.* FutQ, *Methanosphaerula palustries* FutR, *Tannerella sp.* FutS, *Bacteroides caccae* FutU, *Butyrivibrio* FutV, *Prevotella sp.* FutW, *Parabacteroides johnsonii* FutX, *Akkermansia muciniphilia* FutY, *Salmonella enterica* FutZ, and *Bacteroides sp.* FutZA.

Syngenes were constructed comprising the 12 novel $\alpha(1,2)$ FTs in the configuration as follows: EcoRI – T7g10 RBS – syngene – XhoI. FIG. 5A and FIG. 5B show the syngene fragments after PCR assembly and gel-purification.

The candidate $\alpha(1,2)$ FTs (*i.e.*, syngenes) were cloned by standard molecular biological techniques into an exemplary expression plasmid pEC2-(T7)-Fut syngene-rcsA-thyA. This plasmid utilizes the strong leftwards promoter of bacteriophage λ (termed P_L) to direct expression of the candidate genes (Sanger, F. et al. (1982). J Mol Biol 162, 729-773). The promoter is controllable, e.g., a *trp-cI* construct is stably integrated into the *E. coli* host's genome (at the *ampC* locus), and control is implemented by adding tryptophan to the growth media. Gradual induction of protein expression is accomplished using a temperature sensitive *cI* repressor. Another similar control strategy (temperature independent expression system) has been described (Mieschendahl et al., 1986, Bio/Technology 4:802-808). The plasmid also carries the *E. coli rcsA* gene to up-regulate GDP-fucose synthesis, a critical precursor for the synthesis of fucosyl-linked oligosaccharides. In addition, the plasmid carries a β -lactamase (*bla*) gene for maintaining the plasmid in host strains by ampicillin selection (for convenience in the laboratory) and a native *thyA* (thymidylate synthase) gene as an alternative means of selection in *thyA*⁻ hosts.

The expression constructs were transformed into a host strain useful for the production of 2'-FL. The host strain used to test the different $\alpha(1,2)$ FT candidates incorporates all the above genetic modifications described above and has the following genotype:

$\Delta ampC::P_{trp}^B cI$, $\Delta(lacI-lacZ)::FRT$, $P_{lacI} lacY^+$, $\Delta wcaJ::FRT$, *thyA::Tn10*, $\Delta lon:(npt3, lacZ^+)$, *AlacA*

The *E. coli* strains harboring the different $\alpha(1,2)$ FT candidate expression plasmids were analyzed. Strains were grown in selective media (lacking thymidine) to early exponential phase. Lactose was then added to a final concentration of 0.5%, and tryptophan (200 μ M) was added to induce expression of each candidate $\alpha(1,2)$ FT from the P_L promoter. At the end of the induction period (~24 h) the culture supernatants and cells were harvested. Heat extracts were prepared from whole cells and the equivalent of 0.2OD₆₀₀ units of each strain analyzed for the presence of 2'-FL by thin layer chromatography (TLC), along with 2 μ l of the corresponding clarified culture supernatant for each strain.

FIG. 6 shows the oligosaccharides produced by the $\alpha(1,2)$ FT-expressing bacteria, as determined by TLC analysis of the culture supernatant and extracts from the bacterial cells. 2'FL was produced by exogenous expression of WbgL (used as control), FutO, FutP, FutQ, FutR, FutS, FutU, FutW, FutX, FutZ, and FutZA.

Table 4 summarizes the fucosyltransferase activity for each candidate syngene as determined by the 2'FL synthesis screen described above. 11 of the 12 candidate $\alpha(1,2)$ FTs were found to have lactose-utilizing fucosyltransferase activity.

Table 4. 2'FL synthesis screen results

	syngene	24h OD (induced)	2'-FL culture medium	2'-FL cell extract	
<i>Escherichia coli</i>	WbgL	9.58	5	5	pG204 pEC2-WbgL-rcsA-thyA E640
<i>Prevotella melaninogenica</i>	FutO	12.2	3	2	pG393 pEC2-(T7)FutO-rcsA-thyA E985
<i>Clostridium bolteae</i>	FutP	10.4	1	2	pG394 pEC2-(T7)FutP-rcsA-thyA E986
<i>Lachnospiraceae sp.</i>	FutQ	10.6	3	4	pG395 pEC2-(T7)FutQ-rcsA-thyA E987
<i>Methanosphaerula palustris</i>	FutR	11.9	0	1	pG396 pEC2-(T7)FutR-rcsA-thyA E988
<i>Tannerella sp.</i>	FutS	11.3	2	3	pG397 pEC2-(T7)FutS-rcsA-thyA E989
<i>Bacteroides caccae</i>	FutU	12.1	0	2	pG398 pEC2-(T7)FutU-rcsA-thyA E990
<i>Butyrivibrio</i>	FutV	11.3	0	1	pG399 pEC2-(T7)FutV-rcsA-thyA E991
<i>Prevotella sp.</i>	FutW	10.5	3	3	pG400 pEC2-(T7)FutW-rcsA-thyA E992
<i>Parabacteroides johnsonii</i>	FutX	10.7	3	5	pG401 pEC2-(T7)FutX-rcsA-thyA E993
<i>Akkermansia muciniphila</i>	FutY	9.1	0	0	pG402 pEC2-(T7)FutY-rcsA-thyA E994
<i>Salmonella enterica</i>	FutZ	11.0	0	3	pG403 pEC2-(T7)FutZ-rcsA-thyA E995
<i>Bacteroides sp.</i>	FutZA	9.9	3	3	pG404 pEC2-(T7)FutZA-rcsA-thyA E996

Example 3: Characterization of cultures expressing novel $\alpha(1,2)$ FTs

Further characterization of the bacterium expressing novel $\alpha(1,2)$ FTs FutO, FutQ, and FutX was performed. Specifically, proliferation rate and exogenous $\alpha(1,2)$ FT expression was examined.

Expression plasmids containing fucosyltransferases WbgL (plasmid pG204), FutN (plasmid pG217), and novel $\alpha(1,2)$ FTs FutO (plasmid pG393), FutQ (plasmid pG395), and FutX (pG401) were introduced into host bacterial strains. For example, the host strains utilized has the following genotype: $\Delta ampC::P_{trp}^B cI$, $\Delta(lacI-lacZ)::FRT$, $P_{lacI} lacY^+$, $\Delta wcaJ::FRT$, $thyA::Tn10$, $\Delta lon:(npt3, lacZ^+)$, $\Delta lacA$

Bacterial cultures expressing each exogenous fucosyltransferase were induced by addition of tryptophan (to induce expression of the exogenous fucosyltransferases) in the presence of lactose. Growth of the cultures was monitored by spectrophotometric readings at A600 at the following timepoints: 4 hours and 1 hour before induction, at the time of induction (time 0), and 3 hours, 7 hours, and 24 hours after induction. The results are shown in FIG. 7, and indicate that expression of the exogenous fucosyltransferase did not prevent cell proliferation. Furthermore, the growth curve for the bacterial cultures expressing the novel $\alpha(1,2)$ fucosyltransferases FutO, FutQ, and FutX is similar to those expressing the known $\alpha(1,2)$ FT enzymes WbgL and FutN.

Protein expression was also assessed for the bacterial cultures expressing each fucosyltransferase after induction. Cultures were induced as described previously, and

protein lysates were prepared from the bacterial cultures at the time of induction (0 hours), 3 hours, 7 hours, and 24 hours after induction. The protein lysates were run on an SDS-PAGE gel and stained to examine the distribution of proteins at each time point. As shown in FIG. 8, induction at 7 hours and 24 hours showed increases in a protein band at around 20-28 kDa for bacterial cultures expressing exogenous FutN, FutO, and FutX. These results indicate that induction results in significant expression of the exogenous fucosyltransferases.

Finally, additional TLC analysis to assess the efficiency and yield of 2'FL production in bacterial cultures expressing novel $\alpha(1,2)$ FTs FutO, FutQ, and FutX compared to known fucosyltransferases WbgL and FutN. Cultures were induced at 7 hours and 24 hours, and run out on TLC. FIG. 9A shows the level of 2'FL in the cell supernatant. The level of 2'FL found in the bacterial cells were also examined. As shown in FIG. 9B, 2'FL was produced in cell lysates from bacteria expressing the novel $\alpha(1,2)$ FTs FutO, FutQ, and FutX at 7 hours and 24 hours after induction.

Example 4: FutN exhibits increased efficiency for production of 2'FL

Fucosylated oligosaccharides produced by metabolically engineered *E. coli* cells to express *B. vulgatus* FutN was purified from culture broth post-fermentation.

Fermentation broth was harvested and cells were removed by sedimentation in a preparative centrifuge at 6000 x g for 30 min. Each bioreactor run yields about 5-7 L of partially clarified supernatant. A column packed with coarse carbon (Calgon 12x40 TR) of ~1000 ml volume (dimension 5 cm diameter x 60 cm length) was equilibrated with 1 column volume (CV) of water and loaded with clarified culture supernatant at a flow rate of 40 ml/min. This column had a total capacity of about 120 g of sugar. Following loading and sugar capture, the column is washed with 1.5 CV of water, then was eluted with 2.5 CV of 50% ethanol or 25% isopropanol (lower concentrations of ethanol at this step (25-30%) may be sufficient for product elution.) This solvent elution step released about 95% of the total bound sugars on the column and a small portion of color bodies (caramelized sugars). A volume of 2.5 L of ethanol or isopropanol eluate from the capture column was rotary-evaporated at 56 C° and a sugar syrup in water was generated. A column (GE Healthcare HiScale50/40, 5x40cm, max pressure 20 bar) connected to a Biotage Isolera One FLASH Chromatography System was packed with 750 ml of a Darco Activated Carbon G60 (100-mesh): Celite 535 (coarse) 1:1 mixture (both column packings were obtained from Sigma). The column was equilibrated with 5 CV of water and loaded with sugar from step 3 (10-50 g, depending on the ratio of 2'-FL to contaminating lactose), using either a celite loading

cartridge or direct injection. The column was connected to an evaporative light scattering (ELSD) detector to detect peaks of eluting sugars during the chromatography. A four-step gradient of isopropanol, ethanol or methanol was run in order to separate 2'-FL from monosaccharides (if present), lactose and color bodies. Fractions corresponding to sugar peaks were collected automatically in 120-ml bottles, pooled.

The results from two fermentation runs are shown in FIG. 10A and FIG. 10B. The cultures were grown for 136 (run 36B) or 112 hours (run 37A), and the levels of 2'-FL produced was analyzed by TLC analysis. As shown in both FIG. 10A and FIG. 10B, the 2'-fucosyllactose was produced at 40 hours of culture, and production continued to increase until the end point of the fermentation process. The yield of 2'-FL produced from run 36B was 33 grams per liter. The yield of 2'-FL produced from run 37A was 36.3 grams per liter. These results indicate that expression of exogenous FutN is suitable for high yield of 2'-fucosyllactose product.

Table 1. Hits from PSI-BLAST multiple sequence alignment query for novel $\alpha(1,2)$ fucosyltransferases

Bacterium names	Accession No.	GI No.	Gene name [bacterium]	% identity FutC	Alias	SEQUENCE	WO 2015/175801
Helicobacter pylori	AAD29869.1	4808599	alpha-1,2-fucosyltransferase [Helicobacter pylori]	98	FutC	MAFKVQICGGLGNQMFQYAFKSLQKHSNTPVLLDITSDWSDRKMQLLELPINLPYASAKELIAKMQHLPKLVRLDALKCMGFDRVSQEIVFEYPELLKPSRLTYFYGYFQDPQRYFDAQISPLIKQTFTLPPPPENNNKKEEYHRKLSLILAAKNSVFHRRGDYVIGCQGLIDYQKKALEYMAKRVPMELFVFCEDLEFTQNLDLGYPFMDMTTRNKEEEAYWDMLLMQSCQHGIANSTYSWWAAAYLIENPEKIIIGPKHWLFGHENILCKEAWVKIESHFEVKSQKYNA	1
Helicobacter mustelae; Helicobacter mustelae 12198	YP_003517185.1	291277413	alpha-1,2-fucosyltransferase [Helicobacter mustelae 12198]	70.85	FutL	MDFKIVQVHGGGLGNQMFQYAFKSLQTHLNIPVLDTTWFDYGNRELGLHLFPIDLQCASAAQIAAHMQNLPLVRGALRRMGLGRVSKSEIVFEYMPPELFPSRIAYFHGYFQDPQRYFEDISPLIKQTFTLPHPTHEAEQYSRKLSQLAAKNSVFHRRGDYMRGLGWQLDISYQLRAIAYMAKRVQNLELFLFCEDLEFVQNLDLGYPFVDMTTRDGAHAWDMMLMQSCKHGIITNSTYSWWAAAYLIKPEKIIIGPSSHVIYGNENILCKDWVKIESQFETKS	2
Bacteroides; Bacteroides vulgatus ATCC 8482; Bacteroides sp. 4_3_47FAA; Bacteroides sp. 3_1_40A; Bacteroides vulgatus PC510; Bacteroides vulgatus CLO9T03C04; Bacteroides vulgatus dn1.KV7; Bacteroides vulgatus CAG:6	YP_001300461.1	150005717	glycosyl transferase family protein [Bacteroides vulgatus ATCC 8482]	24.83	FutN	MRLIKVTGGLGNQMFYAFYLRMKKYYPKVRIDLSMMHYKVHYGYEMHRVFNLPHTFCINQPLKKVIEFLFFKKIYERKQAPNSLRAFEKKYFWPLLYFKGFYQSEFFADIKDEVRESFTFDKNKANSRLNMLELKDENA VSLHIRRGDYLPKHWATTGSVCQLPYYQNAIAEMSRRAVSPSYIFSDDIAWVVKENLPLQNAVVIDWNTD EDSWQDMMLMSHCKHHIICNSTFSWWGAWLNPNMDKTVIVPSRWFOHSEAPDIYPTGWIKVPVS	3
Escherichia coli; Escherichia coli UME4 3065-1	WP_021554465.1	545259828	protein [Escherichia coli]	23.13	WbgL	MSIIRLQGGGLGNQLFOFSFGYALS KINGTPLYFDISHYAENDDHGGYRLNNLQIPEEYLYQYTPKINNYKLLVRGSRLYPDIFLGLFCNEFHAYGYDFEYIAQKWKKYGYWQSEHFFHKHILDLKEFFPKNVSEQANLLAAKILESQSSLSIHRRGDYIKNKTATLTHGVCSLEYKKALNKIRDLAMIRDVFISDDIFWCKENIETLLSKKYNYYSEDL SQEEDLWMLSLANHHIIANSFSWWGAYLGSASSQVIYPTPWYDITPKNTYIPVNHWINVDKHS	1

PCT/US2015/030823

Clostridium bolteae; Clostridium bolteae 90A9; Clostridium bolteae 90B3; Clostridium bolteae 90B8	WP_00257 0768.1	488634090	protein [Clostridium bolteae]	29.86	FutP	MFQYALYKAFEQKHIDVYADLAWYKNKSVKFELYNFGIKINVASEKDINRLSDCQADFVSRIRRKIFGKKKSFV SEKNDSCYENDILRMDNVYLSGYWQTEKYFSNTREKLEDYSFALVNSQVSEWDSIRNKNKSVSIHIRG DYL QGELYGIGCTSLYAEAEIYKMRVPNAKFFVFSDDVWVKQQEDFKGFVIVDRNEYSSALS DMYLMSLCKH NIANSFSFWAAWLNREKEKVIAPRRWLNGKCTPDIWCKKWIRI	WO 2015/175801
	WP_00925 1343.1	496545268	protein [Lachnospiraceae bacterium 3_1_57FAA_CT1]	29.25	FutQ	MVIVQLSGGLGNQMFEYALYLSLKAKGKEVKIDDDVTCEGPGTRPQLDVFQITYDRASREELTEMTDASM DALSRVRRKLTGRRTKAYRERDINFDP LVMKDPALLEGCFQSDKYFRDCEGRVREAYRFRGIESGAFPLPED YLRLKQIEDCQSVSHIRRGDYLDESHGGLYTIGICTEAYYKEAFARMERLVPGARFLLFSNDPEWTRHFFES KNCVLVEGSTEDTGYMDLYLMSRCRHHIANSFSFWGAWLNENPEKKVIAPAKWLNRECRDITYTERMI RL	12
Methanospiraerula palustris; Methanosp haerula palustris E1- 9C	YP_002467 213.1	219852781	glycosyl transferase family protein [Methanospiraceae Methanospiraerula palustris E1- 9C]	28.52	FutR	MIIVRLKGGGLGNQSQYALGRKIAHLHNTLKLDTTWFTTSSDTPTRYRLNNYNIIGTIAAKEIQLIERGRAQ GRGYLLSKISDILLTPMYRRTYVRRMHTFDKAILTYPDNVYLDGYWQTEKYFKDIEILRREVTLKDEPD SINLE MAERIQACHSVSLHVRRGDYVSNPTTQQFHGCCSIDYNNRAISLIEEKVDPSFFIFSDLPWAKENLDIPGE KTFVAHNGPEKEYCDLWMLSLCQHIIANSFSFWGAWLGGQDAEKMVIAPRRWALSESFDTSDIIPDSWI TI	13
	WP_02192 9367.1	547187521	glycosyl transferase family 11 [Tannerella sp. CAG:118]	28.38	FutS	MVRIVEIIGGLGNQMFOYAFSLYLKNSHWDRLVYDIEAMKTYDRHYGLEKLVFNLSLCPISNRLHRNLQK RSFAKHVKSLEYHSECEDEPVRGLRPRYRYRGYVWQNEGYFVDIEPMIREAFQFNVNLSKTKAIASKMIR RELSVSIHVRRGDYENLPEAKAMHGGICSLDYYHKAIDFIRQLDNNICFYLFSDDINWVEENLQLENRCID WNOGEDSWQDMYLMSCCRHHIANSFSFWAAWLNPNKNKIVLTPNKWFNHTDAVGIVPKSWIKIPVF	14
Bacteroides cacciae; Bacteroides cacciae ATCC 43185	WP_00567 5707.1	491925845	protein [Bacteroides cacciae]	28.09	FutU	IMKIVKILGGLGNQMFOYALFSLKERFPHEQVMIIDTSCERNYPLHNGFEVDRIFAQKAPVASWRNLIKVAYP YPNYRFWKIGYILPKRKTMCVERKNFSFDDAVALTRKGDYDGYWQHEEYFCDMKETIWEAFSPPEVDG RNKEIGALLOASDSASLHVRRGDYVNHPLFRGICDLDYKRAIHMYMEERVNPQLYCVFSNDMAWCESH LRA LLPGKEVVYVDWNKGAEVYVDMRLMSLCRHHIANSFSFWGAWLNRRNPQKVVVAPERWVMSPIEDPV SDKWIKL	15
	WP_02277 2718.1	551028636	protein [Butyrivibrio sp. AE2015]	27.8	FutV	MIIILKGGGLGNQMFOYALYKSLKRGKEVKIDDKTGTVNDKLRIPVLSRWGVEYDRATDEEINLTD SKMDL FSRIRRLKLTGRKTRIDEESGKFNPEILEKENAYLVGYWQCDKYFDKDDKDVVREIREAFKKPQELMTDASSWS TLQQIECCESVSLHYVRTDYVDEEHHIHNICTEKYKNAIDRVKQYPSAVFFITDDKEW/CRDHFKGPNFV VELEEGDGTDAEMTLMSRCKHHIANSFSFWAAWLNDSPEKVIAPQKWINNRDMDDITYTERMTKIAL	6

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Prevotella sp. CAG:891	WP_02248 1266.1	548264264	uncharacterized protein [Prevotella sp. CAG:891]	27.4	FutW	MRLVKMIGGLGNQMFIYAFYLQMRKFSNVRLDITDMMHYNVHYGYELHKVFLGRLPTEFCMNQPLKKVL EFLFRTIVERKQHGRLMEPYTCQYVWPLVYFKGYQSERVSEVKDEVRECFTFNPALANRSSQQMMEQIQ NDPOAVSIHRRGDYLNPKHYDTIGICQLPYKHAVSEIKKVSNPVSELDWVYKANLPLENAQYIDW NKGADSWQDMMLMSCCKHHIICNSTFSWAAWLNPSVEKTVIMPEQWTSRQSDVDFVASCGRWVRV KTE	WO 2015/175801
Parabacteroides johnsonii;Parabacter oides johnsonii CLO212C29	WP_00815 5883.1	495431188	glycosyl transferase [Parabacteroides johnsonii]	26.69	FutX	MRLKIMIGGLGNQMFIYAFYLKMKHHYPDTNIDLSDMVHYKVHNGYEMNRIFDLSQTEFCINRTLKILEFL FFKKIYERRQDPSTLYPEYKRYFWPLLYFKGYQSERFFEDIKDDVRKAFSFLNIANPESLELLKQIEVDDQAV SIHRRGDYLLPRHWANTGSVCQLPYKNAIAEMENRITGPSYVFSDDISWVKENIPLKKAIVVVTWNKGED SWQDMMLMSHCRHHIICNSTFSWAAWLNPRKEKVIAPCRWFQHKETPDMPYKWEIKVPIN	18
Akkermansia muciniphila;Akkermansia muciniphila ATCC BAA-835	YP_001877 555.1	187735443	glycosyl transferase family protein [Akkermansia muciniphila ATCC BAA-835]	25.67	FutY	MRLFGGLGNQLFOYAFALSRQGGKARLETSSYEHDDKRVCELHFRVSLPIEGGPPPWAFKRSRIPACLR LFAAPKYPHFREERKRGDPGLAAPPRRHTYFKGYQTEQYFLHCREQLCREFLKTLPTPENARILEDIRSCS ISLHRRDYLNSPILSPPLLEYLRSMAEMEGRLRAAGAPQESLRYFIFSDIEWARQNLRLPALPHVHVIND GGTGYFDLELMRNCRHIIICNSTFSWAAWLNHAEKVIAPRIWFRNREEGDRYHTDDALIPGSWLRI	19
Salmonella enterica;Salmonella enterica subsp. enterica serovar Poona str. ATCC BAA- 1673	WP_02321 4330.1	555221695	fucosyltransferase [Salmonella enterica]	25.99	FutZ	MYSCLSGGLGNQMFIYAFYAAAYILKQYFOSSTLLVDDSYYSQPKRDTVRSLNQNFIYSDFSADEKEIKLL RKFRNPFPKQISEILSALFGKALSDRAFYTFETIKNIDKACLFQYQDADLLNKYQKILPLFLRLRDDLLDICKN LELYSLIORSNNTTALHRRGDYVTNQHAAYHGVLDISYNNHAMEYERERQKQNFIFSDVVRWAQKAF ENDNCYVINNSDYDFSADIMYLSLCKNNIIANSTYSWWGAWLNKYVEDKLVSPKQWFLGNNETSLRNAS WITL	20
Bacteroides sp. CAG:633	WP_02216 1880.1	547748823	glycosyltransferase family 11 [Bacteroides sp. CAG:633]	26.01	FutZ A	MRLKIMITGGLGNQMFIYAFYLRMKKRYPKVRIDLSDMVHYVHHGYEMHRVFNLPHTFCINQPLKKVIEF LFFKKIYERKQDPNSLRFAFEKKYLWPLLYFKGYQSERFFADIKDEVKRAFTDSSKVNARSALLRLDADAN AVSLHRRGDYLPQHWATTGSVCQLPYQNAIAEMNRRAVAPSYVFSDDIAWVKENIPLQNAVVIDWN KGEESWQDMMLMSHCRHHIICNSTFSWAAWLNPHEDKIVIPNRWVFQHCETPNIPYAGWVKVAIN	21
Clostridium sp. CAG:306	WP_02224 7142.1	547839506	alpha-1,2- fucosyltransferase [Clostridium sp. CAG:306]	34.28		MEKIKIVKLOGGMGNQMFIYAFQAFGKGLSKFGCKVLFDKINYDELQKTIINNTGKNAEGICVRKYELGIFNLNI DFATAEQICQEGEKLKACALPGFIRKIFNLKNTVSNRFEKKYGEYDEELKDYSLAYDGYQNPYKFEDI SDKIKKEFTLPEIKNHDYNNKLEKIQFENSFVHVRDDYLNINCEIDLDDYQKAVKYILKHENPKFFVCAE DPDYIKNHFDIGYDFELVGENNKTQDTYENMRLMMACKHAIANSSYSWWAAWLSDDYDNKIVIAPTPWL PGISNEIICKNWIIQIRGISNE	2

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Prevotella sp. oral taxon 306; Prevotella sp. oral taxon 306 str. F0472	WP_00943 4595.1	497004957	protein [Prevotella sp. oral taxon 306]	32.11		MKIVKILGGLGNQMFQYALYLSLQSEFPKERVALLSCFNGLHNGFELERIFSLTAQKASATIMRIAYYP NYLLWRIGKRLLPRKTMCLLESSTFRYDESVLTREGNRYFDGYWQDERYFVACREKVLKAFITPAFKRTENLS LLRKLDKNSVAIHVRGDIYGNQYQIGICDLDYRAAIDKISTYVTPSVFCISFNDAWQCQTHLOPYLKAPVY VTWNTGTESYRDMQLMSCCAHNIAANSFSSWWGAWLNNQNEKVVIAPKRWLNMDDCCQPLPASWVKI	WO 2015/175801
Brachyspira sp. CAG:484	WP_02191 7109.1	547139308	glycosyl transferase family 11 [Brachyspira sp. CAG:484]	30.14		MLVVKLMGGLGNQMFQYAFKALGDKNILFYGDYKKHSLRKVEINRFFCKAVYIPRELKYLKVFVKFKDKIE YMRSGYVPEYLRDGNHYYIGFWQTEKYFKQIRPRLKDFTPRKLDREAGIISKMQQINSVSVHRRDYYV DESHYGDNTLDYYKRAIEYISSKIENPEFFSDDMAYVKEKFAGLKPHSFIDINSNNYSYKDLILMKKNCHN IANSTFSWWGAWLNNENEKIVIAPAKWFTVGENDKDIVPDEWIKL	24
Thalassospira profundimaris; Thalassospira profundimaris WP0211	WP_00888 9330.1	496164823	glycosyl transferase family 11 [Thalassospira profundimaris]	30		MVIVKLLGGLGNQMFQYATGRAVASRLDVELLDVSAFAHYDLIRRYELDDWNITARLATSEELARSGVTAA PPSFFDRIARLRLDLPVNCFREASTYDPRILEVSSPVYLDGYWQSERVFLDIEKKLRQEFQLKASIDANNHSFK KKIDGLGKQAVSLHVRGDIYVTPQYASVHGVCSLDYYRAAVDYIAEHVSDPCFFVSDDEWVQTNLNIK QPIVLVDANGPDNGAADMALMMA CRHIIANSFSSWWGAWLNNLNDKIIVAPKKWFGGRANHDTTDLVP DSWVRL	25
Acetobacter sp. CAG:267	WP_02207 8656.1	547459369	alpha-1 2- fucosyltransferase [Acetobacter sp. CAG:267]	29.9		MAVSPQESKYSAHVSPDKPLRVLGGLGNQMFQYAFGLAAGDVLWDNTSFLTNHYRSFDLGLNISGDF ASNEQIKKCKNEIRFKNILPRIRKKNLGFVILKTNVCEQINRYEPPELLSKDGVYDGVQTEKYFKPLRE RLLDHFTLTPLDAANLMLAKIRAADAVAVHIRRGDYLNPRSPFTYLDKDYFLNAMDYIGKRVDPKPHFFIS SDTDWVRTNIQTAYPQTIVEINDEKHGYFDLELMRNCRHNIIANSTFSWWGAWLNNTPDKIVVAPKQWFR PDAEYSGDIVPNDWIKL	26
Dysgonomonas mossii; Dysgonomonas mossii DSM 22836	WP_00684 2165.1	493896281	protein [Dysgonomonas mossii]	29.9		MVTVLLSGGLGNQMFQYAAKSLAIRLNTALSVDLTYFSKKTQATVRPYELGIFNIEDVVETSSLKAKAVIKAR PFIQRHRSFFQRFVFTDYAILYQPTFEALTGGVIMSGYFQNEYSYFKNISELLRKDFSFKYPLIGENKDVAGQI SENQSVAVHIRRGDYLNNKNSQSNFAILEKDYEKAINYSAHVKNPEFYVFSDFDWIKDNLNFKFEPVTFID WNKGKDSYIDMQLMSLCKHNIIANSFSSWWGAWLNNSEERKIVAPERWVFWDEQKNELLDCFYPQGWIKI	27
Clostridium sp. KLE 1755	WP_02163 6924.1	545396671	glycosyltransferase, family 11 [Clostridium sp. KLE 1755]	29.83		>gi 545396671 ref WP_021636924.1 glycosyltransferase, family 11. [Clostridium sp. KLE 1755]MIIIEISGGLGNQMFQYALGQKFIISMKGKVKYDLSFYNDRVQTLRQFELDFDLDCPVASNSELSRFGK GNSLKSRLLKQKLGWDKEIYEENLDGYQPRIFELDDIYLSGYWQSELYFKDIQELRLTYTFPIQLDYMMNGVFL RKIENSNSVSHIRRGDYLNNLKIYGNICTLNNYNNKALQIAKKTNPFIIVFTNDIEWVVRKELEIPNMVIVDC NSGKLSYWDMYLMSKCKANIVANSFSSWWGAWLNNKNIENRIIISPKRWLNNHEQSTLTCNDNWRICGDD	8

Gillisia limnaea; Gillisia limnaea DSM 15749	WP_00698 8068.1	494045950	alpha-1,2- fucosyltransferase [Gillisia limnaea]	29.28	MFISKNTVIILVGGGLGNQMFQFAIAKIIAEKESEVLDITFYTELTKKPRHFLGIFNSSFAIASKKEIDYF TKLSNFKFKKGLNYPYTFIHESSENFKAQVLELKAPIYLNQYFQSFYFLGKEYVIRKIFKFPDEALDKDNDNI KRKIIGKTSVSLHIRRGDYVNNKTKQFHGNCTIDYYQSAIAYLSSKLTDFNLJFFSDDIHWVVRQQKFNISNQKI YVSGNLHNHNSWKDMYMLMSLCHHNIANSFSSWWGAWLNKNPEKIIAPKRWFADTEQDKNSIDLIPSEWY RI	WO 2015/175801
Methylobacterium mobilis; Methylobacterium mobilis JLVW8	YP_003048 467.1	253996403	glycosyl transferase family protein [Methylobacterium mobilis JLVW8]	29.19	MLVSRIIGGLGNQMFYAAARAASLRISVQLKLDLSGFETYDLHAYGLNFNINVEDVAKKDDYFAGAPESLLKK IKKYLRLGLQLLESFESDLSFDSKVLNDNTYLDGYWQCYERYFIDEDKQIRQDSFKFAPDALNQRYLELDSV NAVSVHRRGDYVNSNTTNEIHGVCLDYYQRAAEFRARIGPENLHFFVSDDDTDWVKENISFGSDTTFIS HNDAAKNYEDMRLMSACKHHIANSFSSWWAAWLNPSKQVVIAPRQWFKSTLLNSDDIVPASWVRL	30
Runella slithyformis; Runella slithyformis DSM 19594	YP_004658 567.1	338214504	glycosyl transferase family protein [Runella slithyformis DSM 19594]	29.14	MIIVLGGGLGNQLFQYAFGRHLATVNOQKELKLDTSALTSTWNTNRSYALDAFNIRAQEAATPEEIKALAGKP NRLLQVRGRKVGITPIQYFQEPHFHFYSSALSISKSSHYLEGYVWQSEKYFEAITPIIRFEAFITSPSTHAQTIKEKI SNGTSVSIHLRRGDYVKTSSKANRYLRPLTMDYYQKADYINQVRKNPNFFLSDDIKAWKSQVTFPPPTTHFST GTSAHEDLWLMTHCRHHIANSFSSWWGAWLNQPPDKIVIAQPKWFESTERFDTKDLLPEPWQL	31
Pseudomonas haloplanktis; Pseudomonas haloplanktis ANT/505	WP_00295 8454.1	489048235	alpha-1,2- fucosyltransferase [Pseudomonas haloplanktis]	29.1	MIKVAIGGLGNQLFQYATARAIAEKRGDGVVDMDSFSSVYKTHPCLNKRCKATYESKPKLNKLLSNEKIR NILLQKLGFIKKYFETQLPFNEVDVLLNNSINYLTYFQSEKYLSIRECLDELTLIEDLNIAETA VSKAIKNAKNSI SIHIRGDYVNEGANKTHGVCDSDYFKKALNYFSEKLLDEHTLFISSDIEWCRNLSFDYKMFNFDGSS ERPEVDMVLMISQCKHQVISNFTSSWWGAWLNKNDKVVVAPKEWFKSTDLSTDIVPNQWIKL	32
uncultured bacterium	EKE06679.1	406985989	glycosyl transferase family 11 [uncultured bacterium]	28.67	MLTLKGGGLGNQMFQYAAASHNLAKNKKTKINFDLSSFFSDIEVRDIDKRDYLLDKFNISADISFDQKNSISGRK FLVKVISKFFGEVYRRLKFLSSKYLGDYFQSEKYFNVEEDIRKDFTLKDEMGVEAKKIEQQIVNSKNSVSLHIR RGDYVDDDLKTNHGVCLDYYKRSIKYLKENFGEINIFVSSDDIAWVKENLAFENLQFVSRPDIKDYEELML MSKCEHNIANSFSSWWGAWLNENKNIIPKEWFKFNINEKHIVPKSWIRL	33
Clostridium sp. KLE 1755	WP_02163 6949.1	545396696	glycosyltransferase, family 11 [Clostridium sp. KLE 1755]	28.57	MVIVQLSGGLGNQMFYALYLSLAKAGKVVKIDITCYEGGTRPKQLDVFVGSYERATKQELTEMTDSSLD PVSRIIRKLTGRKTAYREKIDINFPQVMERDPALLEGCFQSEKYFQDQREQVREAYRFRGIESGAYPLPEAY RRLEKEIADCKSVHIRRGDYLEESHGGLYTGICTEQYQEAFAARMEKEVPGAKFFLNSNDPDWTRHEFKGE NRILVEGSTEDTGYLDLYLMSCKKHNIANSFSSWWGAWLNNDNPKEKVTAPARWLNRECRDIYTERMIRI	1
Francisella philomiragia; Francisella philomiragia subsp. philomiragia ATCC 25015	WP_00428 7502.1	490414974	alpha-1,2- fucosyltransferase [Francisella philomiragia]	28.57	MKIKIQGGGLGNQMFQYAFYKSLKNNCIDCVYDKNYDLYKLYGFELNRIKFNIDLSFARKYHKKEVLGKLFESI IPSKFIVKFNKNYLOKNFAFDKAYFEIDNCYLDGYWQSEKYFKITKDIYDAFTPELDSINFELKNIQDYNLV SIHVRRGDYVNHPLHGGICDLEYNKAISFIRSKVANVHFLVFNIDILWCKDNKLDRTVYIDHNRMDSYK DMHLMSLCKHNIANSFSSWWGAWLNQNDKIVAPSKWFWFNDDKINQKIDICPNWVRI	5

Pseudomonas fluorescens;Pseudomonas fluorescens NCIMB 11764	WP_01733 7316.1	515906733	protein [Pseudomonas fluorescens]	28.52	MVIAHLIGGLGNQMFQYAAARALSSAKKEPLLLDTSSFSYTLHQGFELSKLFAGEMCIARDKNDINHVLWSQ AFPRIRNFLHRPKLAFRLKASLIIEPSFHYWNGIQKAPADCYLMGYWQSERVFQDAEEIRKDFTFKLNMSPPQ NIATADQILNTNAISLHVRRGDYVNNSSVYAACVVEYQAAIQLLSKRVDAPTFFVFSDDIIDWVKNNLNIGFPH CYVHNKGSSESYNDMLMSMCQHNIANSFSWWGAWLNSNADKIVVAPKQWFINNTNVNDLFPAPW VTL	WO 2015/175801	37
Herbaspirillum sp. YR522	WP_00811 7381.1	495392680	glycosyl transferase family 11 [Herbaspirillum sp. YR522]	28.48	MIATRLIGGLGNQMFQYAAAGRALRVGSPLLDVSGFANYELRRYELDGRIDATAASAQQLARLGVNATP GTSLLARVLKVPQPADRLREASTYDARIEQASAPVLDGYWQSERVFARIRQHLLDEFTLKGDWGSND AAMAAQIATAGAGAVSLHVRRGDYVNSAHTAQYHGVCSLDYRDVAHIGGRVEAPHFVFSDDHEWVR ENLQIGHPATFVQINSADHGIYDMMMLMKSCRHHIANSFSWWGAWLNPAAEDKIVVAPQRWFKDATNDT RDLIPAAWVRL		
Prevotella histicola;Prevotella histicola F0411	WP_00882 2166.1	496097659	rotein [Prevotella histicola]	28.43	MKIVKILGGLGNQMFQYALYLSKETFPQENVTVDLSCFHGYHLHNGFEIARIFSLHPDKATVMEILRIAYYP NYFFWQIGKRVLPQRKTMCTESTKLLFDKSVLQREGDRYFDGYWQDERYFIDCRRTILNTEKFPPTDDNNL ALLKMDTNSVSIHVRRGDYVGNKLYQGICDLYREAIMKISSYSPSMFCVFSNDIEWCRDNLSEFIKAPIY YVDWNSGTESYRDMQLMSCCGHNIANSFSWWGAWLNQNSKIVIAKRWLNKNCGFMPLSRWVKI		38
Flavobacterium sp. WG21	WP_01749 4954.1	516064371	protein [Flavobacterium sp. WG21]	28.42	MIVVQLIGGLGNQLFQYAAAKALALQTKQKFSLDVSQFESYKLHNVALNHNVISKNYKPNRYLRKIKSFYQ KNVFKYEVDFGYNPDLIHLKGGIIFLEGYFQSEKYFIKYEKEIREDFELRPLKKTAAIAKIESVNSVSIHRRGD YINNPLHNTSKEEYNNKALEIVENKINNPVYFVSDDMEWVKANFSTKQETIFIDFNDASTNFEDLKLMTSCK HNIIANSFSWWGGWLNKNPDKIVIAKRWFWFNDDSDINTNDIIPTNWVKI		39
Polaribacter franzmannii	WP_01894 4517.1	517774309	protein [Polaribacter franzmannii]	28.42	MIIVRVGGLGNQMFQYAYAKALQKQGYQVKIDITKFKKYNLHGGYQLDQFKIDLETSSPIANVLCRIGLRRS VKEKSLDFDEKFLIPQREYIKGYFQTEKYFSSITPILRKQFIVQKELCNTTLRYLKEITIQKNACSLHIRRGDYISDE KANSVHGTCDLPYKKSIRIQDEYKDAHFFIFSDDISWAKNLTNKNITTFIEHIVMPHEDMHLMSLCKHNIT ANSFSWWGAWLNQHENKTVIAKPNWFWVFNRENEVACANWVQL		40
Polaribacter sp. MED152	YP_007670 847.1	472321325	glycosyl transferase family 11 [Polaribacter sp. MED152]	28.42	MVVVRILGGLGNQMFQYAYAKSLAEKGYEVQIDISKFSYKYLHGGYHLDKFRIDLETANSSSAFLSKIGLKTIK EPNLLFHKDLKLVNNNAFIKGYFAEQYFSDIREILNQFKIKKELAKSTLAIKNQJELLKTTCSLHVRRGDYISDK KANKVHGTCDLDYSSAIEHISKQNSNVHFEVSDDIWVKDNLNITNATYIDHNVPHEDMYLMTLNHNHI TANSFSWWGAWLNQNPDKIVIAKPNWFWVDKENEVACKSWITL	PCT/US2015/030823	1

Methanococcus maripaludis; Methanococcus maripaludis C7	YP_001329558.1	150402264	glycosyl transferase family protein [Methanococcus maripaludis C7]	28.19	MKIQLKGGGLGNMFQYALYKSLKRGQEVLLDISWYLKNNAHNGYELEWVFGLSPEYASIRQCCKLGDPI NLIYVVRKVPKTHFEKSNFNDNNVFEVTNGYFEGYVQENYFKNFRSEILNDFSKNIDKRNAEFSEY LKSINSVSVHVRGGDYVTNQKALNVHGNICNLEYNNKAINLANNLNKPKFVIFSDDITWCKSNLGDIDPVVY DWNTGPYSYQDMYLMISNCKNIIANSFSWWGAWLNQNTKFKVSPKKVWVNDNRNNVNIIVNGWIK	WO 2015/175801
Gallionella sp. SCGC AAA018-N21	WP_018293379.1	517104561	protein [Gallionella sp. SCGC AAA018-N21]	28.15	MIAHIIIGGLGNMFQYAAGRALSLARGVPFKLDSFGEGYDLHQGFELQRFVFNCAAGIAEAEVRDSL GW QFSSPIRRIVARPSLAVLRSTFVVEPHFYWAGIKQVDPNCLAGYWQSEQYFQSHAAVIRTDFAKPPLSG QNSKLAMQIAQGNVSLHRRGDYANNPKTTATHGCLSDYYRAAIQHIAERVSQPHFFIFSDDIAWVKS NL AINFPHQYVDHNQGTESYNDMRMLMSLCQHNIIANSFSWWGAWLNTNAHKIVIAPKQWFANTTHVADLI PSSWERL	43
Azospira oryzae; Dechlorosoma suillum PS	YP_005026324.1	372486759	Glycosyl transferase family 11 [Dechlorosoma suillum PS]	28.04	MOSPACIAGARAWVVGYGMAEAMQPVVVGSLGGLGNMFQYAAGRALAHLRGLHPLSLDLSWFQGRG DRHFALAPHIAASLERAWPRLPAMQAQLSRLSRRWAPRIMGAPVFREPFFHYVPFAAALAAPVFLEGY WQSERVRELREPQLQDFSLRQPLPASCQPIAAIGNSDAICVHVRGGDYLSNPVAAKHGVCPVDYQQGV AEISASLARPHCFVSDDPWVRGSLAFPCMTVVDVNGPAEAHFDLALMAACQHFVIANSSLSWWGAW LGQAAGKRVIAPSRWFLTSDKDARDLLPPSWERR	44
Prevotella paludivivens	WP_018463017.1	517274199	protein [Prevotella paludivivens]	28	MKIVKIIGGLGNMFQYALAMALNKNFTDEVKLDIHCNFGYTKHQGFEDRVFNGEFELASYRDRVAKVAY PYNFKLWRIGSRIFDRRHIMISEDTSFKIMPEVITSHNYKYDGYWQHEEYFKNIHDEILDFAKFPKQDER NKALAEERLSDSNSISIHRRGDYLNDELFGTGCIEYKKAIEEINERTVPTLCVFSNDIHWCKENIEPLNGKE TIW/DWNTGSDNYRDMQLMTKCKHNIIANSFSWWGAWLNNTKDKIVIAPRIWYNTKEKVSVPVANSWIKL	45
Gramella forsetii; Gramella forsetii KT0803	YP_860609.1	120434923	alpha-1,2-fucosyltransferase [Gramella forsetii KT0803]	27.96	MSNKNPVIVEIMGGGLGNMFQYAVAKLAEKNSSVLLVDTNFYKEISQNLKDFPRYFSLGIFDISYKMG TEN GMVNFKNLSFKNRVSRKGLGNYPKIFKEKSYRFDADLFNKKTPYILKGYFQSYKYFVIGVESKIRQWFEFPYEN L GVGNEEIKSILEKTSVSVHRRGDYVENKTKTEFHGNCSELYYKNAITYFLDIVKEFNIVFFSDDISWVRDEFK DLPNEKVFVTGNLHENSWKDMYLMISLCDHNIIANSFSWWAAWLNNNSEKNVIAPKKWFADIDQEQLSL DLLPPSWIRM	46
Mariprofundus ferrooxydans; Mariprofundus ferrooxydans PV-1	WP_009849029.1	497534831	alpha-1,2-fucosyltransferase [Mariprofundus ferrooxydans]	27.92	MIIVOFTGGGLGNMFQYALGRRLLSHDVELKFDLSFYQHDIILRDFMLDRFQVNGQVATEKEIAYTNTPIF ALDRPLDLRLVRWGLYRGIVSVSDEPPGKQALMVVNSRVLAAPNTYVQGYWQSEKYFMPIRQKLLDDFSL VD KADQANGAMLEKIRQCHSVSLHVRGGDYVSNPLTNHSHGTGCLYYEKAIALIGSKVDDPHFFVSDPE WTRDHLKCRFPMYVTVCNSADSCEWDMELMRHCRDHIANSFSWWGAWLNMMNPDKNVVPVAPAAWFN NF-SADTSLDLPDSWVR	7

Bacillus cereus; Bacillus cereus VD107	WP_00217 4293.1	488102896	protein [Bacillus cereus]	27.91		MKIQVSSGLGNQMFQYALYKKSINDNDVFLDSSTSYMMYKQNHNGYELERIFHIKPRHAGKEIDNLSLDL SELISRRKLF GAKSMVVELKEFEYDPIFEKKEFYKGYWQNNYFKDIEQLRKDFVTEKLDKRNEKLANE IRKNVSIHRRGDYLLNKVYEEKFGNANLEYLLKAINLVKKIEDPKFYIFSDIDWAOKNINLITNDVVYISH NOGNESYKDMQLMSLCKHNIIANSTFSWWGAFNNNDKIVVAPKKWINKGLEKVELPENWITY	WO 2015/175801
Firmicutes bacterium CAG:534	WP_02235 2106.1	547951299	protein [Firmicutes bacterium CAG:534]	27.81		MIIRMTGGLGNQMFQYALYKRAMGKEVKMDDFTYEGRAPLSLWAFGIEYDRASREELCRMTDGF DPVSRIRKLFGRKSLMEKDCNFDPEILNRDPAYLTGYFQSEKYFADIEEVRQAFRFSERIWEIPSQLLER IRSYEQJKTMAVSVHRRGDYLLQNEEAYGGICTERYKTAIEYVKRQQDASFFVTNDPDYAGEWILKNF GQEKERFVLIJEGTQEENGYL DLYLMSLCRHHILANSSFSWWGAYLNP SREKMVIVPHKWFNGQECRDIYME NMIRIAKEQS	49
Sideroxydans lithotrophicus; Sideroxydans lithotrophicus ES-1	YP_003525 501.1	291615344	glycosyl transferase family 11 [Sideroxydans lithotrophicus ES-1]	27.81		MVISNIIGGLGNQMFQYAAARALSLEVPKLDISGFTNYALHQGFELDRIFGCKIEIAEADVHEILGWQSA SGIRRVVSRPGMSIFRRKGFVVEPHFSYWNIGIRKITGDCYLAGYWQSEKYFLDAAVEIRKDFSKLPLD SHNA ELAEKIDQENAVSLHRRGDYANNPLTAA THGLCSLDYRKSIKHIAGQVRNPYFVFSDDIAWVKDNLEIFP SQVVDYNHGMSFNDMRLMSLCKHHIIANSSFSWWGAWLNPKEKVVIAPERWFANRTDQDILLPPGW VKL	50
zeta proteobacterium SCGC AB-137-C09	WP_01828 1578.1	517092760	protein [zeta proteobacterium SCGC AB-137-C09]	27.81		MIVSQIIGGLGNQMFQYATGRALSHRLHDTFFDLDDGFGSYQLHQGFELSNVFOCEVNVATRSQMQALLG WRSFSSVRRLLMKRSLKWARHVRVMIEPHFHYWRSRFAEINEGCYLSGYWQSERYFKPIENIRQDFKFNHLL KGVNLDLAQQMTEVNSVSLHVRRGDYASDANTNHTHGLCLDYYRDAILYAQNTPVAPSFIFSDDIEWCRE HLKLSFPATYIDHNKGSNSYCDMQLMSLCHHHIIANSSFSWWGAWL NTRLDKIVIAPKQWFANGNRTD DLI PAEWLVM	51
Pedobacter heparinus; Pedobacter heparinus DSM 2366	YP_003090 434.1	255530062	glycosyl transferase family protein [Pedobacter heparinus DSM 2366]	27.8		MKIIRFLGGLGNQMFQYAFYKSLQHRFPV KADLQGYQEYTLHNGFELEHIFNIKVNSVSSFTSDILFYNNKW LYRKLRRLNLRNTYIEEKKLFSFDP SLLNNPKSAYWGYWQNFQYFEHIADDLRKDFQFRAPLSAQNQEVLD QTKLSNISLHRRGDYIKDPLLGGLCGPEYQTAINTYTSKVNAARFFISDDIDWCANLKLQDCSFISWNGK TSSYIDMQLMSSCKHHIVANSSFSWWAAWLNPNPDKIVIAPEKWTNDKINVRMSFPQGWISL	52
Methylophilus methylotrophus	WP_01898 5060.1	517814852	protein [Methylophilus methylotrophus]	27.78		MFQYAMGLSLAENNTPLKLDLSQFTDYKLNHNGFELSKVFNCSAETASVTQIETLLGICKYSFIRILKNTYLN LRPAQYVVEPFYWGDNVFLGDNVYLEGYWQSQYKFIIDYESTIRTHFTFKNLSGENKLSDRIKGSNSVSL HIRRGDYVTKNNNAFIGTSLIYQNAIEYFSTKIADPIFFISDDITWAKSNLRLANEHYFVGHNGQEDSHFD MQLMSLCKHHIIANSSFSWWGAWLNP SKDKIIIPKPKWFA SGLNDQDLVPKDWLRI	3

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Rhodobacterales bacterium HTCC2255	WP_00803 3953.1	495309205	alpha-1,2- fucosyltransferase [Rhodobacterales bacterium HTCC2255]	27.7	MIYTRIRGGGLGNQLFQYSAARSLADYLNVSGLDTREFDENSPYKMSLNHFNIRADLNPPDLIKHKDGIAYI IDHIKGNQKVKYKEPFLSFDKNLFSNVDGTYLKGYWQSEKYFLNRNRKNILSDINLIKKTDFNTINLKEIKKSTSI SLHIRRGDYLNSESYNETHGICLSYTTDAVEYKRNRLGENIKVFAFSDDDPDVWLENLKLSDIKIINNNTSANS FEDLRMLNCDHNIIANSSFSWWGAWLNNQPEKIVISPKKWWYNNKKQLQONADIVPSSWLKY	WO 2015/175801
Spirulina subsalsa	WP_01730 2658.1	515872075	protein [Spirulina subsalsa]	27.69	MAKIIARIGGIGNQJFIYAAARRLEINNAELVLDVSQGVHDLQYRQHYQLDHFHIPCRCATPAERFEFFSR VRRYLKRQLNQRLPFEQRRYVIEQSIDFDRLEFKPRGTVHLLEGYQWSEDFYKDIETIRQDLQIQPPTDPTN LAIVQHIHQHTSAVHIREFDQPNADTMNNAPSDYYHRAVEAMETFPVGAHYVLFSDQPEAAKSRIPLPDE RVTLVNHNRRGNKLAYADLWLMTCQCFHIANSTESWWGAWLAENKKQKQVIAPGFKEKREGVSWWGFKGL LPKQWIKL	55
Vibrio cyclitrophicus	WP_01043 3911.1	498119755	glycosyl transferase family 11 [Vibrio cyclitrophicus]	27.67	MVIVKITGGGLGNQLFQYATGSALANKLSCELVLDLSFYPTQTLRKYLAKFNINARVATDREIFLAGGGNDFFS KALKKLGTLTIIFPEYKEQESIKVVGKIDLCCKSGAYLDGYWQNPVYFSQNKIELTREIFLRAQLSPSALAWKDDHI SOASNSVSLHVRRGDYVENAHTNNIHGTCSLEYQYHAIEKIRSEVHNPFVFFSDDDIEWCKNLNLSLAEFEV DNITSAIDDLMLMRQCKHSIIANSTESWWGAWLKLDDGLVIAPRNNWFSSASRNKGIYPKEWHIL	56
Lachnospiraceae bacterium NK4A179	WP_02278 3177.1	551039510	protein [Lachnospiraceae bacterium NK4A179]	27.65	MRSVVDIKGGYGNQLFCYSFGYAVSKETGSELIIDTSMMLDMNNVKNYQLGVLTITYDSHISYKYGKDFLSR KTGLNRLRKKSAIGFTVVFKEQYVYDPSVFEIKRDTYFDGFWQSSRYFEKYSDDLRLKMLPKKISNAAEKL AEDARDCLSVSHIRRGDYVSLGWTLKDDYIKALDIKERYGSEPVFFVSDNKKYADDFFSAAGLYRLMD YETDDAVRDDMFLMSRCSHNIMANSSYSWWGAFNLNDNKDVTVCIPETGVWGGDFYPEGWMKVTASSG K	57
uncultured bacterium	EKE02186.1	406980610	glycosyl transferase family protein [uncultured bacterium]	27.57	MIIVNLYGGGLGNQMFQYALGRHLAEKNNTLKLDISAFESYKLRKYELGNLNIKEFALPEEISRLSTLPTGKIER FIRKTLRKPVKPESYIKENITGGFNPKILDQNNIYLEGYWQSEKYFIEIDIRKEFSKFPATGKNKEILENINI NSVSLHIRRGDYVTNPEVNVQVHGVCSLDYKSCVDIEKKLESFYIFSDDIEWYKNNLQIQSQVYYVDHNT VDNAIEDMRLMFSCKHNLANSFSSWWGAWLNSNPDKMVITPRKWFNNTTYDSNDLIPERWIKL	58

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WO 2015/175801	59	protein [Bacteroides fragilis]	492366053	WP_00582 2375.1	Bacteroides fragilis; Bacteroides fragilis HMW 616	MKIGIIVTGPYIKFWNEFYSSQLYFCVEAEKNEYFTDSSLASQRLPNVHMHLIEDKGWVWVSSKSFIC EIRNQLTSYDIYFYLNGNFKFSPICYDEILPOAEHNYLTALSFHYTHPHDYPYDRNKNCAFIYPYGGQKYF QGGFYGGRTQEVLSLSEWCRDAIEADFNKVIARFHDSEVINRLLTQHPKVLNDKYAFQDIWPVEGEYKAIV LNKEEVPEDNNLOEMKQNYIDPSLFLNDELKFIPIVQLYGGNGMQMFGYAFYLRHISTQERKLLIDPAP CKRYGNHNGYELPSFKICQDIHISDETKNNIRKLRKGTSLSEEVRAASMQSFKKQPIIFYSGCWQCCTVY ETVKDEIKDFIDESKLNPSAQMIRIRRSNSVSHIRNDYLGNNFELYGGICTKSYEKAISQMYTLLKDE PIFYFTDDPEWVRNFALDKSYLDWNKNDNWQDMYMSACRHHIANSSFSWWAAWLGFGPEKKVI APSTWLNQMOTDILPTEWIKIPITPDKKILDRICNHLHLSSYMKQLGNSGKMGVWIFFHYARYTONPLVE NYAGDLFDELYEIEHKGISFSLDGLGIAWAVEYL VHEQFIEGNTDLSLAIDFKVMQIDPRRFTDYSFETGL EGIACVYLSRLLSPRVCSSTLTDVYLKDLTEACRKYVPDKANYTRLFLNIESKEVGSFKDVLQMVLNHSEK AFGSDGLTWQTLTMIMR	27.46	3
WO 2015/175801	60	protein [Butyrivibrio sp. AE3009]	551034739	WP_02277 8576.1	Butyrivibrio sp. AE3009	MHIIQLKGGGLGNQMFQYALYKELRSRGKEVKIDDVTGFVDDLRTPVLQRFGEYDRATREEVVKLTDSKMDI FSIIRRLKLTGRKTCRIDESGTFNP'DILELDEAYLVGYWQSDKYFRNEDVIAQLRQEQKRPQEIIMTDSASWA TLQJIECCQSVSLHRRTDYIDEHNHHLNCTEKYKGAIDRISQYPSAVFFITDDKEWCRNHFGRGNFFV VELAEKENTDIAEMLLMSSCKHHICANSSFSWWSAWLNDSPEKMWVVPKNWINNRMDDDIYTRDMTKM AI	27.46	3
WO 2015/175801	61	protein [Bacteroides ovatus]	490447027	WP_00431 7929.1	Bacteroides ovatus; Bacteroides ovatus CL02T12C04	MKQTIILSGGLGNQMFQYAFFLSMKAKGKSCSLDTTLFQTNKMHNGFELKSVFDIPDSPNOASALHSLIKM LRRYKPKSLTIDEPTYTCPDALSKSKSFLMGDWLSPKYFESIKDVVNAVRFHNIKNKNDVDTANEMHGNNS VSIHRRGDYLLKPYCVCNENYRQAEIQKDRVDNPIFYVFSNEPSWCDSEFMKEFRVNFKIVNWNQKDS YQDMYLMTOQCKHNIANSTFSWWGAWLNNNTDKIIVAPSKWFKNSEHNINCKEWLLIDTSK	27.43	3
WO 2015/175801	62	protein [Desulfospira joergensenii]	550911345	WP_02266 4368.1	Desulfospira joergensenii	MGKYYETVWNGGLGNQIFQFSAGFALSRLNLDVLNISTFDSQKRNFEITYTPKIKNSFACIKDDDDPGVFS RLRIPFLNFKKIKQFHESHFFDPAFFDIREPVRIEGYFQSYKFEKYSQDLKDILLDPLTSRLKTVLKVISSKES VSVHRRGDYISDQGINEVHGTLEAYYLSIKLMEKMFPESEFFLFTDDPHYVEENKFLEDTSCISDNDCLP YEDMYLMANCHHNIANSFSWWGAWLNNQNPKEIVAPRKWFSRKILMEKPVMDLLPDDWILL	27.42	3
PCT/US2015/030823	3	protein C819_03052 [Lachnospiraceae bacterium 10-1]	507817890	EOS74299.1	Lachnospiraceae bacterium 10-1	MNIIRMTGGLGNQMFQYALFLRLKAQGEVDFDRTEYKGEEARPILLWAFGIDYPAAGEEVVELTIDGV MKFSHRLRRKLFGRKSEYREKSCNFDQQLKEPAYFTGYFQSERYFEEVKEQVRKAFQFSGKIWGSVSKEL EERIREYQTKIENKQMPVSVHRRGDYLENDEAYGGICTDAYYRKAEMMEKEFNTVFYFNSDGTGWAKQ WIDHFYKEKSRFVIEGTTEDTGYLDLFLMSKCRHIANSSFSWWGAWLDPDQEKIIVAPSKWVWNNQDMK DIYTRMIKISPKGEVR	27.39	3

Bacteroides dorei; Bacteroides dorei DSM 17855	WP_00783 2461.1	495107639	protein [Bacteroides dorei]	27.33	MVVVVGAGLANRMFQYAFALSLREKGLDVFIEDSFIPRFDFERTKLDVSFVNVIQRCDKNSFPLVLRED RFYKLLKRISYMSDNRVIERWNLDYLPYIHKASTNCIFIGWISYKYFQSSDADAVRKAFTKPLDSIRNVELAT KLVTENSAVHFRRKNIDYLNKLPNTCPSPYYEAINIKYVVPNPKFYFSDNWDVWVRENIRGVETAVDWN PSSGHSCHDCMQLMSLCKHNIA NSTYSWWSAYLNENNNKIVVCPKDWYGGMVKKLDTIIPESWIING	WO 2015/175801
Firmicutes bacterium CAG:24	WP_02191 6201.1	547127421	protein [Firmicutes bacterium CAG:24]	27.33	MVIVKMSGGLGNQMFQYALYRIQQTGKDVKLDLFSFQDKNAFRFRSLDFIPIEYQTANLEECRKLGECSYR PVDKIRKMFGLKESYQEDLDKGYQPEILEMNPVYLDGYWQCERYQDIKEKILEDTYFPKISIESRLQERI KNTESVSIHRRGDYLDAAANYKIYGNICTIEYYQSAISRMRKLCEKPNFYLSNDPEWAKEIFGDTEDITVEEDK ERPDYEDMFLMSRCKHNIIANSFSFWAAWLNQENKRVIAPKVWFNNHSVTDVICDDWIRIDGDHKGGA	65
Clostridium hathewayi CAG:224	WP_02203 1822.1	547299420	epsH [Clostridium hathewayi CAG:224]	27.3	MIYVNIIRGLGNQLFIYAFARALQKSTNQJITLNYTSFRKHYNNNTAMDLEQFNIPEDIMFENSKELPWFA NT DGKVIIRLRHYFKLIRSIQKMNVLMMWLGDEYEVKVNKRDRDIVDGFQSSRYFKSVYKELNLIKPMEM SKEIKTMGDLINQKESVCVSRRGDYVTVKNRDYYICDEKYLNTSIRMVVELVPMVTWVFISDDADWVK DNIVFPGEVYQPPRVTPLTLYLMKACKHFIISNSFSWWGQYLSNNDNKIVIGPAKWYVDGRKTDIEEE WIKIEV	66
Syntrophus aciditrophicus SB; Syntrophus aciditrophicus	YP_462663. 1	85860461	alpha-1,2- fucosyltransferase [Syntrophus aciditrophicus SB]	27.3	MVIVRLTGGIGNQMFQYAAARRVSLVNNAPLFLDLGWFEQETGSWTPRKVYELDAFRIAGESASVGDIKDFKS RRQNAFFRRLLPLFKKRIFHTRQTHIEKSYNFDPEILNLQGNVYLDGYWQSEKVSFSDVSEIRREFSFQTDPAE RNRKILERIASCESVSIHRRGDYVTLPDANAFHGLCTPAYRLAVEQISRKVVPEVFFVFSDDIAWARGNLKL GFETCFMDQNGPDRGDEDLRLMIACRHHIIANSFSFWWGAWLCSNPEKIVYAPRKWFNNGLDTPDNIPAS WIRI	67
Bacteroides cacciae; Bacteroides cacciae ATCC 43185	WP_00567 8148.1	491931393	protein [Bacteroides cacciae]	27.27	MKIVKIIGGLGNQMFQYALYLSKKYPKEKIKIDISMFETYGLHNGFELKRIFIDIDA EYASREEIRELSFYKIYKL QRIFRKIFPVKTECEVEKYDFKFMSEVW/SNCDRYEGYQWQWYFIEAQTEVRSFTTFKELVGRNAKVIREI QYAKMPVSLHRRGDYLLHHKLFGLGLD LNYKKAIDVNLNNYDTPQFYLSNDIEWCKTYILPLVQGYPFILVD WNSGVESYIDMQLMSCCRINIANSFSFWAAWLNDSSEKIVIA PKLWAHSPYKGEIQLKSWLLF	68
Butyrivibrio fibrisolvens	WP_02275 6304.1	551011888	protein [Butyrivibrio fibrisolvens]	27.24	MIIEMSGGLGNQMFQYALYKSMHLKGLDVTIDKSIYRDVDHKEQVLDLDRFPNVSIEADRKLSSTLRGYGY NDSIIDKIRNKLNSKRNLYHEDLDKGYQPEIFEFDNVNLNGYWQCERYFKDIKNEIKKDFIFPCTQSGDDKIK ALTIEMESCNLSLHVRRGDYLLKPGUEIYGNICTEYKYKSEIYKERVDPNVPVYFISNDMAWVRDNFKSDDFR YVNEGDGAFDGMTDMYLMTRCRHNIVANSFSWWGAW/LNKHDDNNIVICPNRNVVNTHTVTDICEDWIRI DV	69

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Parabacteroides distasonis; Parabacteroides distasonis CL03T12C09	WP_005857874.1	492476819	protein [Parabacteroides distasonis]	27.24	MIVGGNDYCKVNVNIIIGLGNQMFQYAFALSLKEHFKKEIRIDISHFNVLVNVKVGAAHLHNGYELDKIFFNIELKKANAWQLMKLTWFIIPNYLISRIARKILPVRNSEYIQNSDDCFYDPMVYNKQSGCYEGYVWQAIGYYE SMRD KLKIFQHPSPGKQYIENMESSNGIHIRRGDYLLSDNFRGICEVDYKRAIDKILQDGEKHVYFL FSNDQKWCEEYILPLLGNYEIIFVTGNIGRDSWDMFLMTHCKDLIIANSFSWVGAFNLKRGGRVVTPKR WMNRNIRYDLWMPWIRI	WO 2015/175801
Geobacter uraniireducens; Geobacter uraniireducens Rf4	YP_001230447.1	148263741	glycosyl transferase family protein [Geobacter uraniireducens Rf4]	27.21	MIARLQGGGLGNQMFQYAVGLHLALTHNVELKIDITMFSYKWHYSLRPFNIRESIATEEIEIKALTVDKMDR PYKKIDNFLCRLLKRSQKISATHVKEKHFIHYDPDILKLPDNVYLDGYWQSEKYKFEIENIRQTFIKNPQLGRDK ELACKILSTESVCLHIRRGNYVTDKTTNSVLGPCDLSYSCNICKSLAGNNKDPHFFVFSDNHEWVSKNLKLDYP TIYVDHNNEKDYEDLRLMSQCKHHIIANSTFSWWSAWLCSPDKVIYAPQKWFRVDEYNTKDLLPSNWLI L	71
Lachnospiraceae bacterium A4	WP_016280341.1	511026085	protein [Lachnospiraceae bacterium A4]	27.21	MIIVKIYEGGLGNQMFQYAFARSIQVNGKKVFLDTSGYTDQLPLCRTSTRRRYQLNCFNIRIKEVEKKNIEKYSFL IQEDMFGLKLSKLAKLHLWMYKVTIQNAQEYKESYLNTRGNVYKGFQWQNPKYFSSIRRLLLKEITPKYKIRI PAELRELLQEDNIVAVHCRRGDYQYIRNCLPVNYKKAMAYMEKKGVPYRLFSSDDLSWVKRQFGNKDN NYIYEDYGKFEDYQELMIMSRNFIANSTFSWWAAWLCSEYENKVVIMPRVWTVYVGGQGVEMSDPPAD WIRI	72
Colwellia psychrerythraea; Colwellia psychrerythraea 34H	YP_270849.1	71282201	alpha-1,2-fucosyltransferase [Colwellia psychrerythraea 34H]	27.15	MKVVRVCGGFGNQLFQYAFYLAVKHKFNETTKLDIHDMAHYELHNGYELERIFNLNENYCSAEKLAQSTK NIFTKLLKEIKYTPPIPTYIKKHLHFSYQEVDLGTDTSIYRGSWQNPQYFNISAEIREKLTFFTEPKSL ALHQEISEHETVAVHIRRGDYLLKHALGGICDLPYQNAIKEIAGLVEKPLFVIFSDDITWCRANINVEKRVFVD WNSGEQSFQDMHLMSLCTHIIANSFSWVGAWLNANPNKIVISPKNWIIHYTDSMGIVPSEWIKVETSI	73
Roseobacter sp. MED193	WP_009810150.1	497495952	alpha-1,2-fucosyltransferase [Roseobacter sp. MED193]	26.96	MITSLHGRGNQMFQYAAARALAHRLGCGVALDGRGAELRGEGVLT RVFDLP LSAAPKLPLKQHAPLRY GLWRGLGLAPFRFRERGLGYNTAFETWEDGCYLHGYWQSERYFEEISDLIRADFTFPDFSNRQNAEMAARI MEDNAISLHVRRGDYVALSAHLCDQAYEALTRLEGLSQDAPTYYVFSDDPDWAKANLPLPCKKVVVD FNGPETDFEDMRLMSLCKHNIIGNSFSWAAWLNANPNQKRVAGPANWFGDPKLSNPDLPSQWLKVA P	74
Cesiribacter andamanensis; Cesiribacter andamanensis AMV16	WP_009197396.1	496488826	Glycosyl transferase family 11 [Cesiribacter andamanensis]	26.89	MMIVRLCGGLGNQLFQYAVGKQLSVKNNIPLKIDDSWLRLPDARKYRLQFFQIEEP LASPQEVEFVGPYES QSLYARLYRKVNMLPRHRRRYFQESGFWAYEPFELMRISQVLEFGWQHAYFTRLHPQVLEALQLREY ROEPYAVLDQIREDAASVSLHIRRGDYSDPNLQFFGVMPLSYQQAVAYMQEQLHAPTFFYFSDDLDA RAHLKLQAPMVFDVIEGGRKEYLEAMRLCRHNILANSFSWVGAYLNTNPHKRVIAPIRQWVADPELKD KVIQIMPDWILL	5

Rhodopirellula sallentina;Rhodopirel lula sallentina SM41	WP_00867 9055.1	495954476	glycosyl transferase family 11 [Rhodopirellula sallentina]	26.89	MIATRLIGLGNQMFQYAYGSLARRRRLVLDVSFAFESYDLHALAIDQFIDISAARMTQAEFARIPGRYRG KSRWAERVANFAGGLQSCDKRPLRLRREKPFGEAEKYLAEAGSDLYLDGYWQSERYPFGLQAEKKEFQLKRG LSDESSRVLDIEQSSMSVAMHVRRGDYVTNAETLRIYRLDAEYKRLCLNDLRQFNSNLNVFVNSNDIQWCCQ DHLVDVGLKQRPVTHINDATTAIEDMELMSQCDHSIANSFSSWWAAFLGRSDAQRRVYYPDPWFNPGTLN GDSLGCANVWVSESSISVSRPSRAA	WO 2015/175801
Butyrivibrio sp. AD3002	WP_02276 2282.1	551018054	protein [Butyrivibrio sp. AD3002]	26.85	MIIRMMGGGLGNQMFQYALYLQKALGKEVKIDDVYGFRRDDPQRDPVLEKMYGITYTKASDAEVVDITDSH LDIFSRRRLKLGKRSHEYIETGLFDPKVFEEETAYLNGYFQSDKYFDPKVEVLAQLRREFVIKPDDVFTSADSW ELYRQIRETESVSIHVRRGDYLLPGTGVTFGGICNDYKRAIDRMVSEHPDAIFVFTSDKEWCEQNVSGKK FRIVDTKEENDDAADLLMSLCKHHILANSSYSWWSAWMNDSPEKTVIVPSKWLNTKPMDDIYTSRMTKI	77
Segetibacter koreensis	WP_01861 1017.1	517440157	protein [Segetibacter koreensis]	26.78	MVVVKLIGGMGNQMFQYAGRHILAIKNCPLYFDHIELENKNTANTPRNYELDIENVQYQKNPFLQSNRFV AKVYHKLFSVQRIKPEDFTFHPHILNVQGNHLYNGYWNENYFKEIEEIRQDFTFKTPANEKIESILQQAATN SVSLHVRRGDYITLLEANQFHGVCSDTYYQKAIKAIKPAIPHLFVFSDDIHWVQKNMPTFEEHTFVDGNT GKNSFEDRLMAACRHNILANSSFSWWAGWLNKNPEKMWIAPEKWFRAVHTDIVPPSWIKM	78
Amphritea japonica	WP_01962 1022.1	518450815	protein [Amphritea japonica]	26.76	MVIVRLIGLGNQLFQYAYALSILEQGYDVKLDASAFESYTLHGFGGLGEYAERLEVATTEEDVMVSRVGRIS TLLRKLQGGKSRVVIKESNFYSYDEKMLTPEDSHYLVGYFQSELYFNKIRGELLALDLKHLKSPYTEASYLAIDA SVSVSMHIRRGDYVSDKAAHNTHGVCSDLYYAAVTFEEERYPDVDFYIFSDDIEVWKENLNVQRAHYISSEE KRFAGEDIYLMSCQDHNIVANSSFSWWGAWLNANEDKIVVAPRQWYADSNMQRLSKTLVPTDWTIRL	79
Desulfovibrio vulgaris;Desulfovibrio vulgaris str. 'Miyazaki F'	YP_002437 106.1	218887785	glycosyl transferase family protein [Desulfovibrio vulgaris str. 'Miyazaki F']	26.76	MRPVVVDIFGGLGNQMFQYAAAKSLAERLGVRLDLVSMFSGDPLRAFSLGEFAITDHVRGKRSLLVRFA RSLGFGSSSKCCEPFHHYWEGINEIEAPVHMHYWQSEYFKAYEDLIRRTFSACEGVASSGKYAGVSSP MSVSVHLRRGDYKEQKNVVHGHILGREYDAAYSIIKQGPCSACFFVTDAINEAVDFSHWVNDVLFVDGN NQYQDMYLMSCQRHHIIANSSYSWWGAWLGAESDGMTVAPKMWFAFDVLKEKSIKDLFEDWIVL	80
Spirosoma spitsbergense	WP_02060 6886.1	522095677	protein [Spirosoma spitsbergense]	26.76	MIISRTSGLGNQLFQYAVARHLSLKNKTSLYVDLSYLYQYHDDTSRNFKLGNFSPYHTLQQSPVEYVSKAT KLLPNRSLRPFFLFQKERQFHDFEQILQSRAGCVILEGFWQSEAYFRDNADTIRDLQLSGTSPFENQYRELI RETPMSVSIHVRSDYVNHPEFSQTGFVGIDYYKRAIELARKELANPRFFVSDDKWESKTNLPLGEDSVFV QNTGLNGDVADLVLMSCQHIIANSSFSWWGAWLNPAGKLVITPKNWKYKNKPAWNTKDLLPTTWLS	PCT/US2015/030823

Lachnospiraceae bacterium 28-4	WP_01629 2012.1	511037988	protein [Lachnospiraceae bacterium 28-4]	26.73		WO 2015/175801 83 MNIIRMSGIGNQMFQYALYLKLSLGKEVKFDDVTEYELDNARPIMLSVFGIDYPKASREELVELTDSMD FLSRVRKIFGRKSGEYHEASADYDETVEKEHAYLCGCFQSERVKDIEYEVREAYFRNVVVEIRGGIETY ERQIGESLSVHIRRGDYLDAADVYGGICTDAYNQAIRYMIKKYENPFFVTNDTFAEAKWCVEVRETEG KRFTVIKGTDEETGYIDLMLMSRCKAHIANSSFSWWGAWLDASPKCVVAPVKWINTRECRDIYEDMVR IGSNGKISFNSCSSL
Lachnospiraceae bacterium COE1	WP_01630 2211.1	511048325	protein [Lachnospiraceae bacterium COE1]	26.71		84 MVVRIWEGLGNLQFQYARALSLRTKDRVYLDISEYEMSPKVRKYELCHFQIKQPVINCGRIFPFVNKDS FYTNNQYLYRYPAGLKEEDCYKRFDFCELGLLYLKGWFQSEKVKFEFESHIREIYPRNKIKITRGLRKILNSD NTSVHIRRGDFGKDHNLPIEYENS KRVLIRVDNYPYFIIFSDDLWVYKEMNFGNLNCFYMDKEYSKDYEE LMIMSRCKHNIIANSTFSWWGAWLNPSSDKIVIAPKKWFLYNPKKDFDIVPNDWIRV
Parabacteroides;Para bacteroides sp. 20_3;Parabacteroides distasonis CL09T03C24	WP_00586 7692.1	492502331	alpha-1,2- fucosyltransferase [Parabacteroides]	26.69	FutZ B	85 MKIVNIIIGGLGNQMFQYAFVALKAKYPNEEVFIDTQHYKNAFIKYYHGNFYHNGEYIDKVPFNATLEPAR PKDLMKVSFYIPNQVLAARVRRIKRTFVTDDQQPVYFIPEALSVIDDCYFDGYWMTPLVFDKDRILKEF TERPFDTKENLELEPLLKQDNSVTVHIRRGDYVGSFGIGICTLDYRNAIREAYNLITSPEFFIFSNDQKWC/M ENMRNEFGDAKVHFIHNRGADSYRDMQLLSIARCNILANSFSWWGAYLNQRKNCFIICPHKWHNTLEY SDLYLPTWIKI
Bacteroides sp. HP50048	WP_00256 1428.1	488624717	protein [Bacteroides sp. HP50048]	26.62		86 MFVIRLIGVGNLQFQYTFGQFLRHKEGVEVCYDIVAFDTV/DKGRNLELQLLDESPLFETSNNFFSKYKSWK KRLFLYGFLKKNNKYTKYAPEEISLFTKGLSYFDGWVWQYPALLRDTINNMEDEFIPKQIPVQIQKYNEIL LNNFAVALHVRRGDYFTSKYAKTYAVCNVEYTSVAVNLMCEKLRSCFKYVFSDDLDVWYKSNLILPSNTVYVK NYDINSVWYIYLMSLCRHIIISNSSFSSWWGATLNRNFHVIAPKYWSTKKNNTLDCNSWIKI
Bacteroides thetaiotaomicron;Bac teroides thetaiotaomicron dnLKV9	WP_01626 7863.1	511013468	protein [Bacteroides thetaiotaomicron]	26.58		86 MKIINILGGLGNQMFQYAMYLALKNASHSEELCSTRFCGYGLHNGYELGRIFGIQVKEASLLQLTKLAYPFF NYKSWQVMRHWLPVRKTMTRGAINIPFDYSQVMREDSVYVYDGYWQNEKNFLHIREEILTATYTFPKFDDK NQELADIIVKSNVACHIRRGDYLLKEINMCVCTSSYAHASIMNEEINPNLYCVFSDDIEWCRNNICELMGE DKKIIFIDWNKGEKSFMDMLMSLCKHNIIANSSFSWWGAWLNRNDKKIVVAPTRWIASEVKNDPLCDSW KRIE
Desulfovibrio alaskensis;Desulfovibrio alaskensis G20	YP_389367. 1	78357918	glycosyl transferase [Desulfovibrio alaskensis G20]	26.56		PCT/US2015/030823 MKFVGWVILGGLGNQMFQFAAAYALAKRMGGELRLDLSGFKKYPLRSVSLDFTVDTPLWHGLPMSQRRF RIPMDAWTRGSRPLVPSPFVMAKEKNFAFSPVYELQSCYLYGYWQSYRQDVEDDRTLFSLSRFATL ELAPVAQLNEVESVAVHLRRGDYITDAASNAVHGVCGIDYQSRMSLVRRSTTKPIFYIFSDEPEVAKKLAT EDDVVMPSPRRQEEEDLLMSRCKHHIIANSSFSWWAAWLGRASGLCIAPRYWFAFRPKLESTYLFDLIPDE WLLL

Prevotella oralis CC98A	ETD21592. 1	564721540	protein HMPREF1199_0 0667 [Prevotella oralis CC98A]	26.56		MDIVVIFNGLNQMSQAFYLAKRKSGSRCHCIFHNSTGFHNGSELDKVFGIKYEKGIFSKLLSKYIDFDGIP KLKKLNSLGIHIREPRNYDYTASLLPRSRWGLNYFVGWVHSEKYYTEILQEIKNTFSKIDDEIKDIDFEYFYS LIHNDINSVSLHIRRGDYVGANEYSYFQGGVATLEYHKAIDEIYQRIENPTFYVFSDDIGWCKTTFLKNNFIF VDCNCGEKSWRDMFLUSQCKHHIIANSTFSWWGAWLSIFHNSITICPKFIKGVIRDVYPTDWIKLSS	8
Comamonadaceae bacterium CR	YP_008680 725.1	550990115	glycosyltransferase [Comamonadaceae bacterium CR]	26.54		MASKISKIIPRIFGGLGNQFIYAAARRLALVNGAELALDVSGFVRDHEYNRHYQLDHFHNIPCRKATAAERLE PFARVRRYLKRKWNQRLPFEQRYLVQESVDFDERLLTFKPRGTVYLEGYVQSEDFKDIQIRADLRHPP TDTVNQQMAERIRATNAVAVHVRFFDAPAOQSALGVGGNNAPGDYVYQRAIKVMQEQAPDAQYIYFSDQP QAARARIPLRDDHVTLVNHNQCDAYADLWLUSQCQHFIIANSTFSWWGAWLGKTPESIVAPIGFEKREG AMFWGFRGILLPDRWVKL	89
Vibrio nigripulchritudo; Vibrio nigripulchritudo AM115; Vibrio nigripulchritudo FTn2; Vibrio nigripulchritudo Pon4; Vibrio nigripulchritudo SO65	WP_02259 6860.1	550250577	WbA protein [Vibrio nigripulchritudo]	26.51		MKDSRIVKLNGLGNQMFQALAFALKKLNVAVKFDTLLDNRTEFKLSLERFGLIVDKLTTEKFKYKGLE SCKYRKICNWNISNFTTINIHKYKKEKRGVYDRGIFDSNVKYIDGYWQNGEYFNDFRSELLNKFNLNGKVS HAIQYLKEITSVQNSVSIHVRRGDYLLDVYRNLTLDYVSEAIKLVITNPDSKFFISNDINWCKSNFKSVDNAI FVDSTVDEFDDMFLMSKCKCTNIIANSTFSWWAAWLNNSGKIVYCPKKWRNDTTEVHKGLPEGWNIIDK	90
Sulfurospirillum deleyianum; Sulfurosp irillum deleyianum DSM 6946	YP_003304 837.1	268680406	glycosyl transferase family protein [Sulfurospirillum deleyianum DSM 6946]	26.48		MIIKIMGGTSSQMHKALGRVLSLKYNVPLKDLTWFDNPKSDTPWEYQLDYFNINATVSEIKKLGNN LFNRIARKIEKFFSIRYKKSINKSFISDFHKLSDIYLDGEWNGFKYFEDYQDTIKNELTLKRGSSINQNTIKE LKSSDSVFLHIRRGDYLSNKNAAAFAHAKCSLDYKAIQIVKEKIDNPIFYIFSDDILWVKKNFVINESCRFME KNQNFEDLLMSYCKHGTANSFGFSLMAGWLNQNKDKMIIVPQTWVNDNRININILNSLEQDNFTIIR	91

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Bacteroides salyersiae; Bacteroides salyersiae WAL 10018 = DSM 18765 = JCM 12988	WP_005923045.1	492689153	protein [Bacteroides salyersiae]	26.41	MKQTIIMSGGLGNQMFQYALYCSMRKIGIRVKIDISLYEFNRMHNGYMLDYAFGLNISHNKINKYSVLWTR LIRSNRAPPELLFREDESFCDDVFTTYKPYIDGCWIDERYFFENIKKIIQSQSFHNIDQKNLMMVANNMMKVCNS VSLHRRGDYLSQSMYCNIESYKSAIEYIISRVESDKFFISDDPEWCKYFMEKFNVDYIEIQHNFSGKDSYKD MYLMTQCKHNIJIANSTFSWWGAWLNNAGKNVVCPSWINGRDNPCLEEWYHI	WO 2015/175801
Bacteroides fragilis; Bacteroides fragilis CL03T00C08; Bacteroides fragilis CL03T12C07	WP_005786334.1	492241663	protein [Bacteroides fragilis]	26.38	MDIILLHNLGNQMSQYAFYLSKKKNGIHTSYICLSNDHNGIELDKVFGVECMGCKKIFLLFILRLMSNRT GELIRKVNLLFSKIKIKLITENLDYSFHPFLSASPYCLAFWVGWGHHPQYSEISSQKEAFFTKRSLDERNICI EKRMREPNSVCLHRRGDYLTGINYELFGKVCNEQYQKAIIDYIEGKLSIDICYVFSNDMEWAKKILLGKNAV FYDWNRGESWKMVLMKSCNLIIPNSTFSWWAAWLCEHPVNI/CPKLFVYGVGEQSDIYLDNWHKIE	96
Bacteroides nordii; Bacteroides nordii CL02T12C05	WP_007486843.1	494751435	protein [Bacteroides nordii]	26.37	MMGIEKTNMIVIVRLWGGIGNQLFQYSFGFEFLREKYQVDVYDIASFSGSKDKLRKLELSVVVPGIPVTTDISFSK YVGTKNRLLRFYGLKNSFEEKYFSDEQLFKYLSKRGDYLQGYWQKTIYAETLRRKGSFFLSQEEPIVLHTIKA KIQEAEGAIALHVRRGDYFSKHIHTFGVCDAAHYEKAVIDMRGRVSNAMIFVSDDLDDWVRRYVNLPTNVI YVPNYDIPQYVWYLYLMSLCRHNISNSSFSSWWGAFNLMMNTNKIVSPSKWTLNSDKTIALDEWFKI	97
Butyrivibrio proteoclasticus; Butyrivibrio proteoclasticus B316	YP_003829743.1	302669783	glycosyl transferase 11 [Butyrivibrio proteoclasticus B316]	26.37	MECSMIIKFCGALGNQLFQYALYEKMRILGKDKVKAISAFSGDGNKRFYDELGIEFNIAADEIAEYLNKRT IRFVPGFLQHRHYFEKPPYVYNNKILSYDDCYLEGWQNYRYFDDIKDELLKHKMFPCLPLEQKLAEMEN ENSVAVHVRMGDYLNLQDLYGGICDADYDRAFSYIEGNISNPVYVYGFSDDDVDKASALLAKHKINWIDYNSE KGAYDLILMSKCKNNIIANSFSSWWGAYLEYNNGKVYVSPNRWMNCFENSNIAYWGWISL	98
Prevotella ruminicola; Prevotella ruminicola 23	YP_003574648.1	294674032	family 11 glycosyl transferase [Prevotella ruminicola 23]	26.33	MRIVKVLGGLGNQMFQYALYKALQKQYPEERVLLDLHCFNGYHKHRGFEIDSVFGVTYEKATLKEVASLAYP YPNYQCWRIGSRILPVRKTMKEPNFTLEPSALSIPDSTYDGYWQHEEYFMHIREELSTYAFPAFDDERN KTTAQLAASTNCSHIRRGDYLTDLPLRKGTNGVYVIAAIKEMQKEVPEKWLVSDDIAW/CQQHLASTLD ATNTIYIDWNTGANSIHDMHMLALCRHHIIANSFSSWWGAWLSQQDGTITAPSNW/MNLKDVCSVPDN WIKI	99
Prevotella salivae; Prevotella salivae DSM 15606	WP_007135533.1	494223898	protein [Prevotella salivae]	26.33	MKIKIIGLGNQMFQYALYALQKQYKDEEIRLDLNCFRGYNKHQGYLLDEIFGRFRFAASLQEVARLAWPY PHYQLWRVGSRLPRRTMVCEPADGSFSPDVLTLGNRYDGYWQDERYFKAYRKEIEAFKFSFVGDG NRHVENMLRNERFASLHVRGDYLDALYQNTCGIDYQYRAISQMNAMAIPSCYFISDDIAW/CKTHIEPL CEGHRPYIDWNKGKAYRDMQLMALCKYHIIANSFSSWWGAWLNDADGEGTITAPQQWYSHGNKPSPAS ESWIKV	PCT/US2015/030823

Bacteroides ovatus;Bacteroides ovatus 3_8_47FAA	WP_00430 3999.1	490431888	protein [Bacteroides ovatus]	26.25	M V V V I A A G L A N K M F Q A F S R G L M S H G L D V F L D Q T S F Q P E W S F E D J A L E E V F P N I E I K N A P N N M F S L A Y K K D L I S R I Y R M S A F F P N N R Y L M E R P F I Y D E L I Y K K A T N N C I F C G L W Q T E L Y F N F C E R D V R R N F V F T P F Q D D Q N I K L A E K M K N E N S V A I H I R K G A D Y L K R N I W D G T C S V E Y N Q A I N Y L K E H V S N P V F Y L F T D N P E W V E E N L K N I D Y K L V D W N P V S G K Q S Y R D M Q L M S C A K H N I A N S T Y S W W G A W L N N N P Q K I V V A P K I W F N P K I E K A P Y I P D R W I R L	108	WO 2015/175801
Loktanella vestfoldensis	WP_01995 5906.1	518799952	protein [Loktanella vestfoldensis]	26.23	M I I T K L I G G L G N Q M F Q Y A A G R S L A M R H G V P L L D I T E L R S Y P K H Q G Y Q E D V F A G R F E I A G L I P L R V L G R K A R K V P K T V A V V S P K W P P M G D H V W V R Q R T H D Y D A A F E S I G A D C Y L S G F W Q S E K Y F A T I A P Q I R E S F R F K E A L T G A N A A I A S R M K E A P S A I H I R R G D Y V T D K G A H A F H G L C A W D Y Y D A A I D H I S R H E P D A R F F V F S D D V V A A Q E R F A N R Q R A E V V A V N S G R H S Y R D M M L M A Q C K H Q I A N S T F S W W A A W L N Q N P D K I V V A P G T W F S G N D G Q I K D I Y C K D W I V I	109	
Flavobacterium sp. ACAM 123	WP_01699 1189.1	515558304	protein [Flavobacterium sp. ACAM 123]	26.14	M D V V I I F N G L G N Q M S Q A F Y S Q K K I N N S T Y F V P F C K D H N G L E L E T V F S L N T K E T L I Q K S L Y L F R I L L T D R L K I V S D P L K W I L N L F K C K I V K E S F N Y N N P E Y L K P S K G I T Y Y G G W H A E K Y F A K E N Q Q K S V F E E T G D L G K I N K E H V K D I A S T N A V S L H V R R G D F M N E A N I G L F G G V S T K A Y F E G A I K L I A T K V D H P H F F V S N D M D W V K E N L S M D T V T Y V T C N S G K D S W K D M C L M S L C Q H N I P N S T F S W W G A W L N K N P H K I V C P S R F L N N D T Y T D I Y P D S W V K I S D Y	110	
Bacteroides fragilis;Bacteroides fragilis 3_1_12	WP_00577 9407.1	492219620	glycosyl transferase family 11 [Bacteroides fragilis]	26.1	M M K L V R M T G G L G N Q M F I A F Y I Q M K T I F P E L R I D M S E M K K Y K L H N G Y E L E D V F S I R P Q T I S A H K W L K R V I V Y A F F S I R E K S E E L S I H K Y T Q H K R W P L V Y Y K G F F Q S E L F F K E S S D T I R D I F S F N T E N A N F T K E W A K I E Q R S S V S I H I R R G D Y T S A K N K I Y G N I C T E E Y Y Q A I S I L K K E P A F F H I F S D D V E W T K A H L K I H L P H Q Y I S W N K G P D S W Q D M M L M S L C R H N I A N S S F S W W G A W L N A Y K D K T V I A P S R W S N V K K T P H I L P E S W I S I D I	111	
Spirosoma panaciterrae	WP_02059 8002.1	522086793	protein [Spirosoma panaciterrae]	26.09	M I I S R V T S G L G N Q L F Q Y A A A R S L S L R N K T A F Y V D L S Y Y L Y E Y P D D T S R S F K L G F F S V P Y R I L Q E S P Y E L S K S T K L F P N R S L R P E F F L K E K Q H F D P T I L Q A H A G C V I M E G F W Q S E C Y F R D H A E I I R R E L Q L S K S P S S E F E G Y H Q Q I Q A T P V P S V H V R R G D Y V N H P E F S K T F G F I G L D Y Y K T A I R H L T K T I K N P H F Y V F S D D K E W A R A N L P L P T D S V F V T N T G P S G D A D L V L M S T C H H I I A N S S F S W W G A W L N P N P D K L V I T P K L W Y K N Q P T W N T K D L L P P T W V S L	2	PCT/US2015/030823
uncultured bacterium	EKE06672.1	406985982	glycosyl transferase family 11 [uncultured bacterium]	26.09	M I I T K L T G G L G N Q L F Q Y A I G R N L I Y N G S D L K L D V S E Y D V S N K G N F R H Y A L D K F N T I Q N F A S K K E T N N F K F G V F K K W L Y K S G I V K N K N Y F L E K K F N F D K E I L K I D N A F L Q G Y W Q S E K Y F I G I R D I L L Q E F S L K E N I E L K F G E I L K E I N E S N S V S I H V R R G D Y V K N P K N L S F H G V C S P K Y S E S T S K I A S L I E K P V F F S D D I E W V K E N L N I T F P V V Y L S G I K N I K S Y E E L V L M S K C K H N I A N S S F S W W G A W L N T N Q K K I V I A P K R F W F N D V K L D T T D L I P E N W I R I		

Thermosynechococcus elongatus; Thermosynechococcus elongatus BP-1	NP_681784.1	22298537	alpha-1,2-fucosyltransferase [Thermosynechococcus elongatus BP-1]	26.07	MIIVHLCGGLGNQMFQYAAAGLAAAHRIIGSEVKFDTHWFDATCLHQGLELRVFGLELPEPSSKDLRKVLGACVHPAVRRLLAGHFLHGLRPKSLVIOPHFYWTGFEHLPDNVYLEGYWQSERYFSNIAJIRQQFRFVEPLDPHNAALMDEMOSQSVSLHIRRGDYFNPNQMRVRHGVDLSEYPAAVATMIKTAERFYVFSDDPQWVLEHLKLPVSYTVVDHNRGAASYRDMQLMSACRHHIIANSTTSWWGAWLNPRPDKVVIAPRHWFENVDFDTRDLYCPGWIVL	WO 2015/175801
Colwellia piezophila	WP_01902.8421.1	517858213	protein [Colwellia piezophila]	26.03	MKIVKIAGGFGNQLFQYAFYLALDKKYAEQVCLDSLDMAKYRLHNGYELEGIFKLDARYCTEEQRIVRKDNNIIFTKLLSSLLKKILGNKNNYILEPKQEHFTFHEKSFGQANTPTYYKGWQDVKYLENIEEELKSSLVFPEFELGKNIELANFISSNSSVSLHVRRGDYVQHKAFGGICDLSYQRAVEQINTLVKDPFIVFSDDIQWCKDNLNLEKAKFVDWNIGENSFRDMQLMTLCKHNIIANSFSWWGAWLNANDDKNVICPKWVHYTSATGVLPSSEWIKIKASV	114
Prevotella maculosa	WP_01996.6794.1	518810840	protein [Prevotella maculosa]	26	MKIVKIIGGLGNQMFQYALALQERWKDEEIKLDLHGFNGYKHQGYQLDMLFGHRFEAATLTDVAQLAWPYPHYQLWRVSGSRLPKRRSMLCEPSKGLLPDSVLKQKGSYYDGYWQDERYFRAIRPQIMAAFKFPDFTDRRNLETEKRLKASEAVSIHVRRGDYLDVLFQGTGTCNIAYYQRAIARLCQLKTPVFCIFSNDMAWCKVHIEPLHKGKELYVDWNRGKESYRDLQMLTLCRHHIIANSFSWWGAWLSKAEQDGTIAPRHWWYAHDAKPSPAELRWIKV	115
Salmonella enterica; Salmonella enterica subsp. enterica serovar Worthington str. ATCC 9607; Salmonella enterica subsp. enterica serovar Cubana str. CFSAN001083; Salmonella enterica subsp. enterica serovar Cubana str. CFSAN002050; Salmonella enterica subsp. enterica serovar Cubana str. CVM42234	YP_008261.369.1	525860034	fucosyltransferase [Salmonella enterica subsp. enterica serovar Cubana str. CFSAN002050]	25.99	MYSCLSGGLGNQMFQYAAAYILKQYFQSTTLVLDSDYYYSQPKRDTVRSLELNQFNISYDRFSFADEKEIKILLRKFKRNPFPKQISEILSIALFGKYALS DRAFYTFETIKNIDKACLF SFYQDADLLNKHKQLILPLFELRDDLLDICKNLELYSLIQRSNNTTALHIRRGDYVTNQHAAKYHGVLDISYNNHAMEYVERERGGKQNFIFSDDDVRWAQKAFLENDNCYVINNSDYDFSAIDMYLMSLCKNNIIANSTYSWWGAWLNKYEDKLVISPKQWFLGNNETSILRNASWITL	PCT/US2015/030823

Bacteroides sp. 3_2_5	WP_00865 9600.1	495935021	protein [Bacteroides sp. 3_2_5]	25.94		MKKVIFSGGLGNQMFQYAFYFLKKKGKAVIDNSLYSEKMHNGFELIKVFDIKESYRTYFLKVHLFIKLLMK IPPVRKLSCKDDVIPIGDHEFDPPYARFYLYGYWQSKIVNYVIEELRAQFIERNIPQMTEIKGDFLSSINSVSIHR RGDYMGIPIYQGGICNEIYYERALSFMKEHFLNPRFYVFSNDSIWAFLLEKFDIDMEIIVTPPIYSYWDMLMS RCRNHIANSTFSWWAAVLNINKDKIVISPTIFKDKDECIDIIFDDWVVKISNI	7 WO 2015/175801
Clostridium sp. CAG:510	WP_02212 4550.1	547662453	protein [Clostridium sp. CAG:510]	25.86		MIMLQMTGGMGNQMFYALYRSIRQKGEVCIEDFTHYDTPEKNCLQTVFHLDIRKADREVYQRLTDSEP DFLHKVKKRLTGRKEKIQEKDAIIFEPEVFTQDDVYMGYFQSGRYFEKAVDFLRKDDTFAWNTFPEKAKKL EQMQAESSVSLHRRGDYMGKFAISYGNICTDAYEAAARMYKMEHFGDCRFYLTDDAEWGRQSESDT VYVDASEGAGAYVDMALMSCRHHIIANSFSWWGAWLDENPDKTVIAPAKWLNISEGKDIYAGLCNCLI DANGSVQGE	118
Rhodopirellula europaea; Rhodopirelli ula europaea SH398	WP_00866 5459.1	495940880	glycosyl transferase family protein [Rhodopirellula europaea]	25.86		MIVTRLIGGLGNLQFYAFGHSLARSTYQTLIDDSAFIDYRLHPLAIDHFTISASRLSDADRSRVPKFLRTPV GRALDKVSRFVPGYQGVLPVRREKPFGRFRESLLARESDLYLDGYWQSEKFPGLRGLSIREEFQLRQPSSETTR RLSAQMKSENSVAIHVRRGDYVTSKAKAQIYRTLDADYRRCLLDLAAHETDLKLYLNSNDV/PWCESNLDVGI PFTPQHTDGTATAHEDLHIAQCRHVIVANSTFSWWGAYLQGLHPTRRRVYVPEPWFHPGTLDGSAMGCD DWISEASLEEQSSIKSSRRRA	119
uncultured bacterium	EKD23702. 1	406873590	glycosyl transferase family protein [uncultured bacterium]	25.82		MIIVKLKGMGNQMFQYAGRNALATKLGTLRLDLTELLDRSPRKDFVRDYDLDFALDVAFAGPTDLKPPT QFRISHLTKYINIFRLLGRPVYSEPHFSEALIKSSDNVLDGYWQSEKYEKEIENSIRDDFKFRQPLEGRAA EMAAQKKNEDRAVCLNVRADFTSKAQEFHFGIGLDYQKAVDILLVSKVGPLHLFISSDDVDW/CAANLK FNYPPTFTVKDYSGKKYEAYLQMLTLCRHYIIPNSTFAWWGAWLNSDPNKIVIAPKQWFKFEASIDTTDIIPT WIRL	120
Bacillus cereus; Bacillus cereus AH1271	WP_00058 7678.1	446510160	protein [Bacillus cereus]	25.74		MIIVKLKGGGLGNQMFQYALGKSLAYYDKPKIDADYIKNNEGYPVPRDFSLSKFNIELDLYQEADKERVGFILK NNFLAKKLRNYFLKGGKYKGYIENPDNLGLFKKELFENHESMYIDGYWQSYLYFNINIRECLIKFNLKPEYT KEMTEIMQRINETNSVAVHRRGDYVKGWTLDTTYKKAIAEIVKNDNPKFYVFSDDTDWVRSNLQELD NAVFIGECNLFQELWLMSTCKHNIISNSTFSWWGAWLNQNDHQVQVVSFAWINGMSVETTSIPDSW KRV	121
Firmicutes bacterium CAG:95	WP_02249 9937.1	548309386	protein [Firmicutes bacterium CAG:95]	25.74		MDIIRMEGGLGNLQFYALYRQLQFMGRTVKMDVTEYGREHQRQM/LWAFDVFHYEATQEEINRLTD GFMDLPSRIRKRLTGRRTKYYAEADSNFDPQVLLKTPVYLTGYFQSEKYEKDKVEGILHTELGFSDRVDGISEVF ADQIRNYQKQIRETSVSLHVRRGDYLEHPEIYGMSCMTEYQAGVRYRERHDPDAEIVFTNDPVTKEKWL QENFLGDFTLQGTSEETGYLDLMLMSQCKHQIMANSFSSWWGAWLNPKNKDKIVVAPEPWFQDGRNFHDI YTEEMIRISPRGEVKKHG	122 PCT/US2015/030823

Prevotella oris;Prevotella oris F0302	WP_00437 4901.1	490508875	alpha-1,2- fucosyltransfera se [Prevotella oris]	25.74	MIAATLFGGLGNQMFIVATVKSLSHYQVPMAFNLNHGFANDYKYHRKLELCKFNCLPTAKWITFDYRGE LNIKRISRRIGRNLCPNYQFVIEEEPHEKRLFEFTNKNIFLEGYWQSPCYFENYSKEIRADFLQKYPPLSKEML EEIYALKATGKTLVMLGIRRYQVEGRDICTYKLCDEYYIKAITIYIQRIPNALFVFTQDKIEWATTTHLPKGAE FYFVKDKQDEYATVADMFLMTQCTHAISNSTFYWWGAWLQCTTKNHIVIAPDSEFINSDCVCKEWIILKRNS LC	1
Escherichia coli	AA037719. 1	37528734	fucosyltransfera se [Escherichia coli]	25.73	MYSCLSGGLGNQMFQYAAAYILQRKLRKQSLVLDSDSYFLDCSNRDTRRRFEELNQFNICYDRLTTSKEKKEISII RHNRYRLPLFVTNSIFGVLLKKNYLPEAKFYEELNNCKLQVKNGYCLFSYFQDATLIDSHRDMILPLFQINEDL LHLCNDLHIYKKVICENANTTSLHIRRGDYITNPHASKFHGVLPMDDYKEAIRYIEDVQGEQVIIVFSDDVKWA ENTFANQPNYYVNVNNECEYSALDMLMSKCKNNIIANSTYSWWGAWLNTFEDKIVVSPRKWFAGNNKSK LTMDSWINL	124

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Leeia oryzae	WP_01815 0480.1	516890767	protein [Leeia oryzae]	25.71	MIIVKIIGLGNQMMQYAFAHACAKRLGVPFKLDITAFESYKLPYGLHNFEITAPIASLEEIEHAKSMGVITE TSFRFDDSLVSADKGMVLDGYWADRYSESVMGELKPVFTLMDPLTPEQQAAMNLSAPNAVALHVR GDVVTNPNCELLPQQYRDAIKLVLDQDPDAVFCFSDPDWVEAHLDPAPKVVVRGGIDNGFVDMIL MSKARHRIVANSTESIWAASRLADQDGLTIVPSQFFRKDDPWLQVYGEVLQPYPQWVRVVDVTGDGKKE AENTSTALLQIAGGDVRGRKLRIGVWGFYEEFYQNNYFNLKNAPIGHELLKPNQLYQYQGAHNLEFVTLDL VADLSTDAVLFFDAPNMRSPVSSVMQLDIKKYLCLECELIKPNWQQLSHLELFRIFTWHDGLVDNHR KVNVTDLMPWIESAQSLTAPFEETARKGYLQKKLICNISGNKLVSHPFELYSKRIEVRWFESHHPHF MGWSADYPSYKGIKIDDKLEVLKGYRFSCLCYENAKELPGYTEIKIDCFKAGWVPVSGAPNIADWIPDNC SGKFPDADALTYLJSMTEEVHADYLENIROQFLGGKAYPESADAFINTITRTIVQDCLPHERTDVSVV NHGNFVSAITSALNQNVSVELLVLDNASTDDSWSQLQFFADYPQVRLNRNWNIGVQHNWNNHATWLAT GRYVMSLADDLPLPGHLEQAVKRLDENPASSLYTTPCLWINEHDQPLGTNLHPGHLESDYVGGGRDEIS KFSYITPSAAVIRRETINRIGSMNLHLKGAIDWDLWIRIAEISPAFIRKQPGVCYRQHSNNNSVDFYASTAP LEDHIRVESIIDRKVAVKYLKAKEEIAHLNDRASSYPENQIQHLLSRINNIDYLRKGAGPVISVIIPTKNR LANALESITYQTFKDFEVVHNDDGGDGGIVDFSDQLQISYVRSOSGGAASRNRAKLAAGRIIAYLDDDD DVYLDHLEKLVDAKGRSEKHYTNCEYLQERKEGRLELGRERRYAGISYRAQLLVSNFIPTTWSHTKELI DTIGDFDESLEIEDWDLRASKVTEFYQVNAITVEVRSRSDRDDHTLRANADKLLAYHQKIYAKHPVENESI LANRQSLNSLRQDVTNENSYQGVWVNARQPNELAVQILAEARMMLQWSKQYQFMVIMVVKQSQQ NILLANTIDSFCCQLYSGWKLVISDFEAPDESFINNEVLGWLTTETVEDENLLTQAFNGVLAEPVSDWVTL GTRLTSTALLKVGDRLLNGGACVIYTDHLYVSDGMIKDPVLPKPAFNLDMLRSQDYIGSSIFFERTDLSA GFASFGARTYACFRMLDNYGPGQTIHLPPEVMTFPENQPENSLRVAAMQLALEEHLHRNNISASIEEGYV TGTFVQYHHSEQPFVSIIPNKKHEFLAPCIETLMKVYQYPAFEVIVDNQSTDPDTLSYEEIESREFANNV VIQYDNPNFSAQCNLGAESARGDFILNNDTEIVQANWLERMMQHAQRNDVGVVGARLVFPETVTIQH AGIVLGGKYPDEVFQPYMNFVDKDVSLNRTKVQNSAVTGACLLVRKSLYQQVGGMNEQLAVLYGD VDLCRLRQLHKSVMWTPFSTLVHHTGKTLNSNSDHEKHLMMVIQTRQREYMLSHWLDIANDPYHRL DKSECNGTIDCTHTPLWDDIPSARPLQGMALVGGSGEYRVNMPFTLERSALAEIVLSNMITSKARLPSTEL ARNAPDVEVQNALADEFIRMLEMYKKYLPVFRIQMLDLDLLEIPDASSFRHFKQKWRDARLRSKLF CDRLVSTEPLRTFAEDMIDDIIVPNMLERSVWGDLSKRRAGKKPRVGVWVAAQQAQAGDLALMTDWWKA TGHEVDWVFEQGMCPDRIYPYVAEVNTEWLTQYKYPQGIALLDLAIPLEINAFNEAKSNLRLLLEYGALG WPVICDIYPIYQTNNAIPVCRVPNDASAWIEAIRSHIADLADATAQKGDQLRQWVVDHYMIEDHAQEWSA LTRPAGK
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Desulfovibrio africanus; Desulfovibrio africanus PCS	WP_00598 4173.1	492830219	Glycosyl transferase family 11 [Desulfovibrio africanus]	25.68	25.68	MFQYAAARALSLRHSASLAADLTWFSQQFDVQTTPREYALPAFLRLPEADKRIVATFRLNPTLRIVSLRH RICFSPRELPRHITELSDYWDGFRDILPPAYLDGYWQSERYSFDYDPIRADFSMLSEQAAMWSAKIASVQ DSISLHRRGDYVNSLATRKAHGIDTERYAKALEWADRIAGATIFAFSDDPWVRANFDFGKHGIVVDGS WTAHEDMHLMSLCSHHIIANSFSFWGAWLSTSQGITIAPKSWFSNPHIWTDPVCPATWERIPC	1	WO 2015/175801
						MAKGIIVMRFLFGLGNQLFYAFALFALSRQGGKARLETSSVEHDDKRVCELHFRVSLPIEGGPPPWAFRK SRIPACLSLFAAPYPHFREKRHGFDPGLAAPRRHTYKGYFQTEQYFLHCREQLCREFLKTLPTPENARI LEDIRSCCSISLHRRTDYLSNPYLSPPPLEYLRMAEMEGRLRAADAPQESLYFIFSDDIWARQNLRLPALP HVHVDINDGGTGYFDELELMRNCRRHHIIANSFWSWAAWLNHAEKIVAPIRWFNREEGDRYHTDDALIP GSWLRI		
Dysgonomonas mossii; Dysgonomonas mossii DSM 22836	WP_00684 3524.1	493897667	protein [Dysgonomonas mossii]	25.66	25.66	MKIVKQGGGLGNQMFQYAIARTLETNKKDIFLDSFLRMNNVSTDCFTARDFELSIFPHLRKAKLNSLQKFL LLSDRVRYKFKIRKIANINFHKNQLENEIGIPFGIKNVVLDGFFQSESYFKHIFDLIKDFFPELDTRNEALKKTI VNNNSVSIHRRGDYVHLKNANTYHGVLSLEYLNCIKRIGEETKEQLSFFIFSDDPEYASKLSLFLPNMQIVD WNLGKNSWKDMALMLACKHHIIANSFSFWGAWLSENGITYAPVKWFWNNESQYNNIINIPSDWVII	128	127
Prevotella oris; Prevotella oris F0302	WP_00437 2410.1	490506359	glycosyl transferase family 11 [Prevotella oris]	25.66	25.66	MDIVLIFNLGNOMSOYAFYMSKKFVPOQSKCMYYKGASNNHNGSELDKLFDIKYSETFFCKLILLFLYENI PRLRYFHILGINIVSEPQNYDYNESILKKTFRGITLYKGGWHSEKYLANKQDVNLNTSEFKIAKEDKNFIDLAK SIEEDTNSVSLHVRGDYLNISPTDHYQFGGVAATTNYKNAVSMYMLRNKQAHFYFSDDITWCKAEYKDL MPTFIECNKNKSWRDMMLMSLCTNHIANSTFSWWGAWLSTKNGITICTEFHNVVTRDIYPETWVQL	129	130
						MIIVRLMGGMGNGQLFYATAFALSKRKSEPLVLDTRFFDHYTLHGGYKLDHFNISARILSKEEESLYPNWQA NLLRYPIIDRAFKKWHVERQFTYQDRIYRMKRGQALLGYWQSELYFQYRKEISAEFTLKEQSSVTAQQISV AMQGGNSVAVHRRGDYLSNPALRTHGICSLGYNNHAMSLNINERINDAQFYIFSDDIWAKENIKIGKTSK NLIFIEGESVETDFWLMTQSKHHIIANSFWSWWGAWLANNTDEQLVICPSPWFDKLNSETDLIPKSWIRLN KDLPV		
Salmonella enterica	WP_00028 6641.1	446208786	protein [Salmonella enterica]	25.66	25.66	MYSLSGGLGNQMFQYAAAYILKQYFQSTTLVLDSDSYYSQPKRD TVRSLELNQFNISYDRFSFTDEKEKILL RKFKRNPFPKKISEILSIALFGKYALSDSAFYAVETIKNIDKACLSFYQDADLLNKHQKILPLFELRDDLDICKN LDVYPLILNNNTTALHRRGDYLTNQHAAYHGVLDTSYNNAMAYVERERGKQNFIFSDDKWAQKAF L GNENCYVNNNGDYDYSALDMLMSLCKNNIIANSFWSWWGAWLNKSEDKLVISPKQFWFLGNNETSLRNAS WIIIL	130	PCT/US2015/030823
						MLIVKVGIGGNQMFQYFYKYLOKNNDVFLDISDYKVHNNHNGFELIDVFNIEVKQADMMSKFKGHVSSK NSIFRYLTSKLFRNIIIGYSEFMDNSGISVRNEKILTDHYFIFGWQDVLVYLOSVEEIEKEAFNFKNVAIGKQNL E LISLSESVESVHIRKGDYANNSDLSIDCILEYEEAMKIIDSKVSEPLYFIFSDDIWCKQKFGKRDNLIVD WNIACKSYIDMLLMMSCKHHIIANSFWSWWGAWLNNSKIVICPKTWDRKKNENHLLNDWIAI		
Carnobacterium sp. WN1359	YP_008718 688.1	554649642	glycosyl transferase family 11 [Carnobacterium sp. WN1359]		25.59			

Mesotoga prima MesG1.Ag.4.2; Mesotoga prima	YP_006346 113.1	389844033	glycosyl transferase family protein [Mesotoga prima MesG1.Ag.4.2]	25.5	MRVWFGGGLGNQMFQYGLYCLKNNQNEVKADCTQYSTTPMNNGFELERLNLDAHANLDVSKLTG GNRLSPRKVWKLFRPKVYFEEKIPFSDPDLKGNRRYKGVWQNMNYLEPCAKELRDVFTFAFSSDNN KRLADEIAKVEAVGVHFRRGDFLKSSNLGLFGGICSDQYLRATQIMIENTVVEPVYFVFCDDPQWAKNSFSD ARFTVIDWNIIGSNYSYRDMQLMSLCKHNIIANSTFSWWAAWLNRPNRVTIAPERMVNRDLDFSGIFPND WIRLQG	WO 2015/175801
Clostridium sp. KLE 1755	WP_02163 9228.1	545399562	glycosyltransferase, family 11 [Clostridium sp. KLE 1755]	25.49	MIVLKLQGGGLGNQMFYAFARTIQEQKKKLLDTSDFQYDQKQREYSLGHFILNENIEDSSGKFNLWYDQR KNPLLKVGKFWPKFQFQTLKLFYVWDYAKYIPVDVSKKHKNILLHGLWQSDKYFSQISEIRKEFAVKDEP SQGNKAWLERISSANAVCVHIRRGDFLAKGSVLLTCSNYYLKAMEIISKVNEPEFFIFSDJEDVKKIFEFPG YQITLVNQSNDPYEELRLMSKCKHFIIANSTFSWWSSLLSENEKIVAPRLWYSDGRDTSALMRDEWIIIDN E	140
Bacteroides plebeius CAG:211	WP_02205 2991.1	547321746	glycosyltransferase family 11 [Bacteroides plebeius CAG:211]	25.42	MDLVTLSGGLGNQMFQAFYWALKRGGKVFYKKNLAAKEHNGYELQTLFGVEEKCVDGLWMTRLLGC PLLGKILKILPHKIRERVLYNYSYLPFLERNGLHWGVYQSEKYFQDVADDIRRIFCFDHLNLNPAATSAALK CMSEQVAVSVHRRGDYLLPCNVATYGGGLCTVEYENAIRYVYKERYPQAVFVFSDDLWDVYRENIPISAGKM VFVDWNRGKDSWQDMFLMSKCHHILANSSFSWWGAWLNTHPEKLVIAPERWANCAPADALPDGWV RIEGVSRR	141
Treponema lecanitholiticum; Treponema lecanitholiticum ATCC 700332	WP_02168 6002.1	545448980	glycosyltransferase family 11 [Treponema lecanitholiticum]	25.4	MAIKIVISGGLGNQMFYAFACALQKCGHKVYVDTSLYRKATVHSGIDFCHNGLETERLFGIKFDEADTAD VRLSTSAEGLNRRIRRYFTKTHYIDTVFKYTPPELLSKNDCCYLEGYWQTEKYFLPIEKDIRRLFTFRPTLSEKS AAVQSALQAQAAVLSASIHVRRGDFLTKTLNVCTETYYNNAIKYAVKKHAYSRFYIFSDDIPWCREHLFCF NAHAVFIDWNTGNDSWQDMALMSMCRNCNIIANSSFSWWAAWLNNASDKTVLAPAIWNRRLQLEVVDY YGYDSDIVPESWIRIPID	142
Bacteroides eggerthii; Bacteroides eggerthii DSM 20697	WP_00429 1980.1	490419682	glycosyl transferase [Bacteroides eggerthii]	25.34	MRLIKMTGGGLGNQMFYAFYLRMKKRHTNTRIDLSMMHYNVHGYEMHVRVFNLPKTEFCINQPLKKVIE FLFFKKIYERKQDPSSLLPFDKKYLWPLLYFKGYQSERFFADMENDIRIAFTFNSDLFNEKTQAMLTQIKHNE HAVSLHRRGDYLEPKHWKTTGSVCQLPYLNAITEMNKRIEQSYVYFSDDIWVKNLPLPQAVFIDWVK GAESWQDMMLMSHCHRHIIICNSTFSWWGAWLNPRENKTVMPEWRFQHCDDTPNIYPDGWIKVPVN	143
Bacteroides stercoris; Bacteroides stercoris ATCC 43183	WP_00565 6005.1	491891563	glycosyl transferase [Bacteroides stercoris]	25.34	MRIKMTGGGLGNQMFYAFYMRMKKHYSNTRIDLSMMHYKAHNGYEMHVRVFNLPPIEFIRINQPLKKVIEF LFFKKIYERKQVPSLLVPYDKYFWPLLYFKGYQSERFFADMADDIRKAFTFNPRLSNRKTKEMSEQIDHDE NAVSIHVRRGDYLEPKYWKTTGCVQCLPYLNAIAEMNKRIEQSYVYFSDDIWVKNLPLPKAFFIDWVK GAESWQDMMLMSRCHRHIIICNSTFSWWGAWLNPRENKTVMPEWRFHRCETPDICPDKWIKVPINQPD SIQ	4

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WO 2015/175801	146	glycosyl transferase 11 [Butyrivibrio proteoclasticus B316]	302671882	YP_003831 842.1	Butyrivibrio proteoclasticus; Butyrivibrio proteoclasticus B316	MIIIIQLKGGMGNQMFQYALYRQLKLGREVKIDDETGFVDDLRIPVLQRFGISYDKATREEIVKLTDSKMDI FSRIRRLTGRKTRIDEESGIFDPRILEVEDAYLVGYWQSDKYFANEVEKEIREAFKRPQEVQMDSVSWTI LQIECCESVSLHIRTIDYIDEHHIHNICTEKYKSAIDEVRNQPSAVFFITDDKDWCRQHFRGPNFEFVD LDEINTDIAEMTLMRSCKHILANSFSFWAAWLNNDNPGKVIAPSKW/INNRKMDDIYARMKKIAI
						25.34
147	147	alpha-1,2-fucosyltransferase [Roseobacter sp. GAI101]	495504071	WP_00822 8724.1	Roseobacter sp. GAI101	MSPVHFPSDRLLRYEHLNSLWKTAMIYTRLLARLGNQMFOYAAGRGLAARLGVDFTVDSRRVHKGDDGV LTRVFDLDWAAPENMPPAQHERPLAYYAWRGLRRDPKIYRENGLGYNAAFETLPDNTYLHGYWQCERYFA HIADDIRAAEFVPRHPMSAQADMARRIASGPSVSLHVRRGDYLTVGAHGICDQTYDAALAAMVQMGLPSP TVYFSDDPQWAKDNLPLTTFEKVVWDFNGPDSYEDMRLMSLCQHNVIANSSFSWWGAWLNNANPQKRV AGPANWFSPNPKLSNPDIPLSRWIRI
						25.34
148	148	protein [Prevotella oris]	490511493	WP_00437 7401.1	Prevotella oris; Prevotella oris C735	MGQDMYSRIFGGGLGNQLFQVATARA VSLRQGVLELVDTRLAPPGSHWAFGLDHFNISARIAESELPPSKD NFFKYVMWRAFGHDPAFMRERGLGYQSRQAQPDGTYLHGYFQSERYFADVL D HLENELRIVTPPDTRNA EYADRIASAGHTVSLHVRRGDYVETSKNSTHATCDEAYYLRALARLSEKSDLKVFVSDDP EWVRDNKLPL YDTPVGHNGDPKPHEDLRMLSCCSDHVIANSTFSWWGGWLD RRPPEARVVGPAKWFFNNPKLVNPDILPE RWIAI
						25.33
149	149	protein [Prevotella oulorum]	490514606	WP_00438 0180.1	Prevotella oulorum; Prevotella oulorum F0390	MKIIVRIIGGLGNQMFOYALALALAKOQOENEEVKLDLSAFRGYKKGFGQLVQCFTTLPAAATWQEVQAQLA WYYPHYQLWRGLGHRVLPVCKTMLKEPDNGAFLEVLQRKGDAYYEGCWQDERYFESHYRPAILOAFTPTF TNPRNLAMQOQINTTESVAIHVRRGDYLHDALFRNTCGLAYFQRAITCILQHVHPVYFVFSDDMAWCRQ HIQPLLOTNEAVFVDWNVHKGASICDLHMLTLCRHHIANSSFSWWGAWLSPHQAGWIIAPKQWYAHEEK MSPAERWLKL
						25.33
PCT/US2015/030823	149	protein [Spirosoma panaciterrae]	522084965	WP_02059 6174.1	Spirosoma panaciterrae	MNRRVAVQLKGGGLGNQLFQYALGRRLSLQLFAELLFDCSVLENRIPVTNFTFRSFDLDMFRIAGRVA TPDL PLFPKSASIRSPWPHLVQLARLWKQGYVYVERGFAYNP KMLRQLSDRVWNGYWSQYRYFEDIAATLRAD CSFPDPLPDSAVGLAGQINATNSICLHIRTDTFLQVPLHQVSNADYVVGRAIYMAERVNDPHFFVFSDDIAW CQTNLRISYPVVPNLAGPKNSLHFLRMRYCKHFITANSTFSWWAAWLPSEPSDGKVIPTPQTWFSDSRSI DDLIPANWIRL
						25.33

							WO 2015/175801
Pseudorhodobacter ferrugineus	WP_02270 5649.1	550957292	alpha-1,2- fucosyltransferase [Pseudorhodobacter ferrugineus]	25.17			MIVMQIKGGLGNQMFQYAAAGRALSLQTGMPLHLDRYYRREREHGYGLGAFNIEASPLDESLLPPLPRESPL AWLIWRLGRRGNLVRENGMGFNPTLSNVTKPAWITGYFQSERFYAAHAATIRAEITPVAAPDLVNARWL AEIAAEPRVSLHVRGVDYRDAAAAGHGSCTPAYERALAHITARMGTAPVYAFSDDPAWVRENRLRP AEIRVPGHNDTAGNVEDLRLMSACRHHIVANSSFSWWGAWLNPRADKIVASPARWFAFPFTNPDIWPE AWARIEG
Escherichia coli; Escherichia coli O127:H6 str. E2348/69	YP_002329 683.1	215487252	fucosyltransferase [Escherichia coli O127:H6 str. E2348/69]	25.16			MMYCCSLGGLGNQMFQYAAAYILKQHFPDTILVLDSSYFNQPKDQIRHLELDQKIFIDRFSSKDEKVKIN RLRKHKPLPLNSFLQFTAKLCKNYSINDASYNPESIKNIDVACLSFYQDSKLLNEHRDLPLFEIRDDLRVL CHNLQYSLITDSKNTSIHVRGVDYRDAAAAGHGSCTPAYERALAHITARMGTAPVYAFSDDPAWVRENRLRP KYSNCLVADADENKESVIDMYLMSLCNNNIANSTYSWWGAWLNPRADKIVASPARWFAFPFTNPDIWPE WIAM
Lachnospiraceae bacterium 3_1_57FAA_CT1	WP_01635 9991.1	511537894	protein [Lachnospiraceae bacterium 3_1_57FAA_CT1]	25.16			MIIKVMGGLGNQMQYALYEKFSIGKNVKLDISWFEEDSSVQEKVFARRSLELRQFKDLQFDTCSAEKEA LLGSGILGKLERKLIPARNKHYESDIHSEVFNMSDAYLEGHWACEKYHDMPLLQEKIQIFPESANSQNT VKRMAKAENSVSIIHRRGDYLDPENEAAMFGGICTNSYKAAEYIKSRVDPDTHFYLFSDDTAYLRENYHGDY TIVDWNKGEDSFYDMELMSCCRHNICANSTSEFWGARLNRTPDKIVIRPAKHKNQSEIEPQLLHELWDNW VIIDGGRIV
Butyrivibrio fibrisolvens	WP_02275 5397.1	551010878	glycosyl transferase [Butyrivibrio fibrisolvens]	25.09			MKPLVSLIVPVLNVEKYLEQCLTSISSQTYDNFEVLVWGKCIDNSENICKWCEKDHFRFIEPQLKSCLGARN VGIDAAKGEYIAFCSDDDCITSDFLSCFVDTALKNSDIVETQFTLQDNLSPIYDYDRNHLGHGFLFETSA PSVWYFVVRDIFTNNLHYPERFGEDISMYSLFSYCNKIDYVEKPTLYRQVPSLLMNNPQGKRKRYESLF DIIDFVTNEFTRLLFQKSWLKLFLQLEMHSAISDSATSDDDEAISMQRQESGYLKKVPVKNTIFEVTLGWG GEIVSSIAKENTLHGVSNNMFNRYFFELLESTRKKLEEMIINFSPDIFLIDISEADYLSYKGNLGTFFVKNW KIGSFIMKMIQTHSNSSIFLLENYMQQAPDHVDNTNELLKMLYDDIKINHPDIICISAPADILNRSSEPELPCI YQLKLVSCLKLHTMYSVINCVCETKGGNGQIFQYFYSKYIEKMTGYRPLLHIGFFDYVKAIPGGTKRIFSLDKLF PDIEITTSKIPCSHVVEKSFISNPGSDIFRYGYWQDIRYFSDVKDEVLESFNVDTSMSKDDVIDFADTIRNANS IAMHIRQDYLNENNVSLEFQLSIDYKSAVDMIRKEYADDLVLFISSDDPEYANSIADSFIEGFVMPPLHKDY EDLYLTLAHHHIANSTFSLWGALLSARKDGIRIAPRNWFKGTPATNLYPDKWLLI
Anaeromusa acidaminophila	WP_01870 2959.1	517532751	protein [Anaeromusa acidaminophila]	25.08			MFCVRIYGGGLGNQMFQYALGRAMAKHYSETAAFDLSWYEQIKPGFEASVCQYNIELSRKDRPKAWYEPIL KRISRHTDKLEMMWFGLFEKHYDYDSTVFERGLCKKNITLDGYWQSYKFSYSAIEDLRLRELTIPKEREELIISRS LPENSVSIIHVRGVDYRDAAAAGHGSCTPAYERALAHITARMGTAPVYAFSDDPAWVRENRLRP GRELGLFDYEELIIMSRCKHNIIMANSTFSWWGAWLNPNKVVIAPRKWFRHKKIKVNDLFPSSWVVL

WO 2015/175801	163	MDIVIFENGLGNQMSQAFYLAKKNDNLNCHVIFDPKSTNVHNGAELKRVFGIELNRNYLDKIISFYGYIFNKRIVNKLFLSVGIRMIYEPKNYDYREELLKPSNFIISFYWGWHSEKYFKDIELEVKVKFKPEVTNSPYFTWFNIKIFLDNNSVSIHRRG DYLDKPSDPYQFNGVCTIDYKAILYKERILEPNFYIFSDININWCMKTFGTENMYVYDCNKGKDSWRDMYLMSECRHHINANSTFSWWAAWLSYSPSNGIVLHPKYFIKDIETKDYYPQKWIMIE	glycosyl transferase family 11 [Bacteroides sp. 2_1_16]	496044479	WP_00876 8986.1	Bacteroides sp. 2_1_16	25.08		
163		MDKVVVHLTGGLGNQMFOYALGRSISNRNCP LLNTSFYDTYDKFCGLSRYNVKAFFIKNSYNNKYYRYVIRLLSRYGACVFGSYEKKIFSDEKVKRSCSVYGTWQSYGYFSDIRDLRDDYEMVVGCLSEEEVEKYVSDIKRVDSVSLHRRGDYFDNKRQSHIGILTMEYKAMSLPDSVVFVSDDIEWVRENLTNTNIVVWLESDN PENEIYMSLCKNNIISNSTFSWWGAWLNKNKYKKVAPRMWYKDNQSSDMLPDSWCLI	glycosyl transferase family protein [Chlorobium phaeobacteroides BS1]	189500849	YP_001960 319.1	Chlorobium phaeobacteroides BS1	25.08		
164		MIVISMGGGLGNQMFEYAFYTLKHLPKSEIKVDTKYAFPYSHNGIEVF KIFGLNPPEANWKEVHSLVKTYPIEGNKAHFIFKFLYRILRKANLVREPTSCCKQKDFTEFYNSEFFELPQNKSFYLYGPFVNYNYFAAIHNEIMDLTYFPEITDVTNIEYKRIESSHSIHIRRGDYITEGVP LVPDYYREALVYINKKIEDPHFFVFTDDKDYCKSLFSDNQNFTIVEGNTGANSFRDMQLMSLCKHNIJANSTFSFWGAFLNKSEKIVAPNIAPAFKDCSCP YICPDWII	protein [Treponema bryantii]	551312724	WP_02293 2606.1	Treponema bryantii	25.08		
165		MVIKLFGLGNQMFIYAAAKGIAQISNQKLTFTDITGTG FEDDSRFRVVELKQNLVSQESRRWMSFRYPLGRILRKISRKIGFCIPLVNFKFIVEKPYHFQNEIMIRIASFSSYILEGYFSYKFKIEAQIREDFKFTKEVIGSVEKEASFITNSRYTPVAIGVRRYSEMKGEGELAWEHYDAAIKYANKVPLNIFIVFSEDIDWVKKNLKLDYPVYFVTSKKGELAAIQDMYLMSLCNHHIISNSFYWWGAYLASTNNHIVIA PSVFLNKDCTPIDWVII	LPS biosynthesis alpha-1,2-fucosyltransferase [Bacteroides fragilis 638R]	375358171	YP_005110 943.1	Bacteroides fragilis 638R	25		
166		MSGGLGNQMFOYALYMKLTAMGREVKFDDINEYRGEKAWPIMLAVFGIEYPRATWDEIVAFDGSMDFSKRLKRLFRGRHPYEVVEQGFYDPKVLSEFNMYLKGFSQSQRYFEDILEEVQETFRPELKMNLPAPLYETTEK YLLRIEGCNNAVGLHMYRGDSRSEELYDGICTEKYEGAVRFIQDKCPDAKFFISNEPKWVKGWVISLMKS QIREDMSREEIRALEDHVLIENNTEYTG YLDMFLMSRCRHNIIISNSFSWWAAFINENPDKLVTAPSRWVN GVPSEDVYVKGMTLIDEKGRVERTIKE	protein [Firmicutes bacterium CAG:534]	547951298	WP_02235 2105.1	Firmicutes bacterium CAG:534	25		
PCT/US2015/030823		MVIVKIGDGLGNQMFNVCYGVSAKHNDNTLLDTSVDNSTRTYDLDKFNIDFTDRESFTNKGGFFHKVYKRLRRSLKNVIVESRTENCPVCVLDVYRRKFIKDKYLGHYFQNLVYFKTCKEDIMRQFTPKPEPSAKADELIHREFA TENTCSVHVVRGGDIKPLSIKYKDALDKIGEAKKDMRFVFSNVRNLAEEYKELGVDAEFWDLGEFTDIEELF LMKACRRHILSDSTFSRWAALLDEKSEEVFPFSPDADKIYMPWEWIMEEYDGNEEKR	glycosyl transferase family 11 [Firmicutes bacterium CAG:882]	547971670	WP_02236 8748.1	Firmicutes bacterium CAG:882	25		

Vibrio parahaemolyticus; Vibrio 10329; Vibrio parahaemolyticus 10296; Vibrio parahaemolyticus 12310; Vibrio parahaemolyticus 10290	WP_00549 6882.1	491639353	glycosyl transferase family 11 [Vibrio parahaemolyticus]	25	MVIVKVGGLGNLFQYAGCAISNRSLCELLDTSFYKQSLRKYELDKFNIAKAVATQKEVFCGGGDDLLS RFLKLNLSLFFPNYKEKESLVYLAESHCKSGFLDGYWQNPQYFSDIKDELVKQIVPIMPSPSPALEWQNI INTKNCVSLHVRGDDYVNNNAHTNSVHGCDLSYREAITNIHETVEKPKFFVFSDSDISWCKDNLGSLGHFTYV DNTLSAIDDLMLMSFCEHHIANSTFSWWGAWLNDHGHTIAPKRWFSSVERNNKDLFPEKWLL	WO 2015/175801 168
	WP_00646 3714.1	493509348	glycosyl transferase family protein [Herbaspirillum frisingense]	24.92	MIVSRIGLGNQMFQYAAAGRALALRRGVPAFIDSRAFADYKTHAFGMQCFCADQTEAPSRLLPNPPAEGR LQRLRRFLPNPLRVYTEKTFDEAVLSLPDGYLDGYWQSEKYFADFADDIRKDFAVKAAAPSAPNQAWLEL IGRTHSVSLHRRGDDYVSNAAAAVHGTCDLGYERAVAHQHQTGOAPELVFSDDDLDWVATNLQLPYT MHLVRDNDAAATNFEDLRLMTACRHHIVANSFSWWGAWLDGRSESIITAPARWFWADTPDARDLVQQR WVRL	169
Rhizobium sp. CF080	WP_00775 9661.1	495034125	Glycosyl transferase family 11 [Rhizobium sp. CF080]	24.92	MIITRILGGLGNQMFQYAAAGRALAIAEALKLDIEMGAYKLRPFDALQFNIAKAAQPDEVPAKPKRGLLR KFTSAFKPDRSSCERVENGLTFDSRVPALRGLSLGYWQSEQYFASSADAIRSDFSLSKPLGPARDQVLARI GAATTVPVSIHVRGDDYVTPSANAVHGTCEPPWYHEAMRRMLDRAGDASFEVSDPEQWARDNLQSSRP MVFIQPNNGRDEDMHLMAACHAHIANSSFSWWGAWLNPRPNKHVIAPIRQWFRAPDKDDRDIVPA TWERL	170
Verrucomicrobium spinosum	WP_00995 9380.1	497645196	glycosyl transferase, family 11 [Verrucomicrobi um spinosum]	24.85	MVISHISEGLGNQMFQYAAAGRRSLYHLGTTKLDDYHYRLHPFRSFQLDRELTSPITDAEISHLCPLEGLAR AIRARLPGLRGATRLRLGNLGLSPYQPRHLSFKETPKQPLIGKVVSEHFFHFDPDVLECPDNVCLVGYW QDERYFGEIRDILLRELTLSPPAGATKAVLERIQRSSVSLHVRGDKTKSSSYHCTSLCYCLAAMSEMRARL QAPTFVFSDDDWVVRQIPCCSSSVIHVDHNRADVSDFRLMKSCDHIIHIIASSSLSWWAAAWLGTNENSF VFSPPADRWLNFNSHFTADVLPPhWIQLDGSLLPAQ	171
Fibrella aestuarina; Fibrella aestuarina BUZ 2	YP_007319 049.1	436833833	glycosyl transferase family 11 [Fibrella aestuarina BUZ 2]	24.83	MTANRVLVNSPMVIAKITSGNLGFQYALGRHLALQGNLSLWFDLRYFHQEVATDTPRKFKLDRFNVRYN LLDSSPWLYASKATRLPLGRSLRLIDTRFEADHFDPTVIRPAAPLTILWGFVQSEKYFAQSTPQIRQELTFN RPLSDTFVGYQQQIEQAEVPSVHVRGDDYVTHPEFSQSFVGLAYYQKALAHQLDLPNATLFFSDDPD WVRANIVTEQPHVFVQNSGPDADVDLQLMSLCHHHVIANSSFSWWGAWLNPRPNKHVIAPIRQWFRAN KPWDTKDILPSGWRL	PCT/US2015/030823 2

Rhodobacter sp. CACIA14H1	WP_02366 5745.1	563380195	alpha-1,2- fucosyltransferase [Rhodobacter sp. CACIA14H1]	24.83	MIHMRLVGGLGNQLFQYACGRAVALRHGTELVDLTRELSRGAHAHAFGLDHFHFAIRARMGASADLPSPRSR VLAYGLWRAGFMAPRFLRERGLGNPAVLAAGDGTLYHGYFQSEAYFRDVPQIRPELEIVTPSPDDNLRW ASRIAGDDRAVSLHVRGDDYASAKGQVHGTCDADYARAAARAGIDPRLYVFSDDPHWARDNLA LDAETVLDHNPPGAAVEDMRMLMGVCRHHIIANSSFSWWGAWRNPAGKVVVAPVRWFADPKLHNPD CPPEWLRV	WO 2015/175801
Rhodopirellula baltica SH 1;Rhodopirellula baltica	NP_868779 .1	32475785	fucosyl transferase [Rhodopirellula baltica SH 1]	24.83	MATSAHLHLSDEKQTLDSKASDRDCATTEASADKCTISISGGLGNQMLQYAAGRALSIIHDCSLQLDLKF YSSKRHSYELDAFPIQAHRSKPSFFSQILSKIQESKHVPTYQEQRKFDPAFFNTEPPVKIRGYFFSEKYFSPY ADQIRTELTPPIPPDOPARDMAIRLKECVSTSLHVRGDDYVTNANARQRFWCCTSEYFEAAIERLPTDSTV FVFSDDIEWAKQNISSRTTVYVNDLKKAGSPETGLRDLWLMTTHAKSHIIANSSFSWWGAWLANSEANLTIA PKKWFNDPEIDSDIVPSSWHRI	174
Spirosoma spitsbergense	WP_02060 4054.1	522092845	protein [Spirosoma spitsbergense]	24.83	MVVVELMGGGLGNQMFQYAFGMQLAHQRQDTLTVSTFLLSNKLLANLRNYRPFELCIFGIDKPKASPFNL LRALLPFDLNTSLRETDDPEAVIPAASARIVCVGYWQSEHYFEEVTVHVREKFIRQPFNSFTSRLANNLNGI PNSVVFHIRRGDDYVTNKGANAHGGLCDRTTYERAVTFMREHLENPLFFIFSDDLLEWVSEQLGPILEPATYVG GNQKNDSWQDMYLMSLCRHAIVANSFSWWGAWLSHASKIVVAPKEWFGKPLLPVKTNDLIPNSWIRI	175
uncultured bacterium	EKD71402. 1	406938106	protein ACD_46C00193 G0003 [uncultured bacterium]	24.76	MNAIIPRITGGIGNQLFIYAAARRMAIANSMNLVIDDTSGEKYDVLVKRFYQLEKFNITSRMAITPTELEPFSK IRRYLKRKINKTYPFAQRAYITQEKSGFDPRLLVFRPKGNVYLDGYWQSENYFKDIEGIRQDLIIKSPSDSLNIA TAERIKNTLAIAVHVRFDMVDISDSSNCQSNYYHTAIAKMEEKIPNAHYFISDKPVLARLAMPLPDDDRITIID HNIGDMNAYADLWLSLCKHFVIANSTFSWWGAWLSDNKEKIVAPDKITSGVTQWGFGLIPDEWIKL	176
Prevotella micans;Prevotella micans F0438	WP_00695 0883.1	494008437	protein [Prevotella micans]	24.75	MDVIVFNGLGNQMSQYAFYLEKRLNRNQTTVEVLNPRSTYELERLFGIPYRSNL MCRMVYKLLDKAYFSNHI RLKKILRTALNAVGIIRLIVEPITRNYSLSNFTHPGLTFYRGGWHSSELNFTSVVTELRRKFIPPSDDDEEFKRISAL IIRTOSISLHRRGDDYLDYSEYQGVCTEYERAEIYIRSHVENPVFFVSDDKKEYAINKFSGGDSFRIVDFNTGE NSWRDMQMLMSLCRHHILANSTFSWWGAWLDSAPEKIVLHPIYHMRDVPTRDFYPHNWIGISGE	177
Thermosynechococcus sp. NK55	AHB87954. 1	564737556	alpha-1,2- fucosyltransferase [Thermosynechococcus sp. NK55]	24.75	MIIVLYGGGLGNQMFQYAAAGLALSRLHAPVLRFDLDWFDGVRILHQGLELHRVFDLPLPRAAPSEMQRVLG SFSHPVLRRLVRRRLWLLPQGYALEPHFYWPGFEALGPAYLDGYWQSEYFSEYQDQAVRAAFRAQAP LDERNRQIVVEEAAACESVSLHVRGDDYVQDPVVRVRRVHGVDSLAYSYPRAVALLMERMREPRFVVFSDDPD WVRANLKLPAPIVIDHNHNRGEHSFRDMQMLMSACRHHILANSSFSWWGAWLNSQPHKLVIAPKRWFNVND DFDTRDLYCSGWTVL	3

Coleofasciculus chthonoplastes; Coleo fasciculus chthonoplastes PCC 7420	WP_00610 0814.1	493031416	Glycosyl transferase family 11 [Coleofasciculus chthonoplastes]	24.73	WO 2015/175801 1 MLS LNK NLFVHIPKSCILKEVYIMISFPNLGKGVRLGNQMFOYAFRLSTARRLGKVFYCPAWSGDSLFTLN DQEERVSQPEGITKQYRQGLNPGFSENAISIQDGTGTEISGFQSDKYDNDPDLVRQWFSLKKEKIASIRDRFSRL NFANSVGMHLRFGDVVGQILKRPMPRRSYKKALSYPNQELILVFSDEPERTKMLDGLSGNLFSLSGHKNY EDLYLMTKQCQHFICSVSTFSWWGAWLGGERTVYIPKEGQYRPGYGRKAEGVSCSWIEVQSLRGFLDDY RLVSRLKRLPKSLMNFY
Bacteroides gallinarum	WP_01866 6797.1	517496220	glycosyl transferase [Bacteroides gallinarum]	24.66	180 MRLIKMTGGLGNQMFIYAFYLRMKKRHTNTRIDLSMMHYNVHHGYEMHHVFNLPKTEFCINQPLKKVIE FLFFKKIYERQDSSNLLPDKKYFWPLLYFKGFYQSERFADMEMDIRKAFNLSGLFNEKTQTMLKQJHEHNE HAVSLHVRRGDYLEPKHWKTGSGVQLPYIYINAEAMNRRIEQPFYVYFSDDIWVWKENLPLPQAVFIDWN KGVESWQDMMMLMSHCRHHIICNSTFSWWGAWLNPKENKTVMIPERWFQHCETPNYIPAGWIKVPIN
Firmicutes bacterium CAG:882	WP_02236 7483.1	547967507	glycosyl transferase GT11 family [Firmicutes bacterium CAG:882]	24.66	181 MNNVEIMGGLGKQLFYAFSRYLQKLGKVNVLKDKDFTTIQFPENNGITKREFVLDKYNTRYVAAAGEKTYR DYCDENDYRDDYAGSDEVLYEGYWNIDFYNNVRKEMQEELKLPKPEFIDNSMAAVEKD MSSCNSVALHIR RSDYLTQVNAQIFEQLTQDYASAVSIEQYTHEKPVLYFSDDPEYAAENMKDFMGCRVTIMPPCEPYQDM YLMTRAKHNIANSTFSWWGATLNNANPDNITVAPSRWMKGRVTNLYHKDWITL
Bacteroides xylanisolvans; Bacteroides xylanisolvans CL03T12C04	WP_00802 1494.1	495296741	protein [Bacteroides xylanisolvans]	24.6	182 MIAVNVNAGLANQMFIYAFGRGLMAKGLDVCDFDQSNFKPRSQWAFELVRLQDAFSPIDIKVMPEGHFK WVFPSLPRNGLERRFQEFMKKWHNFIGDEVYIDEPYGYVDPMEKCATRNCIYKGFQSEKVFHCEDDI RKQFTFLPFDLKNIEVAAKMSQENSVAIHRLKGGDDYMQSELMGKGLCTDYVMKADYMRKHNNPHFY VFTDNPCWVKDNLPEFEVILVDWNEVSGKRNFRDMQIMSCAKHNIIGNSTYSWWAAWLNANQDKIVVG PKRFFNPINSFFSTCDIMCEDWISL
Geobacter sp. M18	YP_004197 726.1	322418503	glycosyl transferase family protein [Geobacter sp. M18]	24.58	183 MIGMVIFRAYNGLGNQMFOYALGRHLLALLNEAELKIDTTAFADDPDLREYELHRLKVQGSIAITPDEIAFFREM ENTHPQAYLRLTKQSRLEDPAILSARGNIYLHGFQWQTEKYFADIREILLDEFEPVPAGEDSIKYLSHMKATNA VALHVRSDYVSNPMTLRLHHGVLPIDYREAVRRRIAGMVPDPVFFISDDPQWAKDNIRLEYPAFCVDAHD ASNGHEDLRLMRNCKHFIANSFSWWGAWLSQNTGKVVAPLKWFAKPEIDTRDIVPLQWIRI
Ruegeria pomeroyi DSS-3	YP_168587.1	56698215	alpha-1,2-fucosyltransferase, [Ruegeria pomeroyi DSS-3]	24.57	PCT/US2015/030823 1 MITTRLHGRNLGNQMFOYAAARGLAARLGTQVALDTRLAESRGEVLTTRVFDLDAQPDQLPPLKGDGLLR HGAWRLLGLAPFRFRREHGLGYNAAIETWDDGTYLHGYWQSERYFAHIAARADFAFPAFSNSQNAEMAA RIGDTDAISLHVRRGDYVALAAHTLCDQRYAAALTRLEGVAGDPVYLFSDDPAWARDNLALPVQKVVV DFNGPETDFEDMRMLMSLRHNIIGNSSFSWWAAWLNALNAHPGKRIAPASWFGDAKLHNPDLPLPDWLKIE V

Lachnospiraceae bacterium 28-4	WP_01629 1997.1	511037973	protein [Lachnospiraceae bacterium 28-4]	24.52	MIIQLAGGLGNQMYYQKLLSLGKKVLDISWFEKRNQNVVARRELENYFKAEYEAETEEERKALVGEFGAGKKGKLPFGTRKIFRETEMYPHFIDFEDRVLGYFACEKYADIMEILQEQQFVPPSGNPNQKMAERIADESLSHIRRGDVLDAENMAMFGNICTEYAGAIEMKKYPSAHFFVFSDDIPYAKETYSGEEFTVVDINRGKDSFFDIWLMVSGCRHNICANSTFSFWGARLNRNKGKVVMRPFHKNQSKQFEPELMHLEWKGWVFIDNRGNIC	WO 2015/175801
Prevotella sp. CAG:1092	WP_02198 9703.1	547254188	glycosyl transferase family 11 [Prevotella sp. CAG:1092]	24.49	MRILVFTGGGLGNQMFEYAFYKHLKSCFPKESFYGHYGVKLEHYGLEINKWFDVTLPPAKWWTLPPVVGFLFYLYKCLVPNSKWLDLQREWKHKDAKVFFPKFTKQYFPKENGWLKWVDEASLCEKNKKLLQVHDEETCFVHVRRGDYLASNFKSIFEGCCTLDYYKRALEYMKNKNNPKVRFICFSDDLWMMRKNLPMDDSAIYVDWNTGTDSPLDMYIMMSQCDNGIIANSFSFYWGAYLGGKTTVIYPQKWVNMEGGNPNI FMD EWLGM	186
Spirosoma luteum	WP_01861 8567.1	517447743	protein [Spirosoma luteum]	24.41	MVISLGGGLGNQLFOYAFGLKLAALQQLTELRLERHLLSKAIARLQYTPRTYELDTFGVEAPAAASLMDTVSCLSRVALSDKTAALLRESTLTPNAINNLRVRDVVCLGYW/OSEEFERPATEQLRKHLVFRKNPAQSRSMADTILSCQNAAFVHIRRGDYVTNTHANQHHLGCDVSYRRACEYVKECIPDVQFFVFSDDPDWAKRELGIHLQPARFIDHNRGADSWQDMYMLMSLCRHAIVANSFSWGWALNPVAERLVVAPGQWFWFNQPVLSQQIIPPHW/HCI	187
Marinomonas posidonica IVIA-Po-181; Marinomonas posidonica	YP_004480 472.1	333906886	glycosyl transferase family protein [Marinomonas posidonica IVIA- Po-181]	24.34	MIIIDLGGGLGNQMFOYACARSLSIELNLPKVYVGLASQTVHNGYELNRVGLDLEFATENDMQKNLGGFFLSKPILRKIFSKKPLNNLKQNFPPENSFNYSLSFYKDSGFLQGYWQTEKYELNHSQILKDFCFVNMDDDETNSIANDIQSGHSISIHVRRGDYLNLKAKAIHGHCSLDYILKAEFLQEKIGESRLFISDDPEWVSENIATRFSDVSVIQHNRGVKSFNDMRLMSMCDHHIIANSFSFWGWALNPQNKIIAPKNWVFTDKMINTIDLIPSSWILK	188
Bacteroides; Bacteroides sp. 4_3_47FAA; Bacteroides sp. 3_1_40A; Bacteroides dorei 5_1_36/D4; Bacteroides vulgatus PC510; Bacteroides dorei CL03T12C01; Bacteroides vulgatus dnLKV7	WP_00583 9979.1	492425792	glycosyl transferase family 11 [Bacteroides]	24.32	MKIVVFKGGLGNQLFOYAFYKYSRKDETFYFNDAWYVNSHNGFELDKYFKTDDLKCCSRFWIILFKTILSKLYHWKIYVVGVSVEYQYPNHLFOAGYFLDKKYYDENTIDFKHLLSEKNQSLKDIQNSNSVGVHRRGDYMTKQNLVIFGNICTQKYHDAIRITEKVNDAVFYVFSDDISWVQTHLIDIPNAVYVNWNTGESSIDMYMLMSSCKYNIANSSTFSYWAAARLNKKTNNMVIYPSKWYNTFTPDIFPESWCGI	189

WO 2015/175801	191	Candidatus Pelagibacter ubique	WP_02016 9431.1	519013556	protein [Candidatus Pelagibacter ubique]	24.32	MTIRIKLTGGLGNQMFAQATGFAIAKKNVRLSLDKYINRKLKNGFELQKIFNIYSKVSFLNKTLSFKSINFTE ILNRIDTTFYNFKEPHFYTSNIIILPKHSELDGYWQSELYNEFAATEIKRIFNFSGLDKSNLLVADDINRNSI SIHIRRGDFLLKQNNHHHTDLKEYYLKAINETSIFKNPKYFISDDTSWTVDNFVIDHPYIIVDINFGARSELD MYLMSLCKSNIIANSFSWWSAWLNINNKDKIYAPKNWFNDKSICTDDLLIPESWNIIIL
							MSVIINMACGLANRMFYAFYLYLQKEGYDAYVDYFTRADLVHENVDWLIRFPEATFRATARDIRKMG GHDCFSRLRRKLLPMITTKVLETSGAFEIILPPKNRDSYLLGAFQSAKMVESVDAEVRRIFTPEFESGKNQYFQ TRLAQENSGLHIRKGDYQERIWYKNTCGVEYYRKAVDLMEKEVDSPSYFTDNPAAWKENLSWLEYKL VDGNPGSGWGSHCDMQLMSLCKHNIISNSSFSSWWGAYLNTLNKIWCPRIWFPNPESTKDFSSNPILLAEG WISL
	192	Butyrivibrio fibrisolvans	WP_02275 6327.1	551011911	protein [Butyrivibrio fibrisolvans]	24.29	MIIKLQGGILGNQLFLYGLYKLNKLRDVKMDIESGFEGDELKPKCLDCMNLEYAIAATRDVTDIRDSYMDI FSRIRRKITGRKTFDYVEPEDGNYDPKVLMTKAYLNGYFQSEKYFGDEESVKALKDELTKGKEDILTSTDLLTKI YHDIKNSVSLHIRRGDYLTPIGIIETYGGICTDEYYDKAIAIMIRETTFPEARFFISNDIEWCKEKFAGDKNIIIFV NTIGINLDSEDNIKIGSKDKIDISEYRDLAELYLMSACKHHILANSSFSWWGAWLSDHEGMTIAPSKWLNKN MTDIYTKDMLLI
							MVTVKIGDMGNQMYNACGYAAAKRSGEKLRDISECDNSTLRDYELDHFRVYVYDEKESFPNRTFWQKL YKRLRRDIRYHVIRERDMYAVDARVFPARRGRYLHGYWQCLGYFEEYLLDLREMTFPAYEQTDVARELM QQFTQTPTCALHVRGGDLGGPNRAYFQQAIAARMQKEKPDVTFIVFTNDLPKAKECLDDGEARMRYIAEFGE ALSDIDEFFLMSACQNIISNSTYSTWAAAYLNTLPGRIVIVPKFHGVEQMALPDWIVLDGGACQKGEIDAV
	193	Roseburia hominis; Roseburia hominis A2-183	YP_004839 455.1	347532692	glycosyl transferase family protein [Roseburia hominis A2-183]	24.22	MATSVHPHLSGKQALDSKAAQQVCSTQAASASDRACITISISGGLGNQMLQYAAAGRALSIHHDCPLQLDLK FYSSKRHSRYELDAFPIQAQRWIKPSFFSQVLDKIQGESKAPTVEEQSKRFDRAFFDIELPARIRGYFFSEKYFL PYADQIRTELTPVPVLDQPARDMAQRLESGMSTSLHVRGDYVSNANARQRFWSCTSEYFEAAIEQMPAD STVFVSDDIEWAKQNISSRPTVYVNDLKLGPETGLRDLWLMTHAKSHIIANSFSFWGAWLSGSEA NLTIAPKKWFNDPEIDDSDIVPTSWRRI
							MATSVHPHLSGKQALDSKAAQQVCSTQAASASDRACITISISGGLGNQMLQYAAAGRALSIHHDCPLQLDLK FYSSKRHSRYELDAFPIQAQRWIKPSFFSQVLDKIQGESKAPTVEEQSKRFDRAFFDIELPARIRGYFFSEKYFL PYADQIRTELTPVPVLDQPARDMAQRLESGMSTSLHVRGDYVSNANARQRFWSCTSEYFEAAIEQMPAD STVFVSDDIEWAKQNISSRPTVYVNDLKLGPETGLRDLWLMTHAKSHIIANSFSFWGAWLSGSEA NLTIAPKKWFNDPEIDDSDIVPTSWRRI
PCT/US2015/030823	194	Rhodopirellula europaea; Rhodopirellula europaea 6C	WP_00865 9200.1	495934621	alpha-1,2- fucosyltransferase [Rhodopirellula europaea]	24.16	MVIAKITSGLGNQLFYALGRHLAIQNQTRLWFDRLRYHRTYETDTPRQKLDRESDYDLDYSPWLVSKA TRLLPGRSLRPLFDTRKEPHFLDPAVPAKAGAFITLDGFQWQSEGYFASNAATIRRELFTTRQPGPMYARYR QQIEQTQTPVSVHIRRGDYVSHPEFSQSGALDDTYQTALAQINGQFPDATTLLVFSDDPEWVRQHMIRFER PHVLVENTGPDADVDDLLQLMSLCHHHIIANSFSWWGAWLNPDPKRVIAPKQWFRNKPWNTADLPAG WVRL
							MVIAKITSGLGNQLFYALGRHLAIQNQTRLWFDRLRYHRTYETDTPRQKLDRESDYDLDYSPWLVSKA TRLLPGRSLRPLFDTRKEPHFLDPAVPAKAGAFITLDGFQWQSEGYFASNAATIRRELFTTRQPGPMYARYR QQIEQTQTPVSVHIRRGDYVSHPEFSQSGALDDTYQTALAQINGQFPDATTLLVFSDDPEWVRQHMIRFER PHVLVENTGPDADVDDLLQLMSLCHHHIIANSFSWWGAWLNPDPKRVIAPKQWFRNKPWNTADLPAG WVRL

Bacteroidetes;Capno cytophaga sp. oral taxon 329 str. F0087;Paraprevotella clara YIT 11840	WP_00861 8094.1	495893515	glycosyl transferase [Bacteroidetes]	24.15	197	WO 2015/175801	MRLIKMTGGGLGNQMIYAMYLKMKITIPDVRIDLSDMVHYQVHYGYEMNKVFHLPRTFCINRSLKKIEFL LFKTLERKQGGSLVPYTRKYHWPWYFKGFYQSEKYFAGIEKEVREAFVDIRASRRSLRAMQEIKAADPHA VSIHVRGDDYLLKHWKALGICGSSYYLNALELEKRVKHPHYFYFSEDLNWRQNLPLIKAEFIDWNKGE DSWQDMMMLMSHCHRHIIICNSTFSWWGAWLNLPLDKVIAPERWTQITDSDADVVPESWLKVSIG
	WP_01815 9152.1	516906936	protein [Smaragdicoccus niigatensis]	24.08			MADVVTLAGGLGNQLFQTAYAKNLEARGHRVTLDTGTVRWTRGLHIDPQICGLKILNATPPAPVPGRLAA TVLRRALATRLRFGPDGRIVRQRTLEFDEQYLNLSNPGRYRVEGYWQCERYFSDVGQTVRKVFLDMLGRH VSYNGLSRLPAMADPSSISLHVRRGDYVTANFIDPLALEYERALEELAVPSPRIFVFSDDLDWATRELGRICD VIPVEPDWTSHPGGEIFLMSQCSHHIANSFSWWGAWLDGRTSSRVVAPRQWFSLETYSARDIVPDRWT KV
Bacteroides fragilis CAG:558	WP_02201 2576.1	547279005	family 11 glycosyltransferase [Bacteroides fragilis CAG:558]	24.05	198		MIHLILGGGLGNQMFQAFARSLALQYNENISFNITLYKELKNEERFSLGHLNINTMCIVETPDENKRIWELF NKQIFHQIARKILPASIRWWWMSNRNIYANVCGPYKYPHPRHSQNTTIHGGFQSWKYFKEHQSMIAE LKVITPISEPNKKILKEIONSNCIVHRRGDFLSAQFSPHLEVCNKDYKAEKAMISSQJENPTFFIFSNTHEDLV WIRKYNIPQNSVVVDLNNPDYELRLMYNCKHILSNSSFSWWAQYLSKSNKIIAPKIWDKRKGIDFSDIY MPEWIIIK
	WP_02265 7592.1	550904402	protein [Desulfovibrio desulfuricans]	24.05			MSFIDVAAAIQRMALVKVDGGLSQMWQYALSLAVGKSSFTVKHDLWSFRHYAKDIRGIENRFFILNSVFT NINLRASENERLFFHIALNRYPDSCNFDPDILALKQPTLYGGYVNAQVVTSAKEIREAYVFAPAVEESNOA MLQTHAAPMPVAVHVRGDIYIGSMHEVLTPTRYFERAFKILAAALQPKPTFFVFSNGMEWTKKAFAGLPYD FVVDANDNDNDVAGDLFLMTQCKHFTISNSSLSWWGAWLSQRAENKTVIMPSKWRRGKSPIPGECMRV EGWHMCPVE
Hoeftlea phototrophica;Hoeftlea phototrophica DFL- 43	WP_00719 9917.1	494373839	alpha-1,2- fucosyltransferase, [Hoeftlea phototrophica]	24.05	200		MHGGGLGNQLFQYAVGRAVALRTGSELLDTRFTSSNPFQYDLGHFSIQAKVANSSELPPGKNRPLAYAW WRKFGSPRFVREQDLGYNARITIEADCYLHGYFOSQKYFEDIASILWKDLFRQAISGENASMAERIQSAP SVSMHIRRGDYLTSAKARSTHGAPDLGYGRALGEIRARSGSDPVVYLFSDDPDWVRNNMRMDANLVTVA INDGKTAFEDRLMSLCDHNIIVNSTFSWWGAWLNPSLDKIIVAPKRWFAADPKLSNPDIPTPPGWLRLGD
	WP_00203 0616.1	487957217	glycosyl transferase, family 11 [Vibrio cholerae]	24.04			MKIISFGGLGNQLFQYAFYLYLKDNDSDFGNIFLDFSFYESQNKRDVIRNFYGVDSLDIIKQSSYVRGKFLILKL INKFRFFNNLLEFVDKENGLEDITLLSTNKVFFDGYWQSYRVYKDYKSNIKELSFYDFKGNILEVRKKICQSSNV CMHVRRGDYVAEKNTKLHVGCSLQYRDALNNKNDNSIDHIFISDDIDWVKNNISFDIPVTVDVFGQ SVPDYAEMILLFSCGKHKVIANSTFSWWGAFSLDRNGVIVSPKKWFAKEEKNYDEIFIEGSLRL

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203						protein [Lachnospiraceae bacterium NK4A179]	551041074	WP_02278 4718.1	Lachnospiraceae bacterium NK4A179
						Glycosyl transferase family 11 [Cecembia lonarensis]	496476931	WP_00918 5692.1	Cecembia lonarensis;Cecembia lonarensis LW9
204						protein [Bacteroides ovatus]	490423336	WP_00429 5547.1	Bacteroides ovatus;Bacteroides ovatus ATCC 8483
205						glycosyltransferase family 11 [Bacteroides coprocola CAG:162]	547668508	WP_02212 5287.1	Bacteroides coprocola CAG:162
206						glycosyl transferase [Bacteroides dorei]	495110765	WP_00783 5585.1	Bacteroides dorei;Bacteroides dorei DSM 17855;Bacteroides dorei C103T12C01
PCT/US2015/030823						protein [Bacteroides]	494936920	WP_00766 2951.1	Bacteroides;Bacteroides des intestinalis DSM 17393;Bacteroides des intestinalis CAG:564

Lachnospiraceae bacterium A4	WP_016283022.1	511028838	protein [Lachnospiraceae bacterium A4]	23.95	MIVIHVMGGLGNLYQYALYEKLRALGREVKLDVYAYRQAEAGAEERWRALELEWLEGIRYEVCTAAERQQL LDNSMLRADRVRRRLTGRRDKTVRECAAYMPEIFEMDDVLYGFWGCEKYEDIIPLLQEKIVFPSSNPKN ADVLRAMAGENAVSVHIRKDYLTADGKRYMGICTDAYYKGAFTYTERVVPFYFSDDPFAKTFQFCE ENMHVVDWNTGRESLQDMALMSRCRHNICANSTFSIWGARLNRHPDKIMIRPLHHDNYEALDARTVHEY WKGWVLIDADGKV	WO 2015/175801
Phaeobacter gallaeciensis; Phaeobacter gallaeciensis DSM 17395 = CIP 105210	YP_006574665.1	399994425	protein PGA1_c33070 [Phaeobacter gallaeciensis DSM 17395 = CIP 105210]	23.91	MIITRLHGRILGNQMFQYAAAGRALADRLGVSVALDSRGAELRGEGLTVLRFVFDLDTATPDLPLRQRAPLGYA LWRGLGQHLGTGPKLRREVGLGYNPDFVDWSDNSYLHGYYWQSERFYAQSAERIRRDFTTPEYSNQNAE MAARIGETNAISLHVRRGDYLTAAHVLCDAQYEEAALAQVLDGLEGGQTVYVFSDDPQWAKENLPLPCDK VVVDFNGADTDYEDMRLMSLCKHNIIGNSFSWAAWLNQTPDRRVAGPTKWFGDPKLNNDILPPDW LRISV	209
Firmicutes bacterium CAG:791	WP_021849028.1	546362318	protein [Firmicutes bacterium CAG:791]	23.88	MSGILGNQMFQYALYLKRLSLGREVCDDKQYDEETFRNSSQKRRPKHLDIFGITYPSAGKEELEKLTGGA MDLPSRIRRKILGRKSLEKNDRDMFDPSPLEETEGYCGGQSPRYFAGAEVEVRKAFTPPEELLCPKEGCSR QEKMILEQSAEYAEIRKANCEAADRGVPGGGSASHLRFQDYVDKDIYGGICTDAYDYDAIRCLKERDPG MIFFVFSNDEEKAGEWIRYQAESENLRGHFVLVKGCODEDHGDLMLTLCRNHVIANSSFSWASFM CDAPDKMVFAPSIWNNQDGSSELARTDIADFMQRISPRGTRLSRPLSVIVTAYNVAPYIGRALDSVCGQ TWKNLEIAVDDGSDDETGAIDLRYAAGDSRIQVHTENRGVSAARNEGIAHARGEYGFVGDGDDRAHPAM YEAMIRGILSSGADMAVRYREVSAAETLDAEEQVASFDPVLRSVLLQQRDVAVQCIFRAGMAEEGKVL RSVWVWNLFRHLLRDNREPEGTSAEDIPFTTRALCLSKVLCPYELLYVYVNRQESIMNTGRAERTLTQEIP AWRTHLELLKESGLSDLAEESEYWFYRRMILSYEEERYRCSETAKEAKELQERILKHRDRILELAEAEHSFGRGD RERKLYVNSPRQYFLLSDLYEKTAVNWKNRPKDT	210
Butyrivibrio proteoclasticus; Butyrivibrio proteoclasticus B316	YP_003829733.1	302669773	glycosyl transferase 11 [Butyrivibrio proteoclasticus B316]	23.84	MRKRIALNGGLGNQMFQYAFARMILEDKHKCLIEFTGFTSTVNDKRLAIQNYNIHKYDFCNHEYNNKIRLLF QKIPFVAVLAGTYKEYSEYQLDPRVFLFNRYFYGYWQNKQYFENISNDIRNELSYIGNVSEKENALLNMLEA HNAIAIHVRRGDYDTQEGYNKIYISLSKEYKRAVSJACKELGDNINPLVYFSDDDIDWCKANLADIGNVTFVDNT ISSADIDMLMMKKSRCRLITANSTFSWWSAWLSDRDDKVLVLPDKWLQDEEKNTKLMKAFICDKWKIVPV	211
Bacteroides sp. 2_1_16	WP_008768245.1	496043738	protein [Bacteroides sp. 2_1_16]	23.81	>gi 496043738 ref WP_008768245.1 protein [Bacteroides sp. 2_1_16] MQVAVARIIGLGNQMFQYAFARALALRIDADLIDTQSGYKNDLFRNLLDSFCISYRKANCFQKY DYILGEKVKSLGKTHFSVIPFMKYISENTSCDFVDGLLKKHLSVYLDGYWQNEAYFKDYASIKKDFQFCQV NDLRTLSEAEIKKSIPTVAIGVRRYQELNSHQNTKTVDLDFYQKAINYESKVDNPTTFIFSEDEQWVKNMLEQ KSNFIMISPKEGNSALNDMYLISLCKHHIVSNSSFWWGWAWLANNKKNIVASDCLFNLPQSPIDSWIKF	2

Desulfomicrobium baculatum; Desulfomicrobium baculatum DSM 4028	YP_003159045.1	256830317	glycosyl transferase family protein [Desulfomicrobium baculatum DSM 4028]	23.76	>gi 256830317 ref YP_003159045.1 glycosyl transferase family protein [Desulfomicrobium baculatum DSM 4028]MAKIVTRIMGGIGNQLFCYAAARRLALVNHAELVIDDVTGFSRDRVYRRRYMLDHFNISARKATNYE RMEPEYRRGLAKYISKLLPFEREYIEQERIEFDPRFLEYRTYNNIYIDGLWQSENKFDVEDIIRDDLIKIPPT DLENINIAKKIKNIQNTIAMHVRWFDLPGINLGNVSTYYHRAIAMMEQINAPHYFLFSDNLEAVHSKLD LPEGRVTFVSNNDGDDNAYADLWLMSCQCKHITANSTFSWWGAWLGESRDSVVLVPRFSPDGGVTSWC FTGLIPERWEQVSSIR	213
Prevotella pleuritidis; Prevotella pleuritidis F0068	WP_021584236.1	545304945	galactoside 2-alpha-L-fucosyltransferase [Prevotella pleuritidis]	23.76	MDIVLIFNGLGNQMSQAFYLAQRQRNNHTVYCVFGPRTQYSLDKLFDIPYRHNNAVLLVLRALDKAHFSN HRWLRLLRPTLQLLGVKMIVEPLSRDFDMRHFTQKGVFVRGGWHSSELNFTAVADAVKRRFRFPEIQDA AVLAVIDRIKSCQSVSLHLRRGDYLGISEFQGVCTEAYYEHAIAFYEQIESPEYFVFSDDPTYAREQFGADPNF HIIDLNHGEDAWCDLLMMTQCRYNIIANSTFSWWGAWLNDNPVKVHPRYHLNGVETRDYPRNWICIE	214
Bacteroides sp. 1_1_14	WP_008763191.1	496038684	glycosyl transferase, family 11 [Bacteroides sp. 1_1_14]	23.75	MKVIWFNGNLGNQVFCYCKEFLHNKYPNETIKYYSNSRSPKICVEQYFRLSLPDRIDSKVRVFEFLGKFFR RIPLKFVPKWYCTRKSLNIEASYFEHYLDQKSFEEKEDSSWLKAKKPDNFSEKYLIFENLICNTNSVAVHIRRGD YIKPGSDYEDLSATDYEQAIKATEVYLDQFFESDDLEFVKNNFKGDNIIYVDCNRGADSYLDILLMSQAK INIIANSTFSYWGAYMNHKKVMYSDLWFRNESGRQMIPNIMLDSWICITKRK	215
Agromyces subbeticus	WP_022893737.1	551273588	protein [Agromyces subbeticus]	23.65	MVGRVGIARRQAAADVSDTDGEGLVAWRRTGEIVLGLQGGIGNQLFEWAFAMALRSIGRRVLFDAVRCRG DRPLMIGPLLPASDWLAAPVGLAGATKAGLLSDRSWPRLVQRSSGYDPSVLERLGGTSYLLGTFSQARY FDGVEHEVRAAVRALLGMLTPSGRRFADELADPHRVAVHVRRGDYVSDPNAAVRHGVLGAGYYDQAL EHAAALGHVRRRVWFSDDDLWDVREHLARDDDLCPADATRDHDGGEIALIASCATRIIANSSFSWWGGWLG APSSPAHPVIAPTWTFADGHSDAAELVPRDWVRL	216
Prevotella salivae; Prevotella salivae DSM 15606	WP_007133870.1	494220705	alpha-1,2-fucosyltransferase [Prevotella salivae]	23.59	MIATTLFEGGLGNQMFYATAKALSLHYRTPMAFNLRQGFQDYKYQRHLELNHFQCQLPTAKWITFNKYE LNIKIRRRIGRNLCPHYQFIKEKEPFHYEKRLFEFTKNIFLEGYWQSPRYFENYSDIIRDFQLKSILPHTITD ELQMLKGTGKPLVMLGIRRYQEVKDKDSYPLCNKDYAKAISHVQEQLPAPLVVFTQEQAWAMNLP TNANLYFVKEKDNAWATIADMYLMTQCQHAISNSTFYWWGAWLQHPHENHIVVAPNFINRDCVCN WILD	PCT/US2015/030823

Carnobacterium sp. WN1359	YP_008718687.1	554649641	glycosyl transferase [Carnobacterium sp. WN1359]	23.57		MIFVDLSEGLNQMFQAYSRYLQELYGGTLYLNTSSFKRNKSTRSYSLNFFLYENVKLP5KFRRVIYFY5K TIRMFIKVIRMINPYSDKYF5MIPYGFYVSSQVFKYLTVP TTKRHNIFFVMGTWQTNKYFQ5INDKIKDELKV KTEPNELNKKLITEINSQ5CVHIRLGDYTNPEFDYLFHVCTSDYYLKGMDYV5KVKEPNFYIFS5SSDIEWI KNNYFKYKYIDLNNDPFDLNNPDMYCNCKHFI5NSTFSWWAQFLSNNDKKIIVAP5KWQK5SNEAKDIY LDHWKLEIE	219	WO 2015/175801
						MLIQIAGGLGNQMQQYAMRKLKAGADRNIKIDTKWDEDKQ5GLAKRKL5EYFTGLPLPVC5ESERA RFTDRSVARKVVEKLVPGMG5RFT5CMYHPEIFELDKYIEGYFACQKYDDIMGELQELFVPTHPDEEINI KNMINLMNEMEMVPSV5HIRRGDYLDPENAA5LFGNIATDAYYDSAMEYFKAIDPDTHTFYIFTNDPEYAREK YADPGRYTIVDHTNGKYSLLDIQLM5HCHRGNICANSTFSFWGARLNRRKDKIPVRTLVMRNNQ5PVTPELMH EYWP5WVLVDKDGKVR		
Clostridium sp. KLE 1755	WP_022762290.1	551018062	glycosyl transferase [Butyrivibrio sp. AD3002]	23.55		MIVIRVMGGLGNQMQQYALYEKFKALGKETRLDTSWFDNASMQENVLARR5LELRFDDNLTYEACTPQER EALLGKEGFNKLERKLP5KNKHFE5EMFHP5EIFKLDNVYLEGHWACEKYHYDIMP5LLQ5KIIFPKTDNIQN NMLKNKMN5SV5HIRRGDYLDPENAA5MFGGICTDSYK5AEGYIRNRVTNP5HFLF5DDPAYLREHYKG EETVVDWNHGAD5FYDMELM5SCCKHNVCANSTFSFWGARLNRT5KKVIRPAKH5NSQ5QAEP5ERMHEL WENWV5IDEGRIV	220	
						MKFFVGGGLGNQLFQY5YRYLKKKYP5ERILGYP5SLKAHNG5IEDK5WFDIELPPT5YLYNKL5GILLYRVNRFLYNHGYRLLFCNRVYP5Q5MKHFFQWGDWQDY5IIKQINIF5ERSELPIGKENMEFLKKMETCNSISVHIRRG DYLTDLHIYGGICT5KY5REAIKFMEQEVEEP5FFF5DDCLYVET5FADIRNKIISHNRDD5R5F5DMYLM5MAH AKNMILANST5F5CWAAYLNR5AKIITPDRWVNT5DFS5KLEALPNEWIKIRV		
Paraprevotella xylaniphila; Paraprevotella xylaniphila YIT 11841	WP_008626629.1	495902050	glycosyl transferase [Paraprevotella xylaniphila]	23.47		MRLIKMTGGLGNQMFIYAMYLKMR5VFPDTRIDLSDMVHYRVHYGYEMNKVFNLPRT5FRINR5LKKIIEFL LFKTIL5ERKQ5G5LVPYIRKYHWPW5YFKG5FYQ5EEYFAGVEKEV5EAFV5DVRVRNRK5SLCAMQEIMADPD AV5IHVRRGDY5LQ5GKHWK5SLGCICQ5RSY5L5NAL5ELEKRV5H5PHYV5F5EDL5DWVRQY5LPLENAV5FIDW5NKG EDSWQDMMLM5SHCRH5IHCNSTFSWWGAWLNP5PDKIV5APERWTQT5TNSADV5VP5ESWLK5V5IG	221	
						MTDRALIAIVKGG5LGNQLFIYAAAR5MALRTGRQLY5LDAVRGY5LADDYGR5FR5LNR5FPI5EAL5MPEQWRVA STL5RHPRAK5LVRAL5NKYLPEAWR5FYVAER5GDT5RPGALWNHGR5NRV5KRV5TLMGYWQ5DEAY5FLDYAEL5LRREL GPPM5PDAPEVR5ARGER5FAG5TES5FL5HVRRCR5Y5PL5LDAGY5QKAV5DLACAE5L5NKP5VFMIFGDD5DIEW5VWN5NI DFRGAGY5ERQDY5DES5DELAD5LWLMTRCR5HAIAN5SSF5SWAAW5LGGAA5G5GR5HR5VWAP5Q5SGLAK5KCAK SWEAVDAQPE		
Thauera sp. 28	WP_002930798.1	489020296	glycosyl transferase family protein [Thauera sp. 28]	23.47			3	PCT/US2015/030823

Subdoligranulum variable; Subdoligranulum variable DSM 15176	WP_007048308.1	494107522	alpha-1,2-fucosyltransferase [Subdoligranulum variable]	23.44		MIYAEAGGLGNQMFIYAFARALGLRCGEAVTLLDRQDWRDGAHTACALEGLNLVPEVKILAEFGFAKRHLPRQNTAKALMIKYEQRQGLMARWDHWRRCAPVLNLLGLHATDGYTPVRRGPARDFLAWGYFQSEAYFADFAPTIRAEIRAKQAPAGVWAEKIRAAACPVALLHRRGDYCRPENEILQVCSPAYYARAAAAAAYPEATLFFVSDIDWAKEHLDTAGLPAVWMPRGDAVGDLNLMALCRGFILSNSTYSWWAQYLAGGRTVWAPDRWFAHTKQTALYQPGWHLIETR	WO 2015/175801
Firmicutes bacterium CAG:24	WP_021916223.1	547127527	protein [Firmicutes bacterium CAG:24]	23.4		MIIVEVMGGLGNMQQYALYRKLESIGKDARLDVSWFLDKERQTKVLASRKLELSWFENLPAKYCTQEEKQAILGKNLIGLKKLLGGSNRHFTESDMYHPEIFLEDAYLSGFWACEAYADILPMLRSQHFFPDPEKGEGWDLAAAAKNKETMERMKQETSIVSIHRRGDYLDAKNAEMFGGICTDAYEAAISYIKEQTPDAHFFVFSDDSYVKNAYPGKEFTVDWNTGKNSLFDMLMSSCNHICANSTFSFWGARLNPSDPKVMIRPSKHKNSQNIIVPEEMKRLWDGWVLDIGKGRII	225
Prevotella sp. CAG:474	WP_022310139.1	547906803	glycosyl transferase family 11 [Prevotella sp. CAG:474]	23.39		MIITKNGGLGNQLFEYACARNILQLKYNDVLYLDIEGFKRSPRHSYLEKFKLSSDVRMLPEKDSKSLILLQAISKLNRLNLAFLKGLPFGTYIWKSSNYRPLKIKNTRGKLYLYGYWQSYEYFENEAIKQELNVKTEIEPCESELLKEINKPHSICVHVRRGDYVSCGFLHCEAYYNRGINHIFDKHPDSNVVVSDDIKWVKANMNFDPVAYVEVDVPDYETLRMLMYMCKHFVMSNSSFSWWASYSLSDNKEKIVAPSYWLPANKDNKSMYLDNWTIL	226
Roseburia intestinalis; Roseburia intestinalis L1-82	WP_006855899.1	493910390	glycosyl transferase family 11 [Roseburia intestinalis]	23.38		intestinalis]MRGNRGMIAVKIGDGMGNQLFNYACGYAQARRDGDLSLVDISECDNSTLRFELDKFHLKYDKKESPPNRLGQKIYKNLRRALKYHVIKEREVYHNRDHYDNDIDPRVYKKGLRNKLYLYGWQHLAYFEDYLDEITAMMTPAYEQSETVKKLQEEFKKTPTCVHVRRGDIMGPAGAYFKHAMIERMIEQEKPGVRYVFTNDMERAEALAPVLESQKKDAVGQAENRLEFVSEMGESDVDEFFLMAACQNQLSNSTFSTWAAAYLNQNPDKTVIMPDDLLSERMRQKNWIIK	227
Bacteroides ovatus; Bacteroides ovatus ATCC 8483	WP_004296622.1	490424433	protein [Bacteroides ovatus]	23.29		MKIVLFTPLGLGNQMFQYLYLRLDNPQNQNIYGYNNRNLNKHNGLEVDKVFDIQLPPTHVISDASAFFIRALGGLGKYHIGKDLSPWKVYFDGYWQNKKEYFQNNVVDKMRREGLNKKNDLILSLRNTNSVSVHVRRGDYCDSCRKDLFLQSCCTPQYYESAISVMKEKQKPVFFVSDIPWVKVNLNIPNAYYIDWNKKNSYLDMYLMSLCTASIIANSTFSFWGAMLGNKKELVIKPKKWIGDEIPEIFPPSWLSL	228
Butyrivibrio sp. AE3009	WP_022779599.1	551035785	glycosyl transferase [Butyrivibrio sp. AE3009]	23.25		MLIIQIAGGLGNMQQYALYRKLYHPDGVRLDLSWFDSEVQKNMLAKREFELAFKGLPYECKPEERAAFLDRNAAQKLSGKVLKLLGLRDNAIPNVFEESRMFHPEIFELDNKYIIGYACQKYDDIMGDLNCLNLFEPHLDPELEKKNLELISKMEKENSVSVHIRRGDYLDPENFKILGNIAIDYEYESAMKYFEDRYEKVHFYFTSDHEYAREHFADESKYITVDWNTGKDSLQDVRLMNHCLGNICANSTFSFWGARLNQRQDKVMIRTYKMRNNQPVDPDTMHDYWKGWILIDETGREV	PCT/US2015/030823

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MTKNEKKLVKFQGGGLGNQLYEAFCWLRQQSYDYEVLDLSYYKIRSAHGELGIWNIFFPINIEVASNWDI IKYSDQIPIMYGGKGADRLNSVRTNVNDRFSSKRKHSYYTEISNTDVSSEVINALNNGIRYFDGYWQNIIDYFKG NIEDLRNKLKFKSEKCDKITDMLRDNVSLHVRRGDYVGEVEKEVGLSYKKAVEYVLDVRVDQAKFFISD DKYAAETAFEWIDNKTWAGYDNELAHVDMMLLSRMKNNIANSTFSLWAAYLNDSMNPLIVYVDPVESLD KKTFSWNGIK	23.23	23.23	23.21	23.2	
MDSQFLKHILSGGFGNQLFOYFFGEYLKEKYNCSISFFSEPALDINQLQIRFFPALRISHNTELRPYHYSFTQ QLAYRCMRKLLLPFLNRKVKIENGSNYQNSFNNTYCFDGYWQSYRYSFAFTPSLQFEDQLINDISADYIN AIEQSEAVFLHRRGDYLNKENQKVFAECPLNYFENANRIKEDIKNVHFFFSNDIQWVSHLKLNDNEVTF IQNEGSCDLKDFYLMTRCKHAIISNTFSWWAAYLINNSDKKVIAPKHWNIDISMNNATKDLIPTWIRL	302669752	517173838	495838392	551041720	
glycosyl transferase 11 [Butyrivibrio proteoclasticus B316]	WP_01836 2656.1	WP_00856 2971.1	WP_02089 5733.1	WP_02278 5342.1	
Butyrivibrio proteoclasticus; Butyr ivibrio proteoclasticus B316	Prevotella nancelensis	Ruegeria sp. R11	Winogradskyella psychrotolerans RS- 3; Winogradskyella psychrotolerans	Lachnospiraceae bacterium NK4A179	Prevotella sp. oral taxon 317 str. F0108; Prevotella sp. oral taxon 317
protein [Prevotella nancelensis]	alpha-1,2- fucosyltransfera se [Ruegeria sp. R11]	alpha-1,2- fucosyltransfera se [Winogradskyell a psychrotolerans]	protein [Lachnospiracea e bacterium NK4A179]	alpha-1,2- fucosyltransfera se [Prevotella sp. oral taxon 317]	
MIITRLHGLGNQMFQYAAAGRALADRAGVPLALDSRGAILRGEGLTRVFDLELADPVHLPLKQTNPRLRYA IWRGIGQKVGAQPYFRRERGLGYNPAFEDWGDNSYLHGYWQSQYFQNSAERIRSDFTFPFSNQNAE MAARIAESTAISLHVRRGDYLTFAAHVLCDDQAYYDAALAKVLDGLQGDPIVYVFSDDPQWAKDNLSLPCEK VVVDNFGPETDFEDMRLMSLCQHNIIGNSSFSWWAAWLNQITPGRRVAGPAKWFGDPKLSNPDIFFHDW LRISV	496522549	WP_00923 0832.1			
MGNQLYAYATAKAMAVALNKKLVDPRLKEAPQRHYDLGLFNIQDEDFGSPFQWLVRWVASVRLGKFF KTIMPAWSYQMIRKKEGFDESLLQQKSRNIVIEGYWQSKFYESIIRPTLLKELSKDKPNAINQKYLDEIESV NAVAVHIRRGDYVANPANAVALHGLCDMDIYKKAIAIKDKVENPYFFITDDPDWAEDNFKISEHQIKIHNI GKODHEDFRLLTNCKYFIANSFSWWGAWLSDYKNKIVISPNKWFNVDVPIITERIPESWIRV					
MITVRIDGGFGNQMFOYAFFLHLKKTITDNKISVDLNCYNPHGSGDIFTRFLAPEQAAPSEIKRFRHNSIYHL LRPLDSAGITNPPYREEDIDDLNSVLNKKRVYLRGYWQDKRYPFSVKDQLIDCFDLGKMDMTGASAEENNVI LEQASEESRSVGVHLRGGDYIGDPVYSGICTPEYEAFAKHVSEKIKDPVFHIFTNDISMIEKCGLSGKYDLKIT DINDEAHGWADLKLMSACRHHIISNSSFSSWWAAFLGEATTEASADVINVIPEYMRQGVSAETLRCPCWTT VTSDDRVPVS					
MKIVCIKGGGLGNQLFEYCRYRSLHRHDNRGVWLYHYDRRRRTKQHGCVWLDKAFHITLNEPLRVKLLVMVLK TLRRLHLKRLYREEDPRAVLIDDYSQHKQYITNAAEILNFRPEQLDYAEIQTTPFAVSVHVRRGDYLLANK SNFGVCSVHYLYLSAAVAVRERHPESRFFVSDMEWAKENLNLPCNVFVEHAQAQPDHADLYLMSLCKGH IIANSTFSFWGAYLSKGSSAIAIYPKQWFAEPTWNVPDIFPAHWMAL					

WO 2015/175801	241							MRIGILYICTGYTVFWNHHFTSCQEQHFLREHEKHYIIFTDGEIAHLNCRVHRIEQQHGLGWPDSTLKRPHM FERIADTLRQNSDFVFFNANMFLRDVGKEFLPTREQALVFHRHPGLFRPAWLLPYERPESTAYIPYSGG SIYVCGGVNGGYTQPYLDFVAMLRNIDIDVERGIARWHDESHINRFVIGRHYKIGHPGYVPDRRLNLPFR IIRVIDKASVGGHTFLRGQTPPEAPEEQSKTVAKLRSLQKRPCMPRAAQDEPILARMGGLGNQMFIYAA ARVLAERQGAQLHLDTGKLSGDSIRQYDLPAFSDAPLWHIPCGCDRIVQAWFALRHVAAGCGMPKPTMQ VLRSGFHLDRFFSIRHSAYLIGYWQSPHYWRGHEDRVRSFDTLRFERPHLRLEALAAVSQPNITISVHLRRG DFRAPKNSDKHLIDGSGYERARKLLEMTPTQSHFYIFSDEPEEAQRLFAHWENTSFQPRRSQDEEDLLMSRC SASIANSSFSWWGAWLGRPKQHVIAPRMWFTRDVLMHTYTLDLFPEKWILL
					23			
								MILIHVMGGLGNQLYQYALYEKMKSLGKKVKLDTYAYNDAAGEDKEWRSLELDRFPAIEYDKATSEDRTKLL DNSGLLTAKIRRKLLGRKDKTIRESEYMP EIFHMDDDVYLYGFWNCERYEDIPLLDQKLQFISNNPRNQ CIEQMCKENAVSIHIRTDTLTVADGARYMGICTEDYKGMAYIEERVSNPVYIFSDDDVEYAKQHYHQD NMHVVDWNSKADSIYDMQLMSCKKHNICANSTFSMWAARLNQNKEMIRPLHHDNYETTTATQVKQN WKNWILLDQNGQVCE
					22.98			
								MTMNIIRMSGGLSQMFQYALYLKLSMGKEVKFDDINEYRGEKARPIMLAVFGIEYPRATWDEITSFTDG SMDLLKRLRRKIFGRKAIEYEEQGFDYDPNVLFNDSMYLRGNFQSEKYFQDIKEEVRLKYRSTLEDMLRPLRY KATKACLDGIESSESVGLHMYRSDSRVDGELYDGICTGNYYKGAVRFIQDKVPDAKFYFSNEPKWVRGWV DLIQSQIQEGMSPSQVKEKMEKRFVMEANTEYTGyLDMMLMSCKKHNIISNSSFSSWWSAWMNDHPEKV VVAPDRWSSDKEGNEITYTGTMLVNEKGRVNYTHENSTVK
					22.96			
								MILSYITGRGNQLFEYAYARSLLKRGKNEELINFSLVRAAGKEIGFDDNLRYFNVSYTELDKDIWLSKGD LQLFIYILFKLDQKLFRIIIRKKEKWFSEFRFGIIFQDYLDNISNLIIPRTKNVFCYGYENPKYFDDIRSILLKEFTPRI PPLKNNDQLYSVIESTNSVCISIRRGDFLCKDFKDRFLVCDKEYLEAMEEAKKRISNSTIFFSDDIEWVRENIH SDVPCYFESGKDPVWEKLRMLMYSCKHFISNSTFSWWAQYLSRNEEKVVIAPDRWSNVNPGKSFLLSNSFIKI PIGILP
					22.96			
								MIYVEINGRLGNMFEIAAASLTDEVTLWCKGDWQLNCIKMYSDTLFKNYPVKSLPNNIRIYEEPEFTFPI PYKENQDILLIKGYFYQYKYLDRKVLKLYPCMPMPVKLDIEKRFGLDLSQYTVSINVRRGDYVNLPHRHPFVGK KFLERAMLWFGDKVHYIISDDIEWCKAHFKQFDNVHYLTNSYPLLDLYIQTACHHNIISNSSFSSWWGAYLN NHPQKIVIAHRWFGMSTNINTQDILLPEWWMIEQCVCYEPKVFELKALPLHAKYLLKRVLK
					22.95			

Prevotella sp. oral taxon 299 str. F0039;Prevotella sp. oral taxon 299	YP_008444 280.1	532354444	protein HMPREF0669_0 0176 [Prevotella sp. oral taxon 299 str. F0039]	22.9	MDSQLLKHILSGGNGQLFQYFFGEYLKEKYNCSFFSEPALDINQLQJHREFFPTLRISHNTELRHFHYAFTQ QLAYRCMRKLLLPFLNRKVKIENGSNYNQNSFNDCYFDGYWQSYRVSFTPSIQFEDQLINDISADYIN AIEQSEAVFLHRRGDYLNKENQKVFAECPLNYFENAVNKIKEGNTYHFFFSNDIEWVKCHLKLNNNEVTF IQNEGSSCDLKDFYLMTRCKHAIISNSTFSWAAAYLINNDKKVIAPRWYNDLSMNNATKDLIPPTWIRL	WO 2015/175801 247
Paraprevotella xylaniphila;Paraprevo tella xylaniphila YIT 11841	WP_00862 8783.1	495904204	alpha-1,2- fucosyltransferase [Paraprevotella xylaniphila]	22.87	MKIVCLKGLGNQMFECRFRDLMDSGNGKVLYFYDRRLKQHDGLRLSDCFEELPSCPWGIRLVVWVGL KICRAIGVLKRLYDDEKDPADVLIDDYSQHRFRIPNARRYFSFRQFLAELQSGVQVMIRAVDYPVSVHVRRGDY LHPSNSSFVLCGVDFRQAIAYVRKRPDARFFFSDDMEWVRENLMEDAVVVEHTELMPPDYMDLYLM TLCRGHIISNSTFSFWGAYLAVDGNMGKMYPRRWFRDPTWTITPIFSEEWVGL	247
Dethiosulfovibrio peptidovorans;Dethi osulfovibrio peptidovorans DSM 11002	WP_00565 8864.1	491897177	glycosyl transferase family 11 [Dethiosulfovibri o peptidovorans]	22.84	MFQYAFGRALALDLGLDLISNFGSDSRPFLSLGIYSLTKNIPFGCYLSTSTRLKVKMTKKLRRWGVWGMID KNMPGVLEVPFPVVLVSLDEVLSKLSHLFVDGYWQSEKYSRYSVDVIRSDFRVIESSAFLAWKKRMLSEPG GSIHVHRRGDYVTDSSANRVHGVLPYLYLRAKELINTISDGLVFYVFTDDPVWARNNLCIGDKTIYVSGEDL KDYELALMSCCDHHVAVANSFSFWGAWLGQDTSVTIAPGRWFRKMDSSFSFVIPDNWIKIWT	248
Lachnospiraceae bacterium 10-1	WP_01622 9292.1	510896192	protein [Lachnospiraceae bacterium 10- 1]	22.83	MIIQVMGGNGQLQYALYRKFRVMGKEARLDSWFLDKEKRGVLAERELELDYFDRLIYETCTPEEQLI GSEGVAAGLKRKFLPGRIRWFHESKIYHPPELLQMENMYLSGYFACEKYADILYDLREKIQFPVNDHPKNIM AQEMQERESVHLRRGDYLDKNTAMFGNICTDAYYCKAIEYMKTLCSKPHFYIFSDDIPIYVRQRTGEEYT VVDINHGRDSFDMWLMMSRCRHNICANSTFSFWGARLNSNDNKIMIRPTIHKNSQVFKVEEMEQWLWPG WKFLSPDGGIK	249
Treponema maltophilum;Trepon ema maltophilum ATCC 51939	WP_01652 5279.1	513872223	protein [Treponema maltophilum]	22.82	MFCAAFVEALKHAGQKVFVDTSLYNKGTVRSIGDFCHNGLETEHLFGIKFDEADKADVHRLSTSAEGLNRI RKYFTKKTHYIDTVFRYTPVLSKSDRYLEGFWQTEKYFLPIESDITLFRFRQPLSEKSAVQSALQAQEPAS LSASIHVRRGDFLHTKTLNVCTETYNNAIEYAAKAYAVAFYVFSDDIQWCREHLNFFGARSVIDWNIGAD SWQDMVLMSCMCRNIANSFSSWWAAWLNAASDKIVLAPAIWNRRLQLEYADRYGYDYSDVIPETWIRIP I	250
Bacteroides massiliensis;Bacteroi des massiliensis dnLKV3	WP_01627 6676.1	511022363	protein [Bacteroides massiliensis]	22.79	MKLVSFTAGLGNQLFQYCFYRYLLNKEPNEKIYGYNNKKWLKHGGIIIEHFFDVKLPRSTRWINLYGQYLRIY KCFSCGVSKDDDDFEMNRTMFVGYWQDQCFSGINISYKKNLVISEKNTWLLGEILKCNVAIHFRRGDYMLP QFKKIFGEVCTVKYKLSIRKVEEKISEPVFFVSDDIDWVKQNFTEFNKYVYVDWNKGNQNSFWDMYLMSSQC SANIIANSTFSFWGAYLNNKNNPFVIYQKQWVVRTNLKQPNIFPKTWMAAL	PCT/US2015/030823

WO 2015/175801		253		254		255		PCT/US2015/030823	
Enterococcus faecium;Enterococcus faecium DO;Enterococcus faecium EnGen0035	YP_006376560.1	389869137	family 11 glycosyltransferase [Enterococcus faecium DO]	22.71	MIVLTGGGLGNQMFQGYARYIQIHREKFIYINDSEVKEADRFNSLGNLNTVNIKVLPRISKPLNETERLV RKIMVRLFGVAGFNESAIFOSLNFYIYHPVSVYKFVESLKTGPKIEGGFQSWKYLETCPEIKQELRVKYPE MGENRLNLISQSESVCHIRRGDYLSPKYKHLNVCDYQYVFESMNYISKLNNPTFFIESNTSDDLWDWIEN YSLPGKIVYKNDNDPDYEELRLMYSCKHFIISNTFSWAWAQLSNNSGVIAPEIWNRLNHDGIADLYMPNWN ITMKVNR	MIGFALGRMGRLANQMFQYASLKGIARNTGVDFCPVHEEAVNDGIGNMLRTEIFDSFDLQVNVGLLNK GHAPVWQERFFHDEELFRMCPDHVDIRGYFQTEKYKHIEDEIREDFTEKDEILNPCKEMIAGVDNPLALHV RRTDYVTNSANHPPTCTLEYEAALKHFDDDRNVIVFSDPAWCKEQELFSDDRFMISENEDNRIDLCLMSLC DDFIANSTYSWWGAWLSANKDKKVIAPVQWFGTGYTKDHDTSDLIPDGWTRIATA	MDIHVLSYGLGNLSQVAFINRRQLMQRAYAFYAFKQKHNGYELDRIFGLKEGLPWYIQLFVRVVRFLGISRR FYSKRTADFVLSLFRIKVIDEAYNYEFDPSLLKPWFGIRILYGGWHDSRYFHPSEAAVRTAFSPPLDDVNDAIL QQIDAVYGVSIHVRRGDYLKGINSLFGGIATLEYRNGAIGWAITYCKHRSLEIKFYVFSDDIDWCKQNLGLR DAVYVSGNSKTDSDWKDILLMSHCRANIIANSTFSWWAAWLNQQPNKVVICPTKFINTDSPNQTIYPAAWH QIEG	MIIVRFHGGGLGNQMFYAFYRYMTNKYGADNVIGDMTWFDNRNYSEHQGYELKKVFDIDIPADYKTLAKIH EYPRYHRFAGRLYLSRMVYAKYKNKHLKPTGEYIMDFGSPQYIHNDADFKLDTNKDYYIEGVFCSDAYIKYYE NQIKKDLTKPNYSQHTKMDMLPKIEETNSVAIHVRRGDYVGNVDFIVTPDYRQAVNYIRERVENPVPFVFS DMDYIKANFDFLGDFVPVHNCCKGDSFQDMYLSRCRHMIIANSFSYFGALLGEKDSITVIAPKKYKADEDLA LARENWVLL	
Bacteroides;Bacteroides sp. 2_1_22;Bacteroides sp. 2_2_4;Bacteroides sp. D1;Bacteroides xylanisolvens SD CC 2a;Bacteroides xylanisolvens SD CC 1b;Bacteroides ovatus CAG:22	WP_004313284.1	490442319	glycosyl transferase family 11 [Bacteroides]	22.67	MDVVIFNGLGNQMSQYAYYLAKKKVNPNTKVIFDIMSKHNHYGYDLERAFGIEVKNKTLIKVLQIYVLSRK FRLKSVGVRTIYEPLNYDYTPLLMQKGPWGINVYVGGWHSEKNFMNVPDEVKKAFFREQPNEDRFNE WLQVIRGDNSSSVHIRRGDYMNIPTGYQLNGVATLDYYHEADYIRQYVDTPHFVYFSNDLDWCKEQF GVENFFIECNQGVNSWRDMYLMSECHYHINANSTFSWWGAWLCKFEDSITVCPERFIRNVVTKDFYPER WHKIKSC	MIGFALGRMGRLANQMFQYASLKGIARNTGVDFCPVHEEAVNDGIGNMLRTEIFDSFDLQVNVGLLNK GHAPVWQERFFHDEELFRMCPDHVDIRGYFQTEKYKHIEDEIREDFTEKDEILNPCKEMIAGVDNPLALHV RRTDYVTNSANHPPTCTLEYEAALKHFDDDRNVIVFSDPAWCKEQELFSDDRFMISENEDNRIDLCLMSLC DDFIANSTYSWWGAWLSANKDKKVIAPVQWFGTGYTKDHDTSDLIPDGWTRIATA	MDIHVLSYGLGNLSQVAFINRRQLMQRAYAFYAFKQKHNGYELDRIFGLKEGLPWYIQLFVRVVRFLGISRR FYSKRTADFVLSLFRIKVIDEAYNYEFDPSLLKPWFGIRILYGGWHDSRYFHPSEAAVRTAFSPPLDDVNDAIL QQIDAVYGVSIHVRRGDYLKGINSLFGGIATLEYRNGAIGWAITYCKHRSLEIKFYVFSDDIDWCKQNLGLR DAVYVSGNSKTDSDWKDILLMSHCRANIIANSTFSWWAAWLNQQPNKVVICPTKFINTDSPNQTIYPAAWH QIEG	MIIVRFHGGGLGNQMFYAFYRYMTNKYGADNVIGDMTWFDNRNYSEHQGYELKKVFDIDIPADYKTLAKIH EYPRYHRFAGRLYLSRMVYAKYKNKHLKPTGEYIMDFGSPQYIHNDADFKLDTNKDYYIEGVFCSDAYIKYYE NQIKKDLTKPNYSQHTKMDMLPKIEETNSVAIHVRRGDYVGNVDFIVTPDYRQAVNYIRERVENPVPFVFS DMDYIKANFDFLGDFVPVHNCCKGDSFQDMYLSRCRHMIIANSFSYFGALLGEKDSITVIAPKKYKADEDLA LARENWVLL	
Synechococcus phage S-SM2	YP_004322362.1	326781960	glycosyltransferase family 11 [Synecococcus phage S-SM2]	22.6	MIGFALGRMGRLANQMFQYASLKGIARNTGVDFCPVHEEAVNDGIGNMLRTEIFDSFDLQVNVGLLNK GHAPVWQERFFHDEELFRMCPDHVDIRGYFQTEKYKHIEDEIREDFTEKDEILNPCKEMIAGVDNPLALHV RRTDYVTNSANHPPTCTLEYEAALKHFDDDRNVIVFSDPAWCKEQELFSDDRFMISENEDNRIDLCLMSLC DDFIANSTYSWWGAWLSANKDKKVIAPVQWFGTGYTKDHDTSDLIPDGWTRIATA	MDIHVLSYGLGNLSQVAFINRRQLMQRAYAFYAFKQKHNGYELDRIFGLKEGLPWYIQLFVRVVRFLGISRR FYSKRTADFVLSLFRIKVIDEAYNYEFDPSLLKPWFGIRILYGGWHDSRYFHPSEAAVRTAFSPPLDDVNDAIL QQIDAVYGVSIHVRRGDYLKGINSLFGGIATLEYRNGAIGWAITYCKHRSLEIKFYVFSDDIDWCKQNLGLR DAVYVSGNSKTDSDWKDILLMSHCRANIIANSTFSWWAAWLNQQPNKVVICPTKFINTDSPNQTIYPAAWH QIEG	MIIVRFHGGGLGNQMFYAFYRYMTNKYGADNVIGDMTWFDNRNYSEHQGYELKKVFDIDIPADYKTLAKIH EYPRYHRFAGRLYLSRMVYAKYKNKHLKPTGEYIMDFGSPQYIHNDADFKLDTNKDYYIEGVFCSDAYIKYYE NQIKKDLTKPNYSQHTKMDMLPKIEETNSVAIHVRRGDYVGNVDFIVTPDYRQAVNYIRERVENPVPFVFS DMDYIKANFDFLGDFVPVHNCCKGDSFQDMYLSRCRHMIIANSFSYFGALLGEKDSITVIAPKKYKADEDLA LARENWVLL		
Geobacter metallireducens;Geobacter metallireducens GS-15;Geobacter metallireducens RCH3	YP_006720295.1	404496189	glycosyltransferase [Geobacter metallireducens GS-15]	22.58	MIGFALGRMGRLANQMFQYASLKGIARNTGVDFCPVHEEAVNDGIGNMLRTEIFDSFDLQVNVGLLNK GHAPVWQERFFHDEELFRMCPDHVDIRGYFQTEKYKHIEDEIREDFTEKDEILNPCKEMIAGVDNPLALHV RRTDYVTNSANHPPTCTLEYEAALKHFDDDRNVIVFSDPAWCKEQELFSDDRFMISENEDNRIDLCLMSLC DDFIANSTYSWWGAWLSANKDKKVIAPVQWFGTGYTKDHDTSDLIPDGWTRIATA	MDIHVLSYGLGNLSQVAFINRRQLMQRAYAFYAFKQKHNGYELDRIFGLKEGLPWYIQLFVRVVRFLGISRR FYSKRTADFVLSLFRIKVIDEAYNYEFDPSLLKPWFGIRILYGGWHDSRYFHPSEAAVRTAFSPPLDDVNDAIL QQIDAVYGVSIHVRRGDYLKGINSLFGGIATLEYRNGAIGWAITYCKHRSLEIKFYVFSDDIDWCKQNLGLR DAVYVSGNSKTDSDWKDILLMSHCRANIIANSTFSWWAAWLNQQPNKVVICPTKFINTDSPNQTIYPAAWH QIEG	MIIVRFHGGGLGNQMFYAFYRYMTNKYGADNVIGDMTWFDNRNYSEHQGYELKKVFDIDIPADYKTLAKIH EYPRYHRFAGRLYLSRMVYAKYKNKHLKPTGEYIMDFGSPQYIHNDADFKLDTNKDYYIEGVFCSDAYIKYYE NQIKKDLTKPNYSQHTKMDMLPKIEETNSVAIHVRRGDYVGNVDFIVTPDYRQAVNYIRERVENPVPFVFS DMDYIKANFDFLGDFVPVHNCCKGDSFQDMYLSRCRHMIIANSFSYFGALLGEKDSITVIAPKKYKADEDLA LARENWVLL		
Lachnospiraceae bacterium NK4A136	WP_022780989.1	551037245	protein [Lachnospiraceae bacterium NK4A136]	22.58	MIGFALGRMGRLANQMFQYASLKGIARNTGVDFCPVHEEAVNDGIGNMLRTEIFDSFDLQVNVGLLNK GHAPVWQERFFHDEELFRMCPDHVDIRGYFQTEKYKHIEDEIREDFTEKDEILNPCKEMIAGVDNPLALHV RRTDYVTNSANHPPTCTLEYEAALKHFDDDRNVIVFSDPAWCKEQELFSDDRFMISENEDNRIDLCLMSLC DDFIANSTYSWWGAWLSANKDKKVIAPVQWFGTGYTKDHDTSDLIPDGWTRIATA	MDIHVLSYGLGNLSQVAFINRRQLMQRAYAFYAFKQKHNGYELDRIFGLKEGLPWYIQLFVRVVRFLGISRR FYSKRTADFVLSLFRIKVIDEAYNYEFDPSLLKPWFGIRILYGGWHDSRYFHPSEAAVRTAFSPPLDDVNDAIL QQIDAVYGVSIHVRRGDYLKGINSLFGGIATLEYRNGAIGWAITYCKHRSLEIKFYVFSDDIDWCKQNLGLR DAVYVSGNSKTDSDWKDILLMSHCRANIIANSTFSWWAAWLNQQPNKVVICPTKFINTDSPNQTIYPAAWH QIEG	MIIVRFHGGGLGNQMFYAFYRYMTNKYGADNVIGDMTWFDNRNYSEHQGYELKKVFDIDIPADYKTLAKIH EYPRYHRFAGRLYLSRMVYAKYKNKHLKPTGEYIMDFGSPQYIHNDADFKLDTNKDYYIEGVFCSDAYIKYYE NQIKKDLTKPNYSQHTKMDMLPKIEETNSVAIHVRRGDYVGNVDFIVTPDYRQAVNYIRERVENPVPFVFS DMDYIKANFDFLGDFVPVHNCCKGDSFQDMYLSRCRHMIIANSFSYFGALLGEKDSITVIAPKKYKADEDLA LARENWVLL		

Bacteroides coprophilus; Bacteroides coprophilus DSM 18228 = JCM 13818	WP_00814 4634.1	495419937	protein [Bacteroides coprophilus]	22.56	MGFVNMACGLNRMFQSYNYLFLKKQGYKVTVDYRSAKLAHEKVAWNSIFPYAEIKQASRLKVLWGGG SDLCVKRRRYFSPSTNVRTTTGAFDASLPANTARNEYIIGVFLNASIVEAVDDEIKKCFTELPTDEMNRLKK EIEECESVAIHVRKGDYQSRWYQNTCSMEYRKAILQMKELQHSKFYFTDNVDWVKENFQEIYTLVE GNPADGYGSHFDMQLMSLCKHNIISNSTYSWWSAFLNRNPEKVVVIAPEIWFNPDSDEFRSDRALCKGWI VL	WO 2015/175801
Bacteroidetes; Capnocytophaga sp. oral taxon 329 str. F0087; Paraprevotella clara YIT 11840	WP_00861 9736.1	495895157	alpha-1,2- fucosyltransferase [Bacteroidetes]	22.53	MKIVCLKGGNGMFEYCRFRDLMESGHDEVLYFDHRRRLKQHNGIRLSDCFEELPSCPWGKLVVWGLK ICRAVGVLKRLYDDEKPEAVLIDDYSQHRFPNARRYFFFRQFLAELQSGFVQVMIRAVDYPVSVHVRRGDYL HPSNSSGLCGDYFQQAIAVVRKRPDARFFFSDDMEWVRNLWMEDAVVVEHTELLPDYVDLYLMTL CRGHIISNSTFSFWGAYLAVDNGMGKIPRRWRFRDPTWTSPFSEEWVGL	258
Butyrivibrio sp. NC2007	WP_02277 0361.1	551026242	glycosyl transferase [Butyrivibrio sp. NC2007]	22.47	MLIQJAGGLGNMQQYAVYTKLREMGDKVLDLSWFDQVQKNMLAPREFELPIFGGTDYEECSAYERD ALLKQGAFAAIAAGKVLKGLRDEANPKVFESEKEMYHPEVELEDKYGKGFACQKYGDIMDKLOEKFIPE HSDPDLHARNMALVERMEREPSVSHIRRGDYLDPNSVEILGNIAETEQQYQGAMDYFTVKEPDTHFYFTSD HEYAREKFSDESKYTIWNNNGKNSVDLMLMSHCKGNICANSTFSFWGARLNKRPKTKVIRTYKMRNN QVNPQIMHDYWKGLWLMDEKGSII	259
Paraprevotella xylaniphila; Paraprevotella xylaniphila YIT 11841	WP_00862 8536.1	495903957	glycosyl transferase [Paraprevotella xylaniphila]	22.45	MKILVFTGGLGNQMFAFYLYLKLFPQERFYGLYKKLSEHYGLEIDKWKVSLPRQPWWVLPVTGLFYL YKQCVNSKWLDLNLQEIKNPRAIVFFPFKTKKYIPDDNIWLEWKVDESGLSEKNRLLSEIRSSDCCFVHVR RGDYLSPTEKSLFEGCCTLSYQRAKSMKEISPFVKVCFSDDIQWVKQNLGNRAVFDWNSGTDSPDL MYLMSQCRYGIMANSTFSYWGARLGRKKRIYYPQKWWNHGTLGLDIFPNTWVKI	260
Blautia hydrogenotrophica DSM 10507; Blautia; Blautia hydrogenotrophica CAG:147	WP_00594 4761.1	492742598	protein [Blautia]	22.44	MEIHVYLTGRLGNQLFQYAFARHLQKEYGGKIICNIYELEHREKAAWVPGKFNEMSNYKLNDSILEDIKLP WFADFSNPIIRIVKVIPIRYFNLMASKGYLLWQKNYSINIPAIRNNEIIVNGWWQDVRFFHDVEAELSNEIVP TTKPISENEYLYNIAERENSVCSIRGGNYLVPKVKKKLFVCDKEYFYNAIELIKSVRNAIFVFSDDLEWVKSYI KLEEKPECKFYESGKDTVEEKLMMTKCKHFIISNSFSWAAQYLAKNENKIVIAPDWFTNGDKNGLYI DDWILIPTQTKDM	261
Geobacter lovleyi; Geobacter lovleyi SZ	YP_001952 981.1	189425804	glycoside hydrolase family protein [Geobacter lovleyi SZ]	22.44	MITVLLNGGLGNQLFQYAAAGRALAEKHDVELLLDSRLQHPKPGDTPRCFELAPFNKASILAEEGRQPLGYSY QACMHRLLKASIPILWGSIIKEQGCQGFDPILFRAPSSCILDGFWQSECVFKQITSLQQLSLKAPSPALRKAS SVLSDATVAVHVRRGDYVTNPAAASHFGICSDYQAAVANILTSYDPSQLVFSDDPAWCOEHLDLGQPF RLAADFGLNGSAEELVLSRCAHQIANSFSWWGAWLNPSPHKLVVVAPCRWFTDPAITNDLLPETVVRPLP	2

Providencia alcalifaciens	AFH02807. 1	383289327	glycosyltransferase [Providencia alcalifaciens]	22.26	MKINGKSSMIKQKIIISHLIGLGNLQFYATSYALAKENNAKIVIDDRFLFKYKLGGRYLDKLNIIKEKISS IDKLLFPLILCKLSQENFIKSTKFFILEKKTSSFKYLTFSDEKHTKMLGYWQNAIYFQYFSELKEMFVPLDIS QEQLDSIQIHAQSQVALHVRRGDYISNKNALAMHIGCSIDYYKNSIQHINAKLEKPEFYIFSNDKILWCEENLT PLFDGNGFHIVENNSQEIIDLWLSQCQHHIIANSTFSWWGAWLANSDSQIVITPDWFWNKEDIPSPVLSHWL KLKK	WO 2015/175801 270
Salmonella enterica	AFW04804. 1	411146173	glycosyltransferase [Salmonella enterica]	22.26	MFSCLSGGLGNQMFQYSAAYILKKNICHACLIIIDDSYFYCQPKQDTPRIFEINQFNIVFDRVTTDEEKRAISKL RKFKKIPLPLFKSNVITEFLFGKSLTDEDFYKVLKKNQFTVKMNACLSFYQDSSLINKYRDLLPLFTINDELLO VQCQLDSYGFCIEHTNTTSLHRRGDYVYTNPHAAKFGHTLSMNNYSQAMNYYVHKLKQKLIIFSDDDVQWA AEKFGGRSDCYVNNVNCQFSAIDMYLMSLNNNIIANSTYSWWGAWLNKSEELVIAPRKWFAEDKESLL AVNDWISI	270
Sulfurospirillum deleyianum; Sulfurosp irillum deleyianum DSM 6946	YP_003304 829.1	268680398	protein Sdel_1779 [Sulfurospirillum deleyianum DSM 6946]	22.18	MIHIMGGGLASQLHKYSVGRALSCLKYNTLKLDFWFDNISGSDTIREYHLDKYNVVAKIATEQEIQKPKNKY LLKINNLFQKFTNWKINYNRYNCNESFISLENFLLPDNIYVEGEWSDRYFSHIKEILOKELTLKSEYMDSTNHF LAKQSSDFAHDDNASKLHCTCSLEYKALQYISKNLKMKLIIFSDDLWLPNFNFLDNVEFEVEFGQDY EEFLMTLSKHNIANSFGSLFFAWLINHNKIIISLSEWVFEELNKYIIDNIKDKNILENLE	271
Pseudovibrio sp. FO- BEG1	YP_005080 114.1	374329930	alpha-1,2- fucosyltransferase [Pseudovibrio sp. FO-BEG1]	22.15	MSVASQVRISGAARRRKLKPTLIVIRGGIGNQLFQYALGRKIALETGMKLFDRSEYDQYFNRSYCLNLFKT QGLSATESEMSAVLWPAQSFQGTVKLCRKFPYQRYIREDELLOQDSETPVLKQSAYLDGYWQWTWEIPFSI MEQLRDEITLKKPMVLERLKLQRIKSGPSAALHVRGYDYSQAHNLQNFGLCSAGYYKGMDFLTERVPGLT FYFSDSPERAREVVPQENNVYFSDPMQDGDHEDLMVMSSCDHIVTANSTFSWWAAFLNGNEDKHVIA PLKWFKNPNDLDSLIVPPHWQRL	272
Prevotella sp. oral taxon 472 str. F0295; Prevotella sp. oral taxon 472	WP_00923 6633.1	496529942	alpha-1,2- fucosyltransferase [Prevotella sp. oral taxon 472]	22.11	MKIVCKGGLGNQLFEYCRYHGLLRQHNNHGVYLYHYDRRTKQ.HGGVWVLDKAFILTLPTPEWVRVKLMVM ALKMLRKLHLKRLYREDDPRAVLIDDYSQHKQFITNAAEILNFRPFAQLDYVDDEITSEPFVSVHVRRGDYLL PANKANFGVCSVHYLLSAAVAVRERHPDARFFVSDDIEWAKMNLNLPNCVFEVHAQPOPQDADHLYLMSL CKGHIANSTFSFWGAYLSMGSAAIAYPKQWFAEPTWNAPDIFLGHWIAL	273
Butyrivibrio fibrisolvens	WP_02275 2732.1	551008155	glycosyl transferase [Butyrivibrio fibrisolvens]	22.08	NILIRVAGGLGNQMQQYAMYRKLKSLGKEVKLDLSWFDVENQEQQLAPRKCELKYFDGVDFEECTDAERA YFTKRSILTALNKVFPATCKFEETEMFHEIYFSFKDKYLEGFLCNKYDDILPFIQNEIVFPKHSDPKMQKRN EELMERMDGWHTASIHLLRRGDYITEPQNEALFGNIATDAYDAIRYVLDKDYQTHFYFISNDPEYAREHYS DESRVITVGTNDGDSLLDMELMSHCYRICANSTFSFWGARLNKRSDEKEMIRTFKMRNNQEVAREMTD YWKDWILIDEKGNRIF	PCT/US2015/030823

Lewinella persica	WP_02057 1066.1	522059857	protein [Lewinella persica]	22.04	MVISRLHSLGNQMFQYAFARRIQLQNVKLRIDLSILLDSRPPDGYIKREYDLDIFKLSPAYHCNPTSLRILYA PGKYRWSQVVRDLARKGYPVYMEKFSVDNTLLDSPDNIYQGYWQSERFSEVANTIRKDFAFQHSIQP QSESLAREIRKEDSVCLNIRRKDYLASPTHNVTDETYENCICQMRERFSGARFFLSDDLVWCREEFFADHD VVIVGHDHAGPKFGNYLQLMAQCHHYIIPNSTFAWWAAWLGERTGSVIMAPERWFGTDEFDYRDVWPER WLKVPN
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OTHER EMBODIMENTS

While the invention has been described in conjunction with the detailed description thereof, the foregoing description is intended to illustrate and not limit the scope of the invention, which is defined by the scope of the appended claims. Other aspects, advantages, and modifications are within the scope of the following claims.

The patent and scientific literature referred to herein establishes the knowledge that is available to those with skill in the art. All United States patents and published or unpublished United States patent applications cited herein are incorporated by reference. All published foreign patents and patent applications cited herein are hereby incorporated by reference. Genbank and NCBI submissions indicated by accession number cited herein are hereby incorporated by reference. All other published references, documents, manuscripts and scientific literature cited herein are hereby incorporated by reference.

While this invention has been particularly shown and described with references to preferred embodiments thereof, it will be understood by those skilled in the art that various changes in form and details may be made therein without departing from the scope of the invention encompassed by the appended claims.

What is claimed is:

1. A method for producing a fucosylated oligosaccharide in a bacterium comprising providing bacterium comprising an exogenous lactose-utilizing $\alpha(1,2)$ fucosyltransferase enzyme, wherein the amino acid sequence of said enzyme comprises at least 22% identity to FutC (SEQ ID NO: 1).
2. A method for producing a fucosylated oligosaccharide in a bacterium comprising providing bacterium comprising an exogenous lactose-utilizing $\alpha(1,2)$ fucosyltransferase enzyme, wherein the amino acid sequence of said enzyme comprises at least 25% identity to FutN (SEQ ID NO: 3).
3. The method of claim 1 or 2, wherein said $\alpha(1,2)$ fucosyltransferase enzyme comprises any one selected from *Parabacteroides johnsonii* FutX, *Lachnospiraceae* sp. FutQ, *Prevotella melaninogenica* FutO, *Prevotellaa* sp. FutW, *Bacteroides* sp. FutZA, *Tannerella* sp. FutS, *Clostridium bolteae* +13 FutP, *Bacteroides caccae* FutU, *Salmonella enterica* FutZ, *Methanosphaerula palustries* FutR, *Butyrivibrio* FutV, *Akkermansia muciniphilia* FutY, *Clostridium bolteae* FutP, or a functional variant or fragment thereof.
4. The method of any one of the preceding claims, wherein said $\alpha(1,2)$ fucosyltransferase enzyme comprises any one of amino acid sequences SEQ ID NO: 10-21 and 292, or a functional variant or fragment thereof.
5. The method of any one of the preceding claims, further comprising retrieving the fucosylated oligosaccharide from said bacterium or from a culture supernatant of said bacterium.
6. The method of any one of the preceding claims, wherein said fucosylated oligosaccharide comprises 2'-fucosyllactose (2'-FL), lactodifucotetraose (LDFT), Lacto-N-fucopentaose I (LNF I), or lacto-N-difucohexaose I (LDFH I).

7. The method of any one of the preceding claims, wherein the bacterium further comprises an exogenous lactose-utilizing $\alpha(1,3)$ fucosyltransferase enzyme and/or an exogenous lactose-utilizing $\alpha(1,4)$ fucosyltransferase enzyme.
8. The method of claim 7, wherein the exogenous lactose-utilizing $\alpha(1,3)$ fucosyltransferase enzyme comprises a *Helicobacter pylori* 26695 *futA* gene.
9. The method of claim 7, wherein the exogenous lactose-utilizing $\alpha(1,4)$ fucosyltransferase enzyme comprises a *Helicobacter pylori* UA948 FucTa gene or a *Helicobacter pylori* strain DMS6709 FucT III gene.
10. The method of any one of the preceding claims, wherein said bacterium further comprises a reduced level of β -galactosidase activity, a defective colanic acid synthesis pathway, an inactivated ATP-dependent intracellular protease, an inactivated *lacA*, or a combination thereof.
11. The method of claim 10, wherein said method further comprises culturing said bacterium in the presence of tryptophan and in the absence of thymidine.
12. The method of claim 10, wherein said reduced level of β -galactosidase activity comprises a deleted or inactivated endogenous *lacZ* gene and/or a deleted or inactivated endogenous *lacI* gene of said bacterium.
13. The method of claim 12, wherein said reduced level of β -galactosidase activity further comprises an exogenous *lacZ* gene or variant thereof, wherein said exogenous *lacZ* gene or variant thereof comprises an β -galactosidase activity level less than wild-type bacterium.
14. The method of claim 10, wherein said reduced level of β -galactosidase activity comprises an activity level less than wild-type bacterium.
15. The method of claim 14, wherein said reduced level of β -galactosidase activity comprises less than 6,000 units of β -galactosidase activity.

16. The method of claim 14, wherein said reduced level of β -galactosidase activity comprises less than 1,000 units of β -galactosidase activity.
17. The method of claim 10, wherein said bacterium comprises a *lacIq* gene promoter immediately upstream of a *lacY* gene.
18. The method of claim 10, wherein said defective colanic acid synthesis pathway comprises the inactivation of the *wcaJ* gene of said bacterium is deleted.
19. The method of claim 10, wherein said inactivated ATP-dependent intracellular protease is a null mutation, inactivating mutation, or deletion of an endogenous *lon* gene.
20. The method of claim 19, wherein said inactivating mutation of an endogenous *lon* gene comprises the insertion of a functional *E. coli lacZ⁺* gene.
21. The method of claim 10, wherein said bacterium further comprises a functional lactose permease gene.
22. The method of claim 21, wherein said bacterium comprises *E. coli lacY*.
23. The method of claim 10, wherein said bacterium further comprises an exogenous *E. coli rcsA* or *E. coli rcsB* gene.
24. The method of claim 10, wherein said bacterium further comprises a mutation in the *thyA* gene.
25. The method of claim 10, wherein said bacterium accumulates intracellular lactose in the presence of exogenous lactose.
26. The method of claim 10 wherein said bacterium accumulates intracellular GDP-fucose.
27. The method of any one of the preceding claims, wherein said bacterium is *E. coli*.

28. The method of any one of the preceding claims, wherein said production strain is a member of the *Bacillus*, *Pantoea*, *Lactobacillus*, *Lactococcus*, *Streptococcus*, *Propionibacterium*, *Enterococcus*, *Bifidobacterium*, *Sporolactobacillus*, *Micromonospora*, *Micrococcus*, *Rhodococcus*, or *Pseudomonas* genus.

29. The method of any one of the preceding claims, wherein said production strain is selected from the group consisting of *Bacillus licheniformis*, *Bacillus subtilis*, *Bacillus coagulans*, *Bacillus thermophilus*, *Bacillus laterosporus*, *Bacillus megaterium*, *Bacillus mycoides*, *Bacillus pumilus*, *Bacillus lentus*, *Bacillus cereus*, and *Bacillus circulans*, *Erwinia herbicola* (*Pantoea agglomerans*), *Citrobacter freundii*, *Pantoea citrea*, *Pectobacterium carotovorum*, *Xanthomonas campestris*, *Lactobacillus acidophilus*, *Lactobacillus salivarius*, *Lactobacillus plantarum*, *Lactobacillus helveticus*, *Lactobacillus delbrueckii*, *Lactobacillus rhamnosus*, *Lactobacillus bulgaricus*, *Lactobacillus crispatus*, *Lactobacillus gasseri*, *Lactobacillus casei*, *Lactobacillus reuteri*, *Lactobacillus jensenii*, *Lactococcus lactis*, *Streptococcus thermophiles*, *Propionibacterium freudenreichii*, *Enterococcus faecium*, *Enterococcus thermophiles*, *Bifidobacterium longum*, *Bifidobacterium infantis*, *Bifidobacterium bifidum*, *Pseudomonas fluorescens* and *Pseudomonas aeruginosa*.

30. The method of any of the preceding claims, wherein said bacterium comprises a nucleic acid construct comprising an isolated nucleic acid encoding said $\alpha(1,2)$ fucosyltransferase enzyme.

31. The method of claim 30, wherein said nucleic acid is operably linked to one or more heterologous control sequences that direct the production of the enzyme in the bacterium.

32. The method of claim 31, wherein said heterologous control sequence comprises a bacterial promoter and operator, a bacterial ribosome binding site, a bacterial transcriptional terminator, or a plasmid selectable marker.

33. A purified fucosylated oligosaccharide produced by any one of the preceding claims.

34. A nucleic acid construct comprising an isolated nucleic acid encoding a lactose-utilizing $\alpha(1,2)$ fucosyltransferase enzyme for the production of said enzyme in a host

bacteria production strain, wherein the amino acid sequence of said enzyme comprises at least 22% identity to FutC (SEQ ID NO: 1).

35. A nucleic acid construct of comprising an isolated nucleic acid encoding a lactose-utilizing $\alpha(1,2)$ fucosyltransferase enzyme for the production of said enzyme in a host bacteria production strain, wherein the amino acid sequence of said enzyme comprises at least 25% identity to FutN (SEQ ID NO: 3).

36. The construct of claim 34 or 35, wherein said $\alpha(1,2)$ fucosyltransferase enzyme comprises any one selected from *Parabacteroides johnsonii* FutX, *Lachnospiraceae* sp. FutQ, *Prevotella melaninogenica* FutO, *Prevotella* sp. FutW, *Bacteroides* sp. FutZA, *Tannerella* sp. FutS, *Clostridium bolteae* +13 FutP, *Bacteroides caccae* FutU, *Salmonella enterica* FutZ, *Methanosphaerula palustries* FutR, *Butyrivibrio* FutV, *Akkermansia muciniphilia* FutY, *Clostridium bolteae* FutP, or a functional variant or fragment thereof.

37. The construct of any one of claims 34-36, wherein said $\alpha(1,2)$ fucosyltransferase enzyme comprises any one of amino acid sequences SEQ ID NO: 10-21 and 292, or a functional variant or fragment thereof.

38. The construct of any one of claims 34-37 wherein said nucleic acid is operably linked to one or more heterologous control sequences that direct the production of the enzyme in the bacterium.

39. The construct of claim 38, wherein said heterologous control sequence comprises a bacterial promoter and operator, a bacterial ribosome binding site, a bacterial transcriptional terminator, a plasmid selectable marker, and/or an origin of replication.

40. An isolated bacterium comprising an isolated nucleic acid encoding a lactose-accepting $\alpha(1,2)$ fucosyltransferase enzyme, wherein the amino acid sequence of said enzyme encoded by said nucleic acid comprises at least 22% identity to FutC (SEQ ID NO: 1).

41. An isolated bacterium comprising an isolated nucleic acid encoding a lactose-accepting $\alpha(1,2)$ fucosyltransferase enzyme, wherein the amino acid sequence of said

enzyme encoded by said nucleic acid comprises at least 25% identity to FutN (SEQ ID NO: 3).

42. The isolated bacterium of claim 40 or 41, wherein said $\alpha(1,2)$ fucosyltransferase enzyme comprises any one selected from *Parabacteroides johnsonii* FutX, *Lachnospiraceae* sp. FutQ, *Prevotella melaninogenica* FutO, *Prevotellaa* sp. FutW, *Bacteroides* sp. FutZA, *Tannerella* sp. FutS, *Clostridium bolteae* +13 FutP, *Bacteroides caccae* FutU, *Salmonella enterica* FutZ, *Methanosphaerula palustries* FutR, *Butyrivibrio* FutV, *Akkermansia muciniphilia* FutY, *Clostridium bolteae* FutP, or a functional variant or fragment thereof.

43. The isolated bacterium of any one of claims 40-42, wherein said $\alpha(1,2)$ fucosyltransferase enzyme comprises any one of amino acid sequences SEQ ID NO: 10-21 and 292, or a functional variant or fragment thereof.

44. The isolated bacterium of any one of claims 40-43, further comprising a $\alpha(1,3)$ fucosyltransferase enzyme and/or an $\alpha(1,4)$ fucosyltransferase enzyme.

45. The isolated bacterium of any one of claims 40-44, wherein said bacterium is *Escherichia coli*.

46. The isolated bacterium of any one of claims 40-45, wherein said bacterium further comprises reduced level of β -galactosidase activity, a defective colanic acid synthesis pathway, an inactivated adenosine-5'-triphosphate (ATP)-dependent intracellular protease, an inactivated endogenous *lacA* gene, or any combination thereof.

47. The isolated bacterium of claim 46, wherein said bacterium comprises the genotype $\Delta ampC::P_{trp}^B cI$, $\Delta(lacI-lacZ)::FRT$, $P_{lacI} lacY^+$, $\Delta wcaJ::FRT$, $thyA::Tn10$, $\Delta lon:(npt3, lacZ^+)$.

Synthetic routes for neutral hMOS

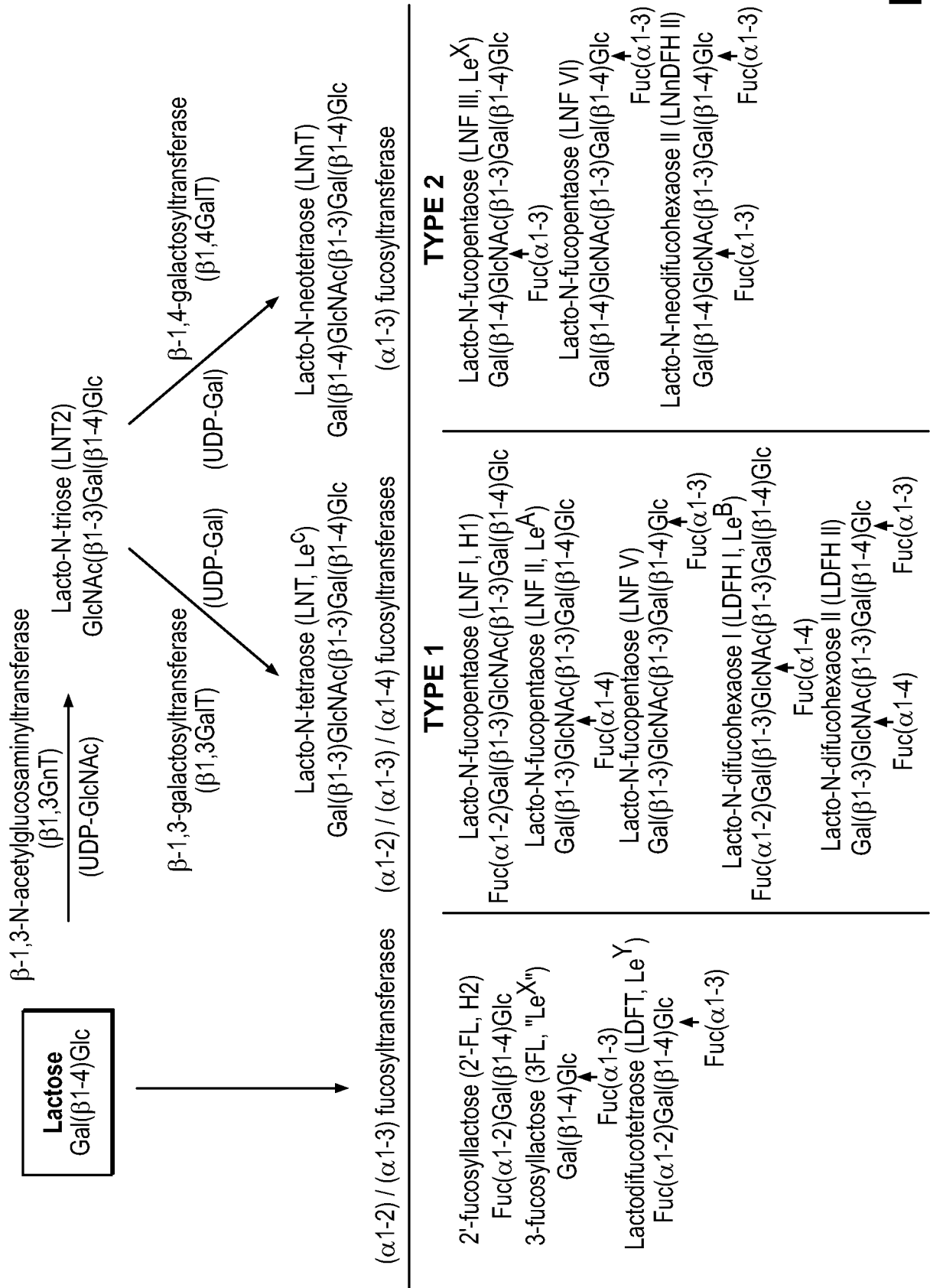


FIG. 1

Metabolic engineering for 2'-FL production in *E. coli*

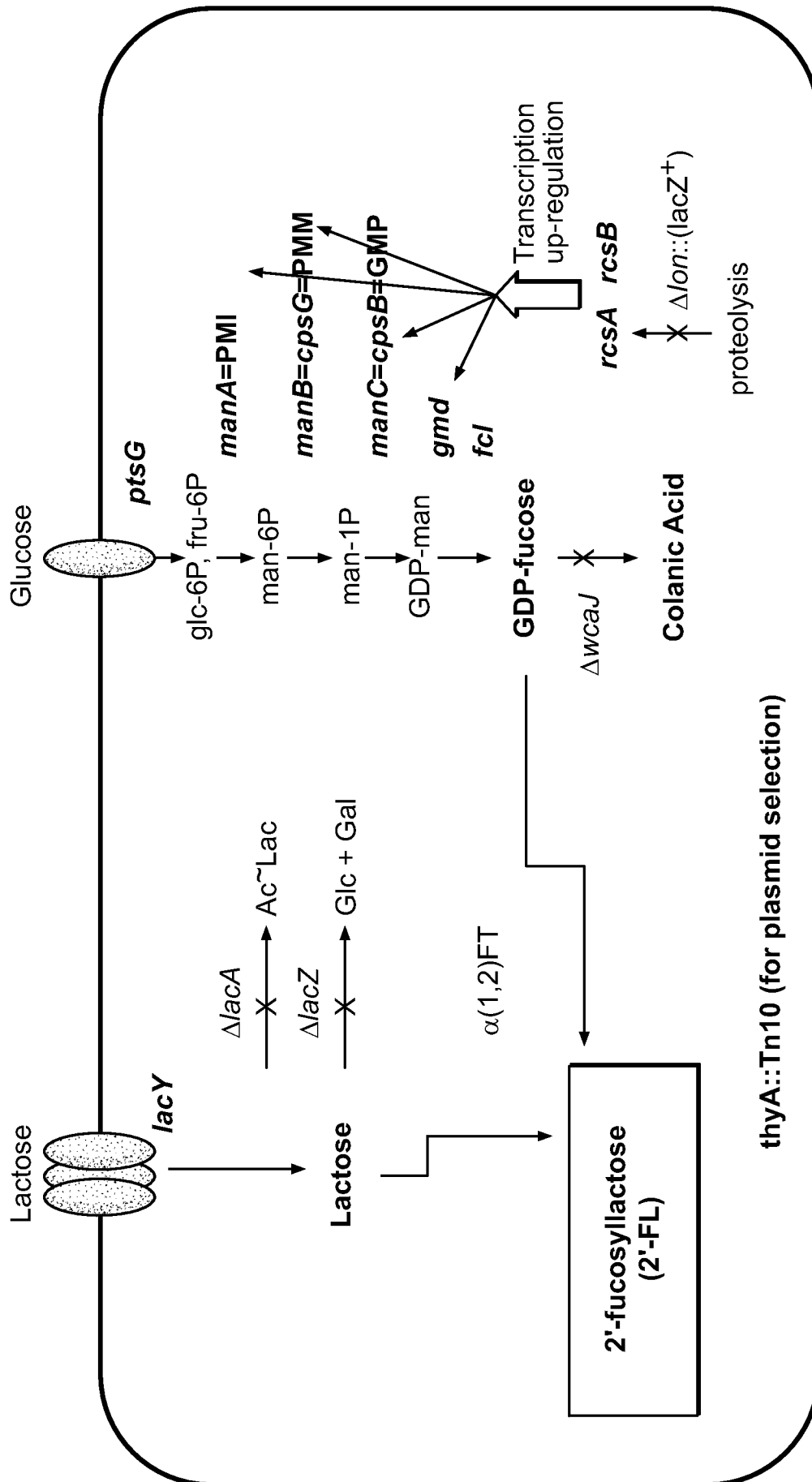


FIG. 2

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	1	2	3	4
H.pylori futC	1	70.10	23.97	22.57
H.mustelae futL	2	70.10	27.04	21.36
Bacteroides vulgatus futN	3	23.97	27.04	31.23
E. coli O126 wbgL	4	22.57	21.36	31.23

FIG. 3A

H. pylori futC	MAFKVVQI	CGGLGNQM	FQYAF	AKSLQKHS	NTPVLL	DI	TSFDWS	DRKMQL	100											
H. mustelae futL	MDFKIVQ	VHGGGLGNQM	FQYAF	AKSLQTHLN	IPVLLD	TTWFDY	GNRELGLH		100											
Bacteroides vulgatus futN	M - - RL	IKVTGGLGNQM	F IYAF - YRR	MKKYYPKVR	IDLSDMMH	YKVHYGYE			95											
E. coli 0126 wbgL	M - - SI	IRLQGGGLGNQL	FQFS	EYALSKING	TPLYFD	ISHYA	ENDDHGGYR		103											
H. pylori futC	LKPSRL	TYFFGYFQD	PRYFDA	ISPLIKQT	FTLPPPP	NNKNNKKE	EEYQ		200											
H. mustelae futL	FEPSRI	AYFHGYFQD	PRYFED	ISPLIKQT	FTLPHPT	EH - - - -	- AEQYS		193											
Bacteroides vulgatus futN	- - - WPL	LYFKGFYQSE	RFADIKDEV	RESFTFDK	- - - -	- NKANSRS			190											
E. coli 0126 wbgL	AQKWSK	KYIGYWQSE	HFHKKH	LDLKEFF	- IPK - - - -	- NVSE	EQAN		199											
H. pylori futC	FVFCED	LEFT - QN	LDLGYPF	MDMTTRD	KEEEAYWD	MLLMQSC	QHGI IANS		298											
H. mustelae futL	FLFCED	LEFV - QN	LDLGYPF	VDMTTRD	GA - - AH	WDMMLMQSC	KHGI ITNS		287											
Bacteroides vulgatus futN	YIFSD	DLAWVKEN	LP - - LQ	NAVYI	DWNTDEL	SWQDMM	LMSSHCKHHI ICNS		282											
E. coli 0126 wbgL	FIFSD	DLFWCKEN	IELLSK	KYNIYY	SEDL	SQEE	DLWMSLANHHI IANS		298											
	LFPI	DLPY	SAKEI	IAKMQHL	- - - -	- PKLVR	DALKCMGFDRVSQE	IVFEYEPKL	100											
	LFPI	DLQC	SAQQI	AAAHMQNL	- - - -	- PRLVR	GALRRMGLGRVSKE	IVFEYMPKL	100											
	MHRV	FNLP	HTFC	INQP - L	KKVIEFL	- - - -	- FFKKI	YERKQAPNSLRAF	- - EKK - YF 95											
	LN	NL - QI	PEEY	LQXY	TRKJ	NNIYKFL	VRGSRLYPEI	FLFLGFCNEFHAYGYDFE	- YI 103											
	CKLS	LILAA - KNS	VFVHIR	RRGDYV	G - - - -	- IGCQL	GIDYQKKALEYMAKRV	PNMEL	200											
	RKLS	QILAA - KNS	VFVHIR	RRGDYMR	- - - -	- LGWQL	DISYQLRAIAYMAKRV	QNLLEL	193											
	LN	MLEI	LDKDENA	VS	LHIR	RRGDY	LQPKHWAT	TG	SV	QCLPY	YQNAI	AEMS	RRVA	SPSY	190					
	LLA	AKIL	ES - QSS	LSIH	IR	RRGDY	TKNTAT	LT	THG	VCSLEY	YKKAL	NKIR	DLAM	IRDV	199					
	TS	SWWA	AYL	EN	PEKI	IGPK	HWLFGHENI	- - -	- LCKE	WVKIES	HFEVKS	QKYNA	*	301						
	TS	SWWA	AYL	IKNP	PEKI	IGPS	HWIYGNENI	- - -	- LCKD	WVKIES	QFETKS	*	- - - -	- 287						
	TF	SWWG	AWLNP	NDKT	VI	VP	SRWFQHSE	APDI	YP - -	- TG	WIKVP	VS	*	- - - -	- 282					
	SF	SWWG	AYL	GT	SA	SQI	VI	YPT	PWYDI	TP	KN	TYI	PI	VN	HWI	NVDK	HSSC	*	- - - -	- 298

FIG.

FIG. 3B

FIG. 4A	FIG. 4B
FIG. 4C	FIG. 4D
FIG. 4E	FIG. 4F

FIG. 4

H. pylori futC	MAFKVVQICGGLGNQMFAQYAFAKSLQKHSNTP--VLLDITSFWDSDR
H. mustelae futL	MDFKIVQVHGGGLGNQMFAQYAFAKSLQTHLNIP--VLLDTTWFDYGNR
Bacteroides vulgatus futN	M--RLIKVTGGLGNQMFYAFYLRMKKYY--PKVRIDLSDMMH--
E. coli 0126 wbgL	M--SIRLQGGGLGNQLFQFSFGYALSKINGTP--LYFDISHYAEN--
Prevotella melanogenica FutO YP_003814512.1	M-K-IVKILGGLGNQMFAQYALYLSQES-FPKERVALDLSSF--
Clostridium bolteae+13 FutP WP_002570768.1	m-v-iikmmglgngMQYALYKAFEQK-HID--VYADLAWYKNKSV
Lachnospiraceae sp. FutQ WP_009251343.1	M-V-IVQLSGGLGNQMFYALYLSLAK-GKEVKID-DVTCYEGPGT
Methanospaerula palustris FutR YP_002467213.1	M-I-IVRLKGGGLGNQLSQYALGRKIAHL-HNTE-LKLDTTWFTT SS
Tannerella sp. FutS WP_021929367.1	M-VRIVEIGGLGNQMFAQYAFSLYLNKSHIWDRLYVDIEAMKT--
Bacteroides caccae FutU WP_005675707.1	M-K-IVKILGGLGNQMFAQYALFLSLKER-FPHEQVMIDTSCF--
Butyrivibrio FutV WP_022772718.1	M-I- QLKGGGLGNQMFAQYALYKSLKKR-GKEVKID-DKTGFVNDKL
Prevotella sp. FutW WP_022481266.1	M--RLVKMIGGLGNQMFYAFYQLQMRKRP--SNVRIDLTDMMH--
Parabacteroides johnsonii FutX WP_008155883.1	M--RLIKMIGGLGNQMFYAFYLMKHHY--PDTNIDLSDMVH--
Akkermansia muciniphila FutY YP_001877555	M--RLFGGLGNQLFQYAFLFALSRQGG--KARLETSSYEHD DK
Salmonella enterica FutZ WP_023214330	M-YSC--LSGGLGNQMFAQYAAAY LKQYFQSTTLVLDDSYYSQPKR
Bacteroides sp. FutZA WP_022161880.1	M--RLIKMTGGLGNQMFYAFYLRMKKRY--PKVRIDLSDMVH--
Clostridium bolteae FutP WP_002570768.1	M-----FQYALYKAFEQK-HID--VYADLAWYKNKSV

FIG. 4A

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KMQL-----ELFPIDLPYASAKEIAIAKMQHLPKLVRDALKCM-----GFDRVSQEI VF 94
ELGL-----HLFPIDLQCASAQQIAAAHMQNL PRLVRGALRRM-----GLGRVSKEI VF 94
-----YKVHYGYEMHRVFNLP-----TEFCINQPLKKVIEFLF-FKKIY-----ERKQAPN 85
-----DDHGGYRLNN-----QIPEEYLQYYTPKINN|YKFLVGRSRLYPEI|FLPLGFCNEFHAY 96
-----HGYHLHNGFELENI|FSVTAQKASAADIMRIAYYYPNYLLWRIGKRFLPRRKGM-----CLESSL 99
-----KFELYN-----FGIKINVASEKDINRLSDCQADF-VSRI RRIKIFGKKKSF-----VSEKNDS 93
RPR-----QLDVFGITYDRASREELTEMTDASMD-ALSRVRRKLTG-RRTK-----AYRERDI 94
DTPRTYRLNN-----YNIIGT|ASAKEIQLIERGRAQGRGYLLSK|SDL LTPMYRRT-----YVRERMH 102
-----YDRHYGLELEKVFNL|SLCPI|SNRLHRNLQ-----KRSFAKH FVKS-----LYEHSEC 90
-----RNYPLHNGFEVDRI|FAQKAPVASWRNI|KVAYPPYPNYRFWKIGKY|LPKRKTM-----CVERKNF 99
RIP-----VLSRWGVEYDRATDEEI|INLTDSKMD-LFSRI RRRKLTG-RKTF-----RIDEESG 94
-----YNVHYGYELHKVFLPR-----TEFCMNQPLKKVLEFLF-FRTIV-----ERKQH-G 84
-----YKVHNGYEMNRI|FDLSQ-----TEFCINRTLKKILEFLF-FKKIY-----ERRQDPS 85
RVC-----ELHHFRVSLPIEGGPPPPWA-----FRKSRIPACLRSLFAAPKYPH-----FREEKRH 89
DTVRSLELNG-----FNI|SYDRFSFAD-----EKEKI|LLRKFKRNPFPKQ|SEIDS|ALPGKYALSDRA 104
-----YHVHGYEMHRVFNLP-----TEFCINQPLKKVIEFLF-FKKIY-----ERKQDPN 85
-----KFELYN-----FGIKINVASEKDINRLSDCQADF-VSRI RRIKIFGKKKSF-----VSEKNDS 80

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FIG. 4B

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H. pylori futC	EYEPKLLKPSR-LTYFFGYFQDPRYF--DAISPLIKQTFTLPPPPEN
H. mustelae futL	EYMPELFEPSR-IAYFHGYFQDPRYF--EDISPLIKQTFTLPHPTEH
Bacteroides vulgatus futN	SLRAFEKKYFWPLLYFKGFYQSERFF--ADIKDEVRESFTF--DK
E. coli 0126 wbgL	GYDFEYIAQKWKSKYIGYWQSEHFFHKHIL--DLKEFFI-----P
Prevotella melaninogenica FutO YP_003814512.1	RFDESVLR-QEGNRYFDGYWQDERYF--AAYREKVLKAFTFPAFKRA
Clostridium bolteae+13 FutP WP_002570768.1	CYENDILR-M-DNVYLSGYWQTEKYF--SNTREKLLEDYSF-ALVNS
Lachnospiraceae sp. FutQ WP_009251343.1	NFDPLVME--KDPALLEGCFQSDKYF--RDCEGRVREAYRFRGIESG
Methanosphaerula palustris FutR YP_002467213.1	TFDKAILT-VPDNVYLDGYWQTEKYF--KDIEELRREVTLKDEPDS
Tannerella sp. FutS WP_021929367.1	EFDEPVYRGLRPYRYRGYWQNEGYFV--DIEPMIREAFQFNVNILS
Bacteroides caccae FutU WP_005675707.1	SFDAAVLT-RKGDICYDGYWQHHEEYF--CDMKETIWEAFSFEPEVDG
Butyrivibrio FutV WP_022772718.1	KFNPEILE--KENAYLVGYWQCDKYFDDKDVVREIREAFKKPQE--
Prevotella sp. FutW WP_022481266.1	RMEPYTCQYVWPLVYFKGFYQSERYF--SEVKDEVRECFTF--NP
Parabacteroides johnsonii FutX WP_008155883.1	TLYPEKRYFWPLLYFKGFYQSERFF--FDIKDDVRKAASF--NL
Akkermansia muciniphila FutY YP_001877555	GFDPGLAAPRRHTYFKGYFQTEQYFL--HCREQLCREFRLKTPLTP
Salmonella enterica FutZ WP_023214330	FYTFETIKNIDKACLSFYQDAD--LLNKYKQLLPLFELR--DD
Bacteroides sp. FutZA WP_022161880.1	SLRAFEKKYLWPLLYFKGFYQSERFF--ADIKDEVKRAFTF--DS
Clostridium bolteae FutP WP_002570768.1	CYENDILR-M-DNVYLSGYWQTEKYF--SNTREKLLEDYSF-ALVNS

FIG. 4C

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NKNNKKEEYQCKLSL|AAKNSV|FVH|IRRGDYVG|-----|GCQLGIDYQKKALEYMAKRV|-----P 196
|-----AEQYSRKLSQ|AAKNSV|FVH|IRRGDYMR|-----LGWQLDISYQLRAIAYMAKRV|-----Q 189
NK-----ANSRSLNMLE|LDKDENA|VSLH|IRRGDY|LQ-PKHWA|TGSVCQLPYYQNA|AEMSRRV|-----AS 187
KN-----VSEQANL|AAK|LESQSSLS|H|IRRGDY|IK-NKTATL|THGVC|SLEYKKALNK|IRD|LAMI|-----196
EN-----L-----SLL-----EKL|DENS|ALH|VRRRGDYVG-NNL|---YQGI|CDLDY|YRTA|IEKMCAHV|-----TP 194
QV-----SEWEDS|I-----R-----NKNSV|S|H|IRRGDY|LQ-GEL|---YGGI|CTSLY|YAEA|EY|IKMRV|-----PN 187
AF-----PLPEDY|LRLEK|IEDCQSV|SVH|IRRGDY|LD-ESHGGLY|TGICTEAYYKEAFARMERLV|-----PG 198
IN-----LEMAERI|---QAC---HSVSLH|VRRRGDYVS-NPTTQQFHGCCS|IDY|YNRA|ISL|IEEKV|-----DD 201
KK-----TKA|IAS-----KMRREL|SVS|IHVRRRGDYEN-PEAKAMHGGI|CSLDY|YHKA|IDF|IRQRL|-----DN 192
RN-----KEIGALL-----QASD---SASLH|VRRRGDYVN-HPL|---FRGI|CDLDY|YKRA|IHMEERV|-----NP 195
LM-----TDASSWSTLQ-QIECCESV|SLHVRR|TDYVD-EEH|IH|HN-ICTEKY|YKNA|IDRV|RKQY|-----PS 196
AL-----ANRSSQQMMEQ|QN|DPPQAVS|H|IRRGDY|LN-PKH|YDT|IGC|ICQLP|YKHAV|SEI|KKYV|-----SN 186
NI-----ANPFS|LFL|LKQ|IEVDDQAVS|H|IRRGDY|LL-PRHWANTG|SVC0L|P|YKNA|IAEMENR|I|-----TG 187
EN-----ARILEDI|-----RSCCS|I|SLH|IRRTDY|LSNP|-----YLSPPPLEY|YLR|SMAEMEGR|LRAAGAPQE 191
LL-----DICKNLELYSL|QRSNN|TTALH|IRRGDY|VT-NQHA|AKYH|GVLD|I|SY|YNHAMEY|VERE|-----RG 205
SK-----VNARSAELLRR|LDADANAV|SLH|IRRGDY|LQ-PQHWAT|TGSVCQLPYYQNA|AEMNRRV|-----AA 187
QV-----SEWEDS|I-----R-----NKNSV|S|H|IRRGDY|LQ-GEL|---YGGI|CTSLY|YAEA|EY|IKMRV|-----PN 174

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FIG. 4D

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H. pylori futC	NMELFVFCEDLEFT-QNLDLGY - - - FMDMTTRDKEFEAYWDMMLMG
H. mustelae futL	NLELFLFCEDLEFV-QNLDLGY - - - FVDMTTRDGA - - AHWDMMLMQ
Bacteroides vulgatus futN	PS - YY I FSDD IAWVKENL - - - PLQNAVY I DWNTDE - DSWQDMMLMS
E. coli 0126 wbgL	- RDVF I FSDD I FWCKEN I ETL LSKKYN I YSEDLSQEE - - - DLWLMS
Prevotella melaninogenica FutO YP_003814512.1	SL - FC I FSND I TWCQHLQPYLKAP - VVYV TWNTGV - ESYRDMQLMS
Clostridium bolteae+13 FutP WP_002570768.1	AK - FFVFSDDVEWVKQED - - - FKG FV I VDRNEYS - SALSDMYLMS
Lachnospiraceae sp. FutQ WP_009251343.1	AR - FFLFSNDPEWTRHEFE - - - SKNCVL VEGSTED - TGYMDLYLMS
Methanosphaerula palustris FutR YP_002467213.1	PS - FF I FSDDL PWAKENLD - - I PGEKT - FVAHNGPE - KEYCDLWLMS
Tannerella sp. FutS WP_021929367.1	N ICFYLFSDDI NWVEENLQL - - - ENRC I I DWNQGE - DSWQDMY LMS
Bacteroides caccae FutU WP_005675707.1	QL - YCVFSNDMAWCESHLRALPGKEV VYVDWNKGA - ESYVDMRLMS
Butyrivibrio FutV WP_022772718.1	AV - FF I FTDDKEWCRDHFK - - - GPNF I VVELEEGDGT I AEMRLMS
Prevotella sp. FutW WP_022481266.1	PH - FYVFSDDL DWVKANL - - - PLENAQY I DWNKGA - DSWQDMMLMS
Parabacteroides johnsonii FutX WP_008155883.1	PS - YYVFSDD I SWVKEN I - - - PLKKAVYV TWNKGE - DSWQDMMLMS
Akkermansia muciniphilia FutY YP_001877555	SLRYF I FSDD I EWARQNLRPALP - - - HVHVD I NDG - GTGYFDLELMR
Salmonella enterica FutZ WP_023214330	KQNF I I FSDDVRWAQKAFLE - - - NDNCYV I NNSDYDFS I DMYLMS
Bacteroides sp. FutZA WP_022161880.1	PS - YYVFSDD IAWVKEN I - - - PLQNAVY I DWNKGE - ESWQDMMLMS
Clostridium bolteae FutP WP_002570768.1	AK - FFVFSDDVEWVKQED - - - FKG FV I VDRNEYS - SALSDMY LMS

FIG. 4E

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SCQHG I ANSTYSWWAAYL I ENPEK I IGPKHWF GHFN I L - - - - - CKEWK I FSHFEVK SQKVNA - 301
SCKHG I TNSTYSWWAAYL I KNPEK I IGP SHW I YGNEN I L - - - - - CKDWK I ESQFETKS * - - - - - 287
HCKHH I CNSTFSWWGAWL NPNMDKT V I VPSRWFQ - - - - - HSEAPD I YPT - - - - - GWIKV - - - - - PVS * - - - - - 282
LANHH I ANSSFSWWGAYL GTSA SQ I V I YPTPWYD I TPKN TY - - - - - I P I VNHW I NVDKHSSC * - - - - - 298
CCAHN I ANSSFSWWGAWL NQNREK VV I APKKWL N - - - - - MEECHFTLPA - - - - - S - - - - - W - - - - - KI - - - - - 288
LCKHN I ANSSFSWWA AAWL NRNEEK I V I APRRWLNGKCT - - - - - PD I WCK - - - - - KW - - - - - RI - - - - - 278
RCRHN I ANSSFSWWGAWL NENPEKK V I APAKWLN GR - - - - - ECRD I YTE - - - - - RW - - - - - RL - - - - - 289
LCQHH I ANSSFSWWGAWL GQDAEKM V I APRRWALSESFDTS D I PD - - - - - S - - - - - W - - - - - TI - - - - - 295
CCRHH I ANSSFSWWA AAWL NPNKNK I VLT P NKWFN - - - - - HTDAVG I VPK - - - - - SWIKI - - - - - PVF - - - - - 287
LCRHN I ANSSFSWWGAWL NRNPQKV VVAPERW MNSP I ED - - - - - PV - - - - - SD - - - - - KW - - - - - KL - - - - - 289
RCKHH I ANSSFSWWA AAWL NDSPEK I V I APQKW I NNR - - - - - DMDD I YTE - - - - - RWTK I - - - - - AL - - - - - 290
CCKHH I CNSTFSWWA AAWL NPSVEKT V I MPEQWTS - - - - - RQDSVDFVASCGRWVRV - - - - - KTE - - - - - 282
HCRHH I CNSTFSWWGAWL NPRKEK I V I APCRW FQ - - - - - HKETPDMPYK - - - - - EWIKV - - - - - PIN - - - - - 281
NCRHH I ANSTFSWWA AAWL NEHA EK I V I APR I WFNREEGDRYHTDDAL I P - - - - - GSWLR I - - - - - 290
LCKNN I ANSTYSWWGAWL NKYEDKL V I SPKQWFLGNNETS - - - - - - - - - - - LNASWITL - - - - - 298
HCRHH I CNSTFSWWGAWLDPHEDK I V I VPNRW FQ - - - - - HCETPN I YPA - - - - - GWVKV - - - - - AIN - - - - - 281
LCKHN I ANSSFSWWA AAWL NRNEEK I V I APRRWLNGKCT - - - - - PD I WCK - - - - - KW - - - - - RI - - - - - 265

```

FIG. 4F

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FIG. 5A

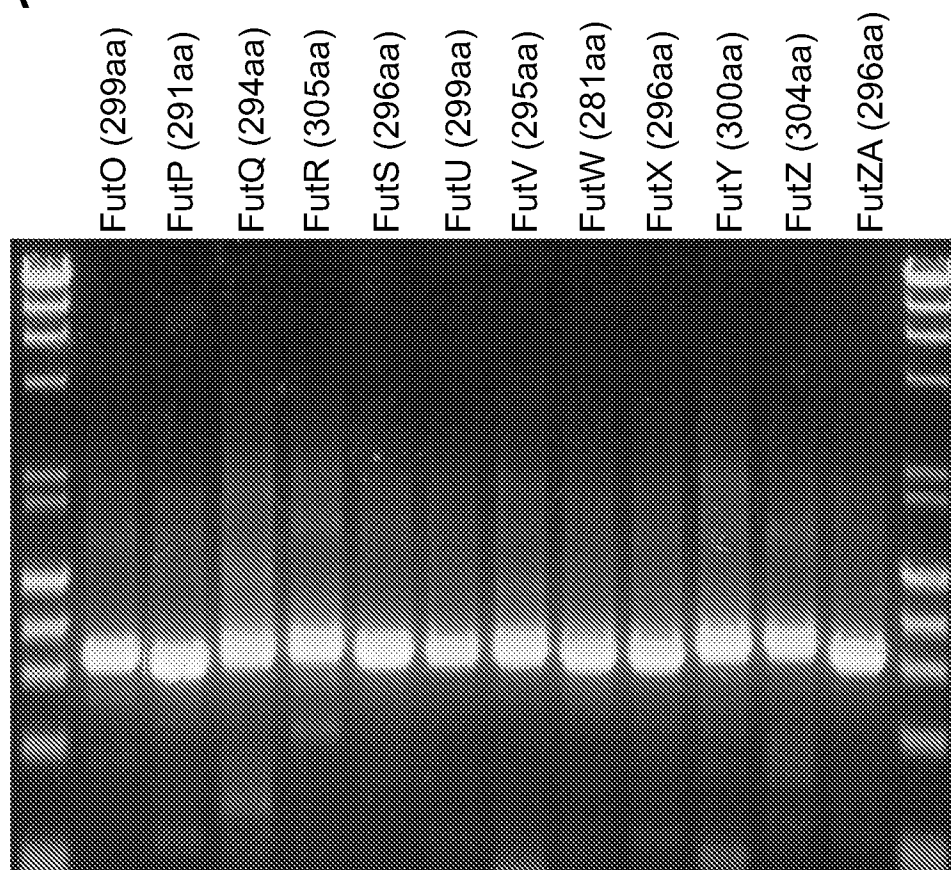
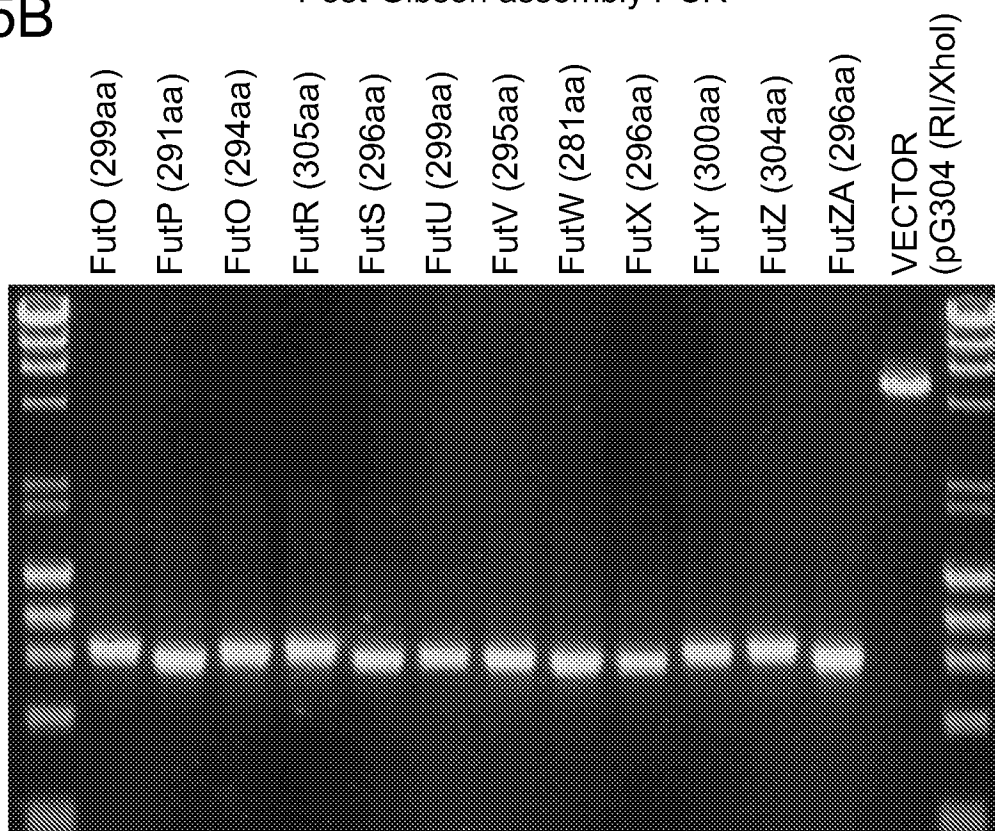
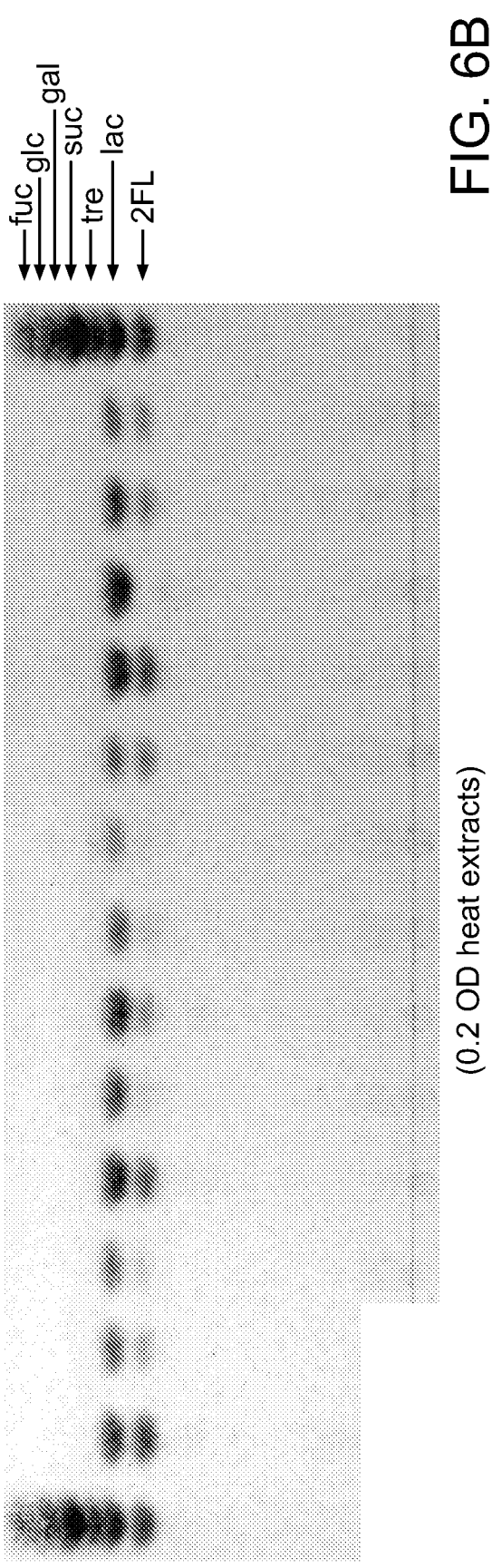
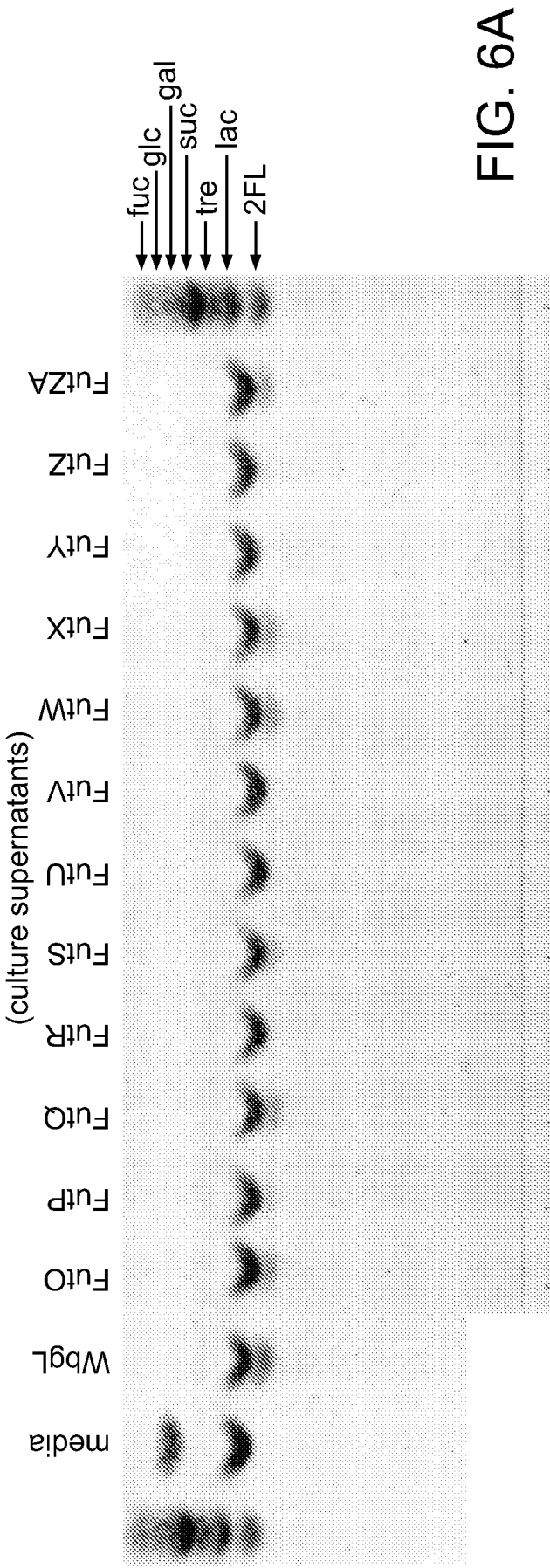


FIG. 5B

Post-Gibson assembly PCR



Gel-purified RI/XhoI syngene fragment



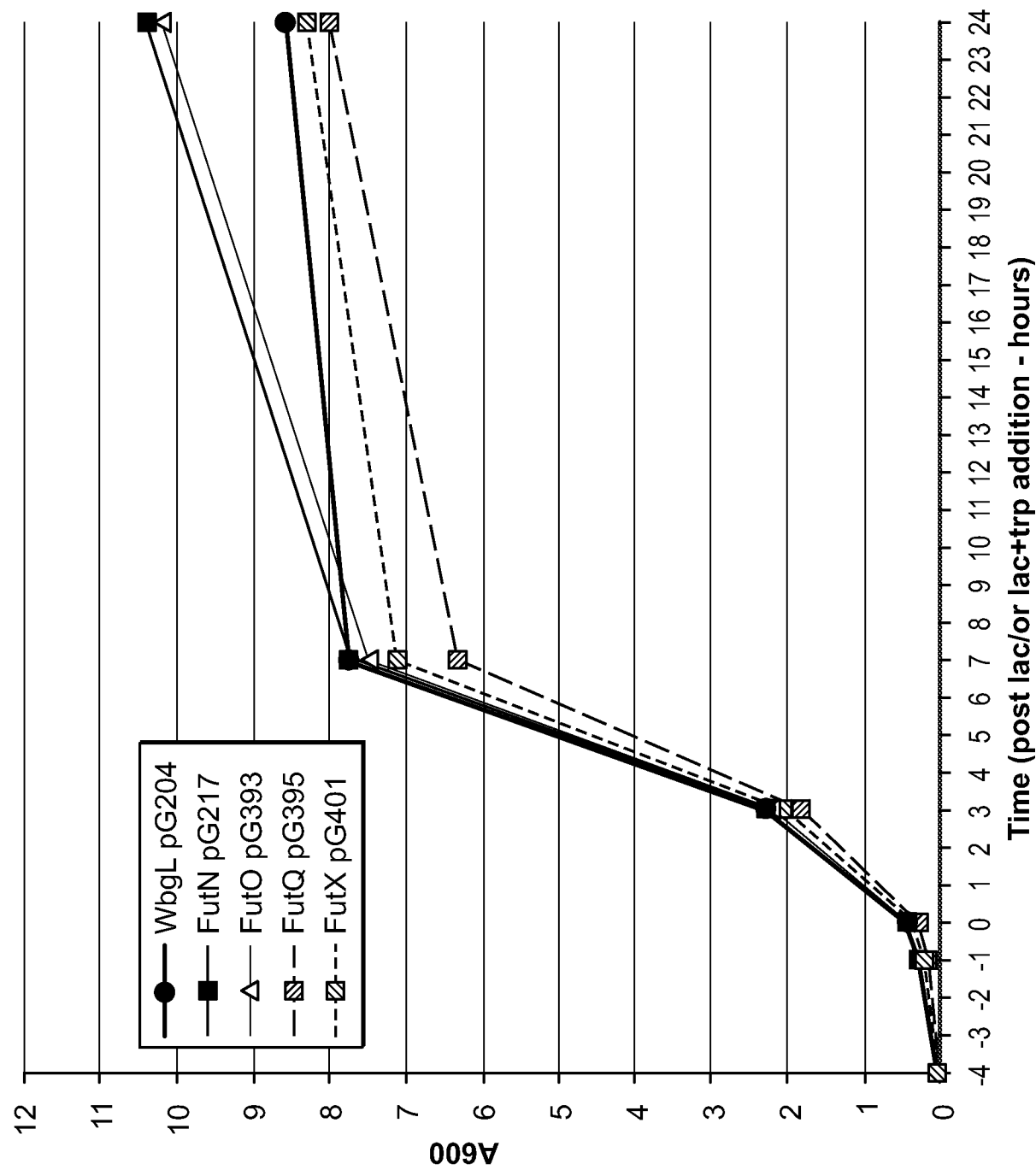


FIG. 7

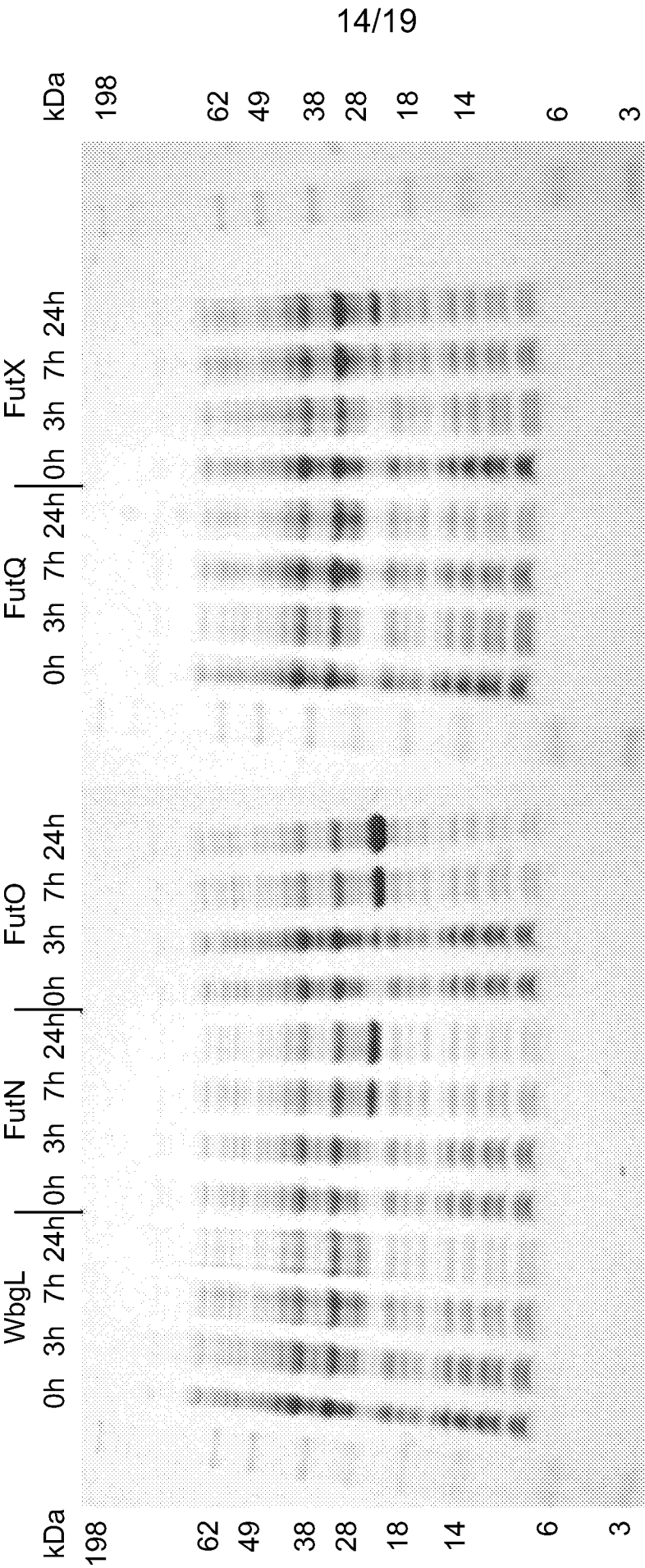


FIG. 8

FIG. 9A

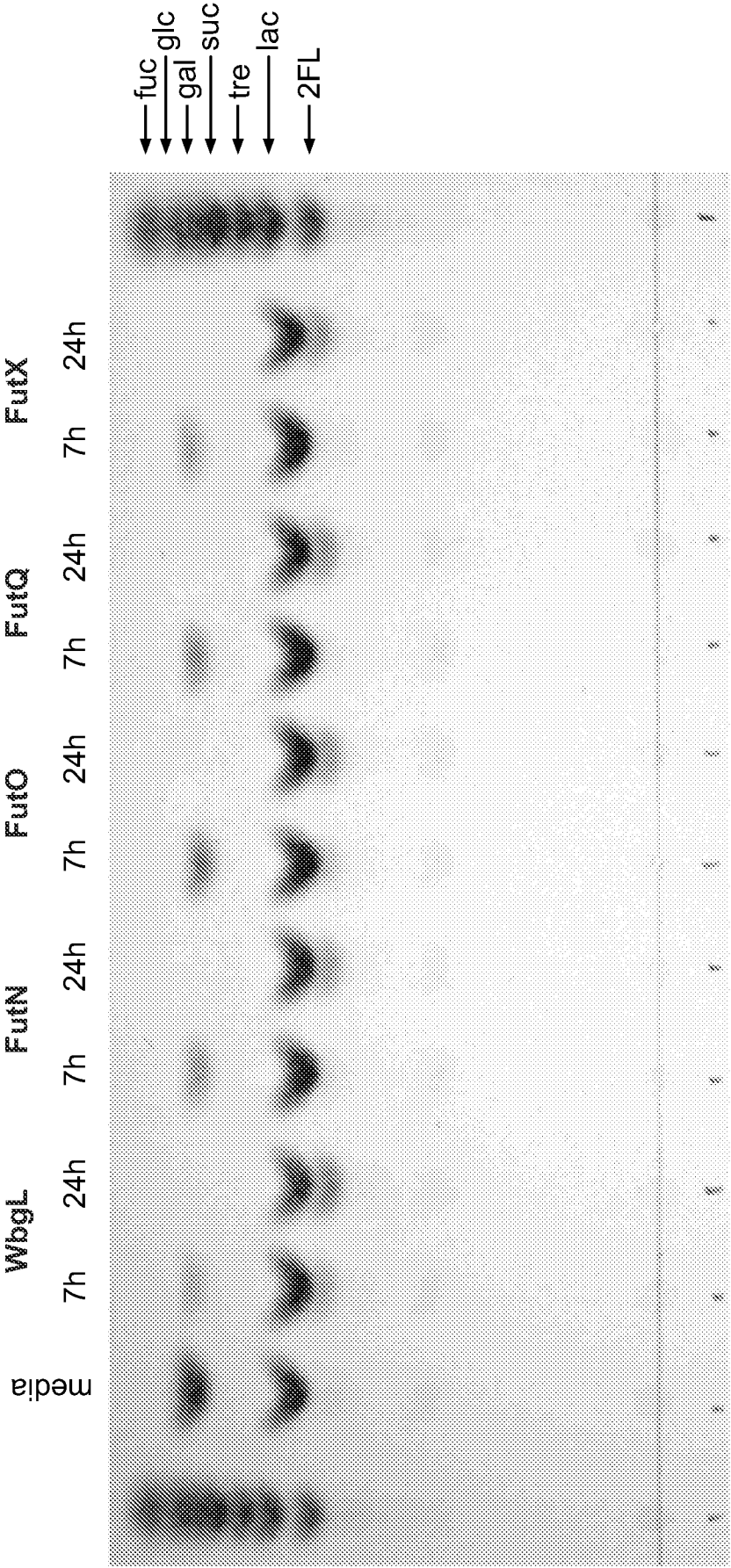
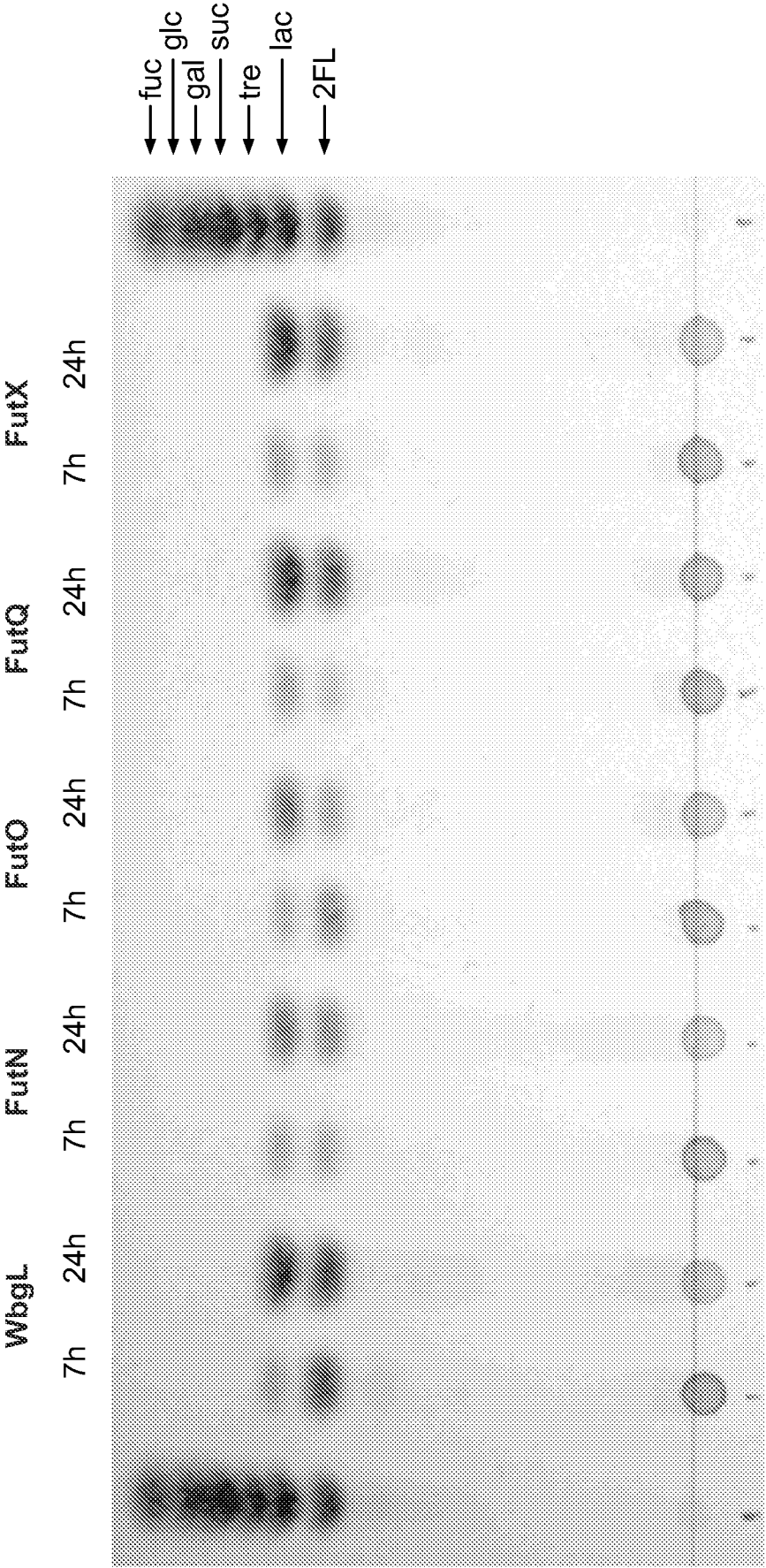
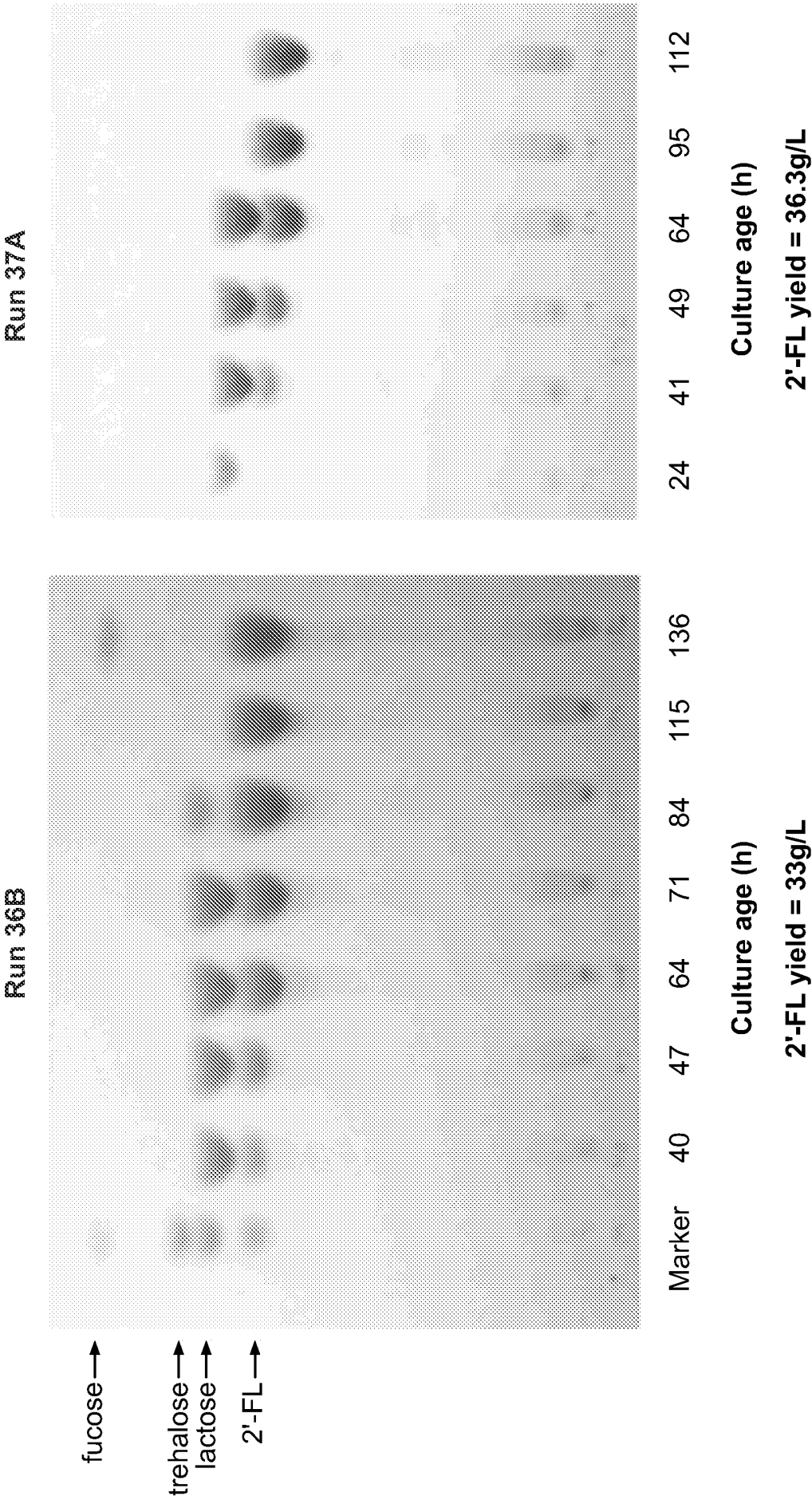


FIG. 9B





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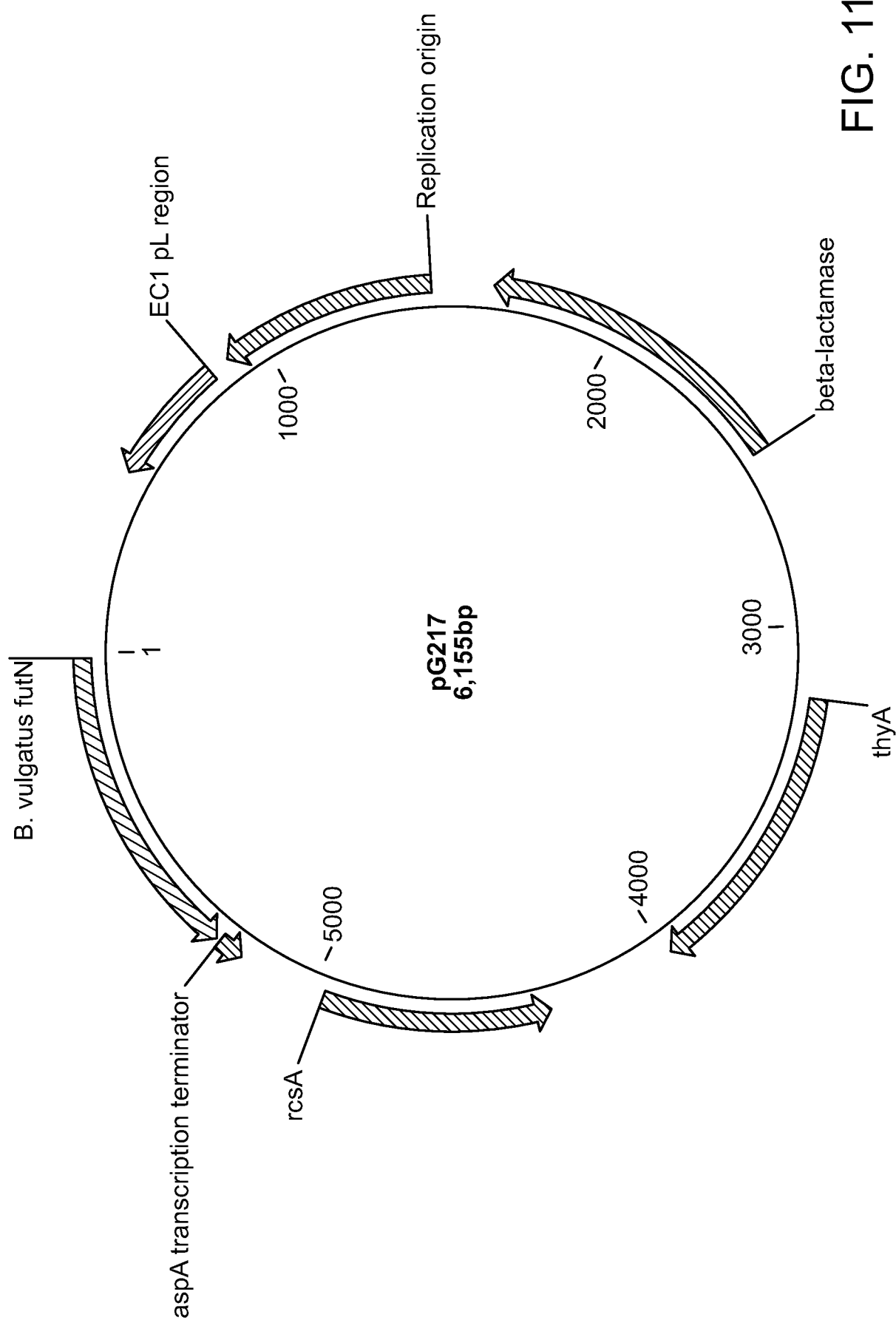


FIG. 11

FIG. 12

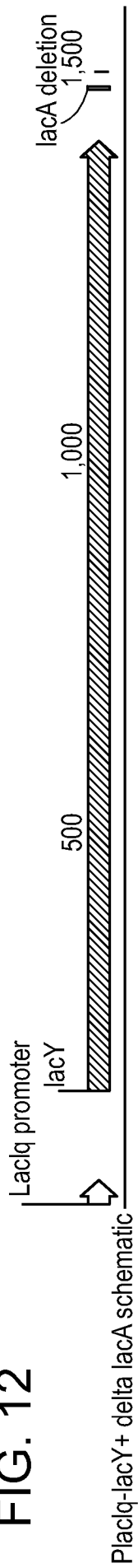
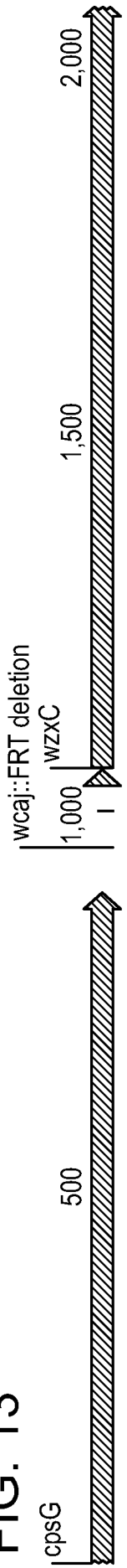
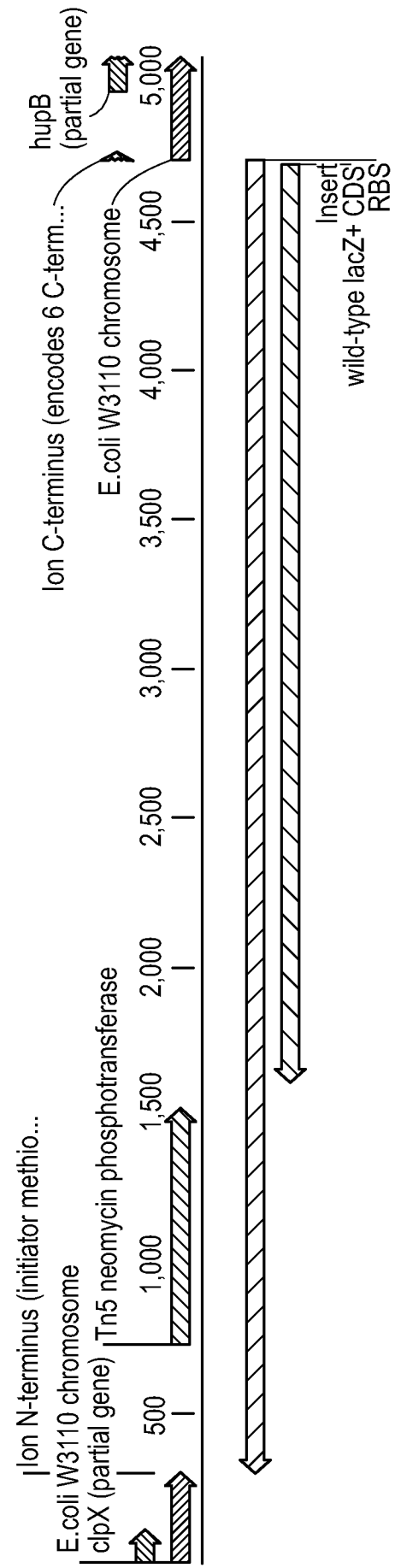


FIG. 13



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FIG. 14



INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 15/30823

A. CLASSIFICATION OF SUBJECT MATTER

IPC(8) - C12N 1/21, C12N 15/63, C12P 19/18 (2015.01)

CPC - C12N 15/63, C12N 9/1051, C12Y 204/01069

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC(8) - C12N 1/21, C12N 15/63, C12P 19/18 (2015.01)

CPC - C12N 15/63, C12N 9/1051, C12Y 204/01069

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

CPC - C12N 15/70, C12N 15/74, C12P 19/18

(keyword limited; terms below)

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

PatBase, PubWEST (USPT,PGPB,EPAB,JPAB), Google Scholar

Search terms: fucosyltransferase, bacteria, bacterium, E. coli, coli, lactose, vector, plasmid, FutC, FutN, FutX, FutQ, FutO, FutW, FutZA, FutS, FutP, FutU, FutX, FutR, FutV, FutY

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 2014/0031541 A1 (HEIDTMAN et al.) 30 January 2014 (30.01.2014) para [0003], [0004], [0012], [0016], [0017], [0079], SEQ ID NO: 8	1-3, 34-36, 40-42
A	US 2010/0120701 A1 (MCCOY et al.) 13 May 2010 (13.05.2010) para [0003], [0009], [0011], [0019], [0035], SEQ ID NO: 21	1, 34, 40

☐ Further documents are listed in the continuation of Box C.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search

11 August 2015 (11.08.2015)

Date of mailing of the international search report

03 SEP 2015

Name and mailing address of the ISA/US

Mail Stop PCT, Attn: ISA/US, Commissioner for Patents
P.O. Box 1450, Alexandria, Virginia 22313-1450

Facsimile No. 571-273-8300

Authorized officer:

Lee W. Young

PCT Helpdesk: 571-272-4300
PCT OSP: 571-272-7774

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 15/30823

Box No. I Nucleotide and/or amino acid sequence(s) (Continuation of item 1.c of the first sheet)

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of a sequence listing:
 - a. ☐ forming part of the international application as filed:
 - ☐ in the form of an Annex C/ST.25 text file.
 - ☐ on paper or in the form of an image file.
 - b. ☐ furnished together with the international application under PCT Rule 13ter.1(a) for the purposes of international search only in the form of an Annex C/ST.25 text file.
 - c. ☒ furnished subsequent to the international filing date for the purposes of international search only:
 - ☒ in the form of an Annex C/ST.25 text file (Rule 13ter.1(a)).
 - ☐ on paper or in the form of an image file (Rule 13ter.1(b) and Administrative Instructions, Section 713).
2. ☒ In addition, in the case that more than one version or copy of a sequence listing has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that forming part of the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments:

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 15/30823

Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
3. ☒ Claims Nos.: 4-33, 37-39, 43-47
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of additional fees.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.
- ☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.
- ☐ No protest accompanied the payment of additional search fees.