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GENE WHICH CODES FOR EICOSAPENTAENOIC ACID SYNTHETASE GROUP AND PROCESS FOR PRODUCING EICOSAPENTAENOIC ACID
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1. An isolated DNA encoding a group of eicosapentaenoic acid synthesis enzymes derived from a microorganism belonging to the genus *Shewanella*.



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(21) 国際出願番号 PCT/JP93/00641 (22) 国際出願日 1993年5月14日 (14. 05. 93) (30) 優先権データ 特願平4/147945 1992年5月15日 (15. 05. 92) JP (71) 出願人 (米国を除くすべての指定国について) 財団法人相模中央化学研究所 (SAGAMI CHEMICAL RESEARCH CENTER) [JP/JP] 〒100 東京都千代田区丸の内一丁目11番1号 Tokyo, (JP) (72) 発明者; および (75) 発明者/出願人 (米国についてのみ) 矢澤一良 (YAZAWA, Kazunaga) [JP/JP] 〒228 神奈川県相模原市鶴野森571 Kanagawa, (JP) 山田章子 (YAMADA, Akiko) [JP/JP] 〒229 神奈川県相模原市若松6-1-27 Kanagawa, (JP) 加藤誠志 (KATO, Seishi) [JP/JP] 〒228 神奈川県相模原市南台1-9-2 Kanagawa, (JP) 近藤 聖 (KONDO, Kiyosi) [JP/JP] 〒242 神奈川県大和市中央林間5-16-4 Kanagawa, (JP) (74) 代理人 弁理士 宇井正一, 外 (UI, Shoichi et al.) 〒105 東京都港区虎ノ門一丁目8番10号 静か堂門ビル 青和特許法律事務所 Tokyo, (JP)		(81) 指定国 AT (欧州特許), AU, BE (欧州特許), CA, CH (欧州特許), DE (欧州特許), DK (欧州特許), ES (欧州特許), FI, FR (欧州特許), GB (欧州特許), GR (欧州特許), IE (欧州特許), IT (欧州特許), LU (欧州特許), MC (欧州特許), NL (欧州特許), NO, PT (欧州特許), SE (欧州特許), US. 添付公開書類 国際調査報告書 873350
(54) Title : GENE WHICH CODES FOR EICOSAPENTAENOIC ACID SYNTHETASE GROUP AND PROCESS FOR PRODUCING EICOSAPENTAENOIC ACID (54) 発明の名称 イコサペンタエン酸合成酵素群をコードする遺伝子及びイコサペンタエン酸の製造方法 (57) Abstract An advantageous process for producing eicosapentaenoic acid (EPA) useful as medicine, pesticide, food, feed and the like by the genetic recombination process by taking a gene coding for an EPA biosynthetase group from a microorganism. EPA is produced by taking a gene coding for an EPA biosynthetase group, preparing a plasmid by linking the gene with a vector, transforming <i>Escherichia coli</i> with the plasmid, and culturing the transformed <i>Escherichia coli</i>.		

(57) 要約

医薬・農薬・食品・飼料等として有用なイコサペンタエン酸（EPA）の生合成酵素群をコードする遺伝子を微生物から取得し、遺伝子組み換え法により、EPAの有利な製造法を提供する。

イコサペンタエン酸（EPA）生合成酵素群をコードする遺伝子を取得し、これをベクターと連結してプラスミドを作成して該プラスミドにて大腸菌を形質転換し、この形質転換した大腸菌を培養してEPAを産生する。

情報としての用途のみ

PCTに基づいて公開される国際出願のパンフレット第1頁にPCT加盟国を同定するために使用されるコード

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DESCRIPTION

Gene Coding for Eicosapentaenoic Acid Synthesizing
Enzymes and Process for Production of

5 Eicosapentaenoic Acid

FIELD OF THE ART

The present invention relates to genes coding for
eicosapentaenoic acid (designated EPA hereinafter)
synthesizing enzymes, plasmids containing these genes,
10 microorganisms transformed with the plasmid, and a
process for production of eicosapentaenoic acid using the
microorganism. The EPA is useful as a starting material
for pharmaceuticals, foods, feeds and the like.

BACKGROUND ART

15 Polyunsaturated fatty acids represented by
eicosapentaenoic acid (EPA) play an important roll as a
component of a biomembrane. So far, the following
pharmacological actions of EPA are known. (1) Platelet
coagulation inhibitory action (thrombolytic action), (2)
20 blood neutral fat-lowering action, (3) actions for
lowering blood VLDL-cholesterol and LDL-cholesterol and
increasing HDL-cholesterol (anti-arterial sclerosis
action), (4) blood viscosity-lowering action, (5) blood
pressure lowering action, (7) anti-inflammatory action,
25 (8) anti-tumor action.

In addition, EPA is a substrate for biosynthesis of
prostaglandins and exhibits an essential function in vivo
in higher mammals including humans. In particular EPA is
important as a substrate for production of three types of
30 prostaglandins, has platelet coagulation inhibitory
action and is studied for applications to treatment and
prophylactic agents for thrombosis. In addition EPA has
especially high activity for lowering plasma cholesterol
level among polyunsaturated fatty acids having said
35 action, and is highly effective in comparison with
linoleic acid and the like usually contained in plant
oil. Additionally, EPA is known as an essential nutrient



for fish.

Thus, epidemiological research by Dyerberg, Denmark (Am. J. Clin. Nutr., 28, 959, 1975) showed a possibility for use as health foods or pharmaceuticals on the basis of thrombosis inhibitory action or lipid-lowering actions of EPA. However, as can be seen from its chemical structure, the chemical synthesis of EPA is very difficult. Accordingly, in Japan, it is recommended to eat blue back fish such as sardine, salmon, saury, and the like.

At present, most commercially available EPA products are fractionation products from fish oil obtained by a boiling process, and the EPA content thereof is about 10 to 30%. Fish oil extracted by a boiling process is a mixed glyceride containing various kind of fatty acids as component fatty acids, and not only is isolation and purification of each component difficult, but also since EPA is a polyunsaturated fatty acid having 20 carbon atoms and five double bonds all of which are cis-type, EPA is also a unstable and highly oxidation-labile fatty acid. Therefore, it is necessary for EPA to be concentrated from fish oil with prevention of oxygen, light, heat and the like. In addition, since fish oil contains various fatty acids in addition to EPA, their fractionation is difficult. Moreover, although various organic solvents used for fractionation of EPA are eliminated under a reduced pressure, complete elimination of the organic solvents is difficult from a technical and economical point of view.

Most EPA preparations used for pharmaceuticals are those having at least 90% EPA concentration, produced by extracting fish oil by various processes, hydrolysing the fish oil enzymatically or under an alkaline condition to generate free fatty acids, optionally converting the free fatty acids to corresponding methyl or ethyl esters, and further purifying by fractional crystallization at a low temperature, applying a urea-addition method,



distillation under a reduced pressure, reverse phase chromatography, or the like. However, since these processes use many organic solvents and heating to near 200°C, it is possible that an EPA concentrate obtained by using such process may be denaturated by residual organic solvent, and polymerization, isomerization or oxidation of EPA. Moreover, where a fish oil is used as a starting material for production of EPA, it is difficult to eliminate docosenoic acid or the like which is considered to be a cause of cardioidiseases, and therefore problems remain in the use for health foods, pharmaceuticals or the like.

On the other hand, recently, processes for production of EPA using microorganisms such as chlorella, unicellular algae Monodus, Eugrena or Diatomaceae have been studied in place of extraction methods from fish oil having drawbacks such as residual fish odor due to incomplete purification and concentration, and the production of EPA using microorganisms has been considered. Recently, fungi producing EPA were reported by Gellerman and Schlenk (J. L. Gellerman and H. Schlenk, BBA, 573, 23, 1979) and Yamada et al. (Meeting of The society of Fermentation Technology, Japan, 1986).

The present inventors sought marine bacteria having an ability to produce EPA to find a new fermentation process for production of EPA using bacteria from which genes can be easily obtained, and which can be cultured in a short time and be easily controlled, and as a result, the present inventors found a new bacteria belonging to the genus Pseudomonas, Alteromonas or Shewanella (K. Yazawa et al., J. Biochem., 103, 5 (1988); K. Yazawa et al., Nippon Suisan Gakkai shi, 54, 1835 (1988)).

It has been suggested that biosynthesis of polyunsaturated fatty acids including EPA works by site-specific aerobic unsaturation of corresponding saturated fatty acids (for example, R. Jeffcoat and A. T. James



(1984) in: S. Numa (Ed.): Fatty acid metabolism and its regulation, Elsevier, Amsterdam, pp85 - 112). However, there is no report relating to biosynthetic enzymes which participate in EPA synthesis, and gene coding therefor.

5 DISCLOSURE OF THE INVENTION

Generally, the ability of a wild strain to produce a useful substance is low, and therefore where it is intended that the ability of the microorganism is industrially used, an improvement, i.e., an increase of productivity of the microorganism is carried out by various methods. The present inventors intended to carry out research for increasing an EPA productivity by finding genes not described in literature for EPA biosynthetic enzymes using gene recombination techniques and introducing the same into another organism, to impart an EPA biosynthesis ability to an organism not having an EPA biosynthesis ability, and eventually to establish an advantageous process for production of EPA.

Accordingly, the present invention provides genes for EPA biosynthetic enzymes, expression plasmids containing said genes, organisms transformed with said plasmid, and a process for production of EPA using said organism.

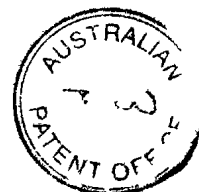
BRIEF EXPLANATION OF THE DRAWINGS

25 Figure 1 represents the structure of the plasmid pEPA containing a group of the present genes.

Fig. 2 represents a restriction enzyme map of a DNA fragment containing a group of the present genes.

Best Mode for Carrying Out the Invention

30 According to the present invention, an EPA producing strain is constructed by extracting DNA from a microorganism having an ability to produce EPA (microbial origin of genes), cutting this DNA with restriction enzymes to excise genes coded for a group of EPA biosynthesis enzymes, introducing the genes into an appropriate vector to construct an expression plasmid, and transforming a host organism with the plasmid to



construct an EPA producing strain. EPA can be produced by this organism.

Gene source

Although organisms which can be used as a gene source according to the present invention are not limited to specific genera, species or strains, usually, microorganisms classified to the genus Pseudomonas, Alteromonas, Shewanella or the like can be used. These microorganisms can be easily obtained from official or public depository institutes for microorganisms.

As examples of microorganisms belonging to the genus Pseudomonas, Pseudomonas putrefaciens SCRC-2181 (FERM BP-2917), SCRC-2201 (FERM BP-2916), SCRC-2271 (FERM BP-2195), SCRC-2341 (FERM BP-2918), SCRC-2451 (FERM BP-2919), SCRC-2642 (FERM BP-2920), SCRC-2792 (FERM BP-2921), SCRC-2878 (FERM BP-1623), SCRC-3011 (FERM BP-2913), and SCRC-3022 (FERM BP-2914) may be mentioned.

As example of microorganisms belonging to the genus Alteromonas, Alteromonas putrefaciens SCRC-2871 (FERM BP-1624) and Alteromonas putrefaciens subspecies sagamifaciens SCRC-1162 (FERM BP-1626) may be mentioned.

As an example of microorganism belonging to the genus Shewanella, Shewanella putrefaciens SCRC-2874 (FERM BP-1625) may be mentioned.

Cloning of Genes Coding for a Group of EPA Biosynthesis Enzymes and Construction of Expression Plasmid

In the present invention, the case wherein Shewanella putrefaciens SCRC-2874 (FERM BP-1625) was used as a source of genes for a group of EPA biosynthesis enzymes is concretely explained. However, as described above, various EPA producing microorganisms can be similarly used as a gene source. A process for cloning genes is described in the Examples of the present invention.

According to the present invention, EPA producing strains can be artificially generated by transforming an



heterogeneous host such as Escherichia coli or a homogeneous host such as Shewanella, or further yeast, fungus or the like. In addition, EPA producing plants can be generated by introducing the present genes into a higher plant such as soybean, sunflower, rape, or the like. As a region for expression of a group of EPA biosynthesis genes, although a control region natively accompanying these enzymes can be used, it is advantageous to prepare another promoter/operator system for increasing an amount of expression or allowing inducible expression. Where E. coli is used as a host, as the promoter/operator system, trp, tac, lavUV5, P_L, P_R or lpp promoter/operator system and the like can be used, and as an SD sequence, an SD sequence of trp leader peptide, lacZ, metapyrocatechase or cII gene can be used. In addition, a transcriptional terminator, for example, rrnBT₁T₂ terminator of E. coli ribosome gene or the like can be provided downstream of a coding region. In addition, for expression of the above-mentioned gene, a host/vector system of Saccharomyces cerevisiae can be used, wherein as a promoter there can be used a promoter of alcohol dehydrogenase gene, a promoter of acid phosphatase gene, a promoter of glyceraldehyde-3-phosphate dehydrogenase gene, a promoter of enolase gene or the like can be used, wherein a plasmid preferably contains a sequence for replication in yeast, and an auxotrophic marker as a selectable maker for selection of yeast containing said plasmid, such as Leu, Trp, His or the like.

For introduction of the genes into a plant, there are a method using a vector, and a direct introduction method. As vectors, Ti plasmid; DNA viruses such as cauliflower mosaic virus (CaMV), Geminivirus, cassava roten virus, tomato golden mosaic virus, and the like; and RNA viruses such as brome mosaic virus (BMV), and tobacco mosaic virus (TMV) can be used, wherein as a



promoter, 35S promoter of CaMV or the like may be mentioned. On the other hand, as direct protoplast introduction methods there can be mentioned a calcium phosphate method, polyethylene glycol method,
5 microinjection, electroporation, liposome method and the like. Moreover, as a direct plant cell introduction method, there may be mentioned a particle gun method.

In addition, a fatty acid composition in a host plant can be changed by using a part of the group of the
10 genes.

Note, that generally, an amount of expression of a particular protein in E. coli is affected by the number of copies of the genes, an efficiency of transcription, stability of mRNA, efficiency of translation, and
15 stability of the protein. To modify control regions such as a promoter, SD region, terminator and the like, a smaller plasmid can be easily treated. The number of copies of the genes depends on the size of the plasmid, and there is a tendency for the plasmid to be smaller as
20 the number of copies increases. For this purpose, a smaller plasmid can be obtained by inserting a DNA fragment containing a group of genes for EPA biosynthesis described in the Examples of the present invention into a plasmid, and repeating subcloning of the plasmid to cut
25 off unnecessary portions of the gene DNA fragment. Smaller EPA biosynthesis enzyme genes thus obtained are included in the present invention. In addition, to enhance stability or activity of the enzyme, the nucleotide sequence of the gene (amino acid sequence) can
30 be modified by a known technique, and the present invention includes such a modified gene.

As a host E. coli of the present invention, any strain derived from E. coli K12 may be used. For example, JM83, JM101, JM103, JM105, JM109, RR1, RB791,
35 W3110, C600, HB101, DH1, AG1, NM554 or the like can be used. As a yeast host, AH22, DC5, D-13-1A, YNN144 or the like can be mentioned.



Note, that a group of the enzymes encoded by the present genes can convert higher fatty acids synthesized by a biosynthetic system natively possessed by the host organism to eicosapentaenoic acid.

5 In practicing the present invention, an organism such as a microorganism transformed with the present genes is cultured in a medium according to a conventional procedure to obtain microbial cells. In this case, for example, a medium having a composition shown in Table 1
10 is prepared.

Table 1

	Yeast extract	0.5%
	Pepton	1.0%
15	Sea water	1/2 concentration
	pH 7.0	

From the microbial cells thus obtained, EPA can be obtained according to a conventional procedure such as
20 extraction with an organic solvent. The details are described in the following Examples.

EXAMPLES

Next, the present invention is explained in more detail by way of Examples.

25 Example 1-1 Preparation of Genomic DNA Containing
Genes Coding for a Group of PEA
Biosynthesis Enzymes

Shewanella putrefaciens SCRC-2874 (FERM BP-1625) was inoculated in 125 ml of a medium (1% pepton, 0.5% yeast
30 extract, 1/2 concentration artificial sea water), and cultured at 15°C for 18 hours with shaking (OD610 = 8.6). The resulting microbial cells were washed once with 1M NaCl, and suspended in 20 ml of 1M NaCl. The suspension was allowed to stand at 55°C for 30 minutes, 20 ml of
35 0.1M EDTA was added thereto, and after being allowed to stand at 55°C for 15 minutes, the suspension was centrifuged at 10,000 rpm for 10 minutes. To the



precipitate, was added 10 ml of TES buffer (1 mM EDTA, 0.1 mM NaCl, 10 mM Tris-HCl, pH 8.0) containing 100 mg of lysozyme, and the cells were suspended. After being allowed to stand at 37°C for an hour, 1 ml of 10% SDS was added to the suspension, which was then allowed to stand at 60°C for an hour. 11 ml of neutralized phenol was added to the suspension, which was then gently shaken for 5 minutes and centrifuged at 6,500 rpm for 5 minutes to obtain the upper layer. 20 ml of ethanol was added to the layer and the mixture was gently shaken. Precipitated DNA was wound onto a glass bar, washed in ethanol, dissolved in 10 ml of TES buffer, and the solution was allowed to stand at 4°C overnight. 0.5 mg of RNase A was added to the solution, which was then gently shaken at 37°C for 3 hours, and after 1 mg of proteinase K was added thereto, the solution was further shaken for 4.5 hours; 5 ml each of neutralized phenol and chloroform were gradually added to the mixture, which was then gently shaken for 5 minutes and centrifuged to recover the upper layer. 10 ml of chloroform was added to the layer, which was then gently shaken and centrifuged to obtain the upper layer. 20 ml of ethanol was added to the layer, which was then gently shaken, and precipitated DNA was wound onto a glass bar. The DNA was washed in ethanol, and dissolved in 3 ml of TES buffer. An amount of DNA thus obtained was about 2.8 mg. Next, 200 µg of the DNA was partially digested with restriction enzyme Sau3A1 and subjected to electrophoresis on 0.3% agarose, and DNA fragments larger than about 20 Kb were isolated by electroelution. The DNA fragments were extracted with phenol/chloroform, and precipitated with ethanol, and the precipitate was dissolved in 500 µl of TE buffer (1 mM EDTA, 10 mM Tris-HCl, pH 7.4).

Example 1-2 Insertion of Chromosomal DNA

Fragments into Vector

As a vector, cosmid pWE15 (STRATAGENE) was used. 10 µg of pWE15 was completely digested with restriction



enzyme BamHI, treated with calf intestine alkaline phosphatase at 37°C for an hour, extracted with phenol/chloroform, and precipitated with ethanol, and the precipitate was dissolved in 10 µl of TE buffer. 1.5 µg of the vector DNA thus obtained was mixed with 1 µg of the chromosomal DNA prepared in Example 1 and partially digested with restriction enzyme Sau3A1, and these DNA were ligated using T4 DNA ligase at 26°C for 10 minutes. One fourth of the reaction mixture was packaged according to a conventional method to form phage, which was then infected to E. coli K12/AG-1.

Example 1-3 Screening of Recombinant EPA
Producing Strain

The E. coli suspension infected with the phage of Example 1-2 was plated on an LB agar medium (trypton 1%, yeast extract 0.5%, NaCl 1%, Agar 2%) containing 50 µg/ml ampicillin and cultured at 37°C overnight. The developed colony was inoculated in 1.5 ml of LB medium containing 50 µg/ml ampicillin, and cultured at 25°C for 1 to 7 days with shaking. The culture was centrifuged to collect the microbial cells, and after removing the medium, the cells were suspended in 0.5 ml of methanol saturated with hydrogen chloride. The cell suspension was sealed and incubated at 80°C for an hour to methyl-esterify fatty acids. After allowing the suspension to cool, it was extracted three times with 0.3 ml hexane, the hexane layer was dried and the residue was dissolved in 20 µl of methanol. 2 µl of the solution was spotted on a silica gel plate which was then developed three times with a developing solvent of hexane and ether 19:1, dried in air, and colored with iodine.

In this way, as a result of tests of about 390 recombinant clones, one clone showing a thin layer chromatography spot at the same position as a standard methyl ester of EPA was obtained. From the clone, cosmid was extracted using an alkali/SDS method. This cosmid was designated as pEPA. The pEPA is a cosmid wherein a



San3A1 fragment of about 37 Kbp was inserted into BamHI site of pWE15.

Example 1-4 Preparation of Restriction Enzyme Map of pEPA

Cosmid pEPA was prepared from transformant AG-1/pEPA. The pEPA was cleaved with various restriction enzymes, and a restriction enzyme map was prepared (Fig. 1).

Example 1-5 Analysis of Sequence

An entire nucleotide sequence of a Sau3A1-Sau3A1 fragment containing a genomic DNA insert in the cosmid pEPA is shown in SEQ ID NO: 1. In the nucleotide sequence, 9 open reading frames, ORFs 2 to 10, can be identified, and these nucleotide sequences and corresponding amino acid sequences are shown in SEQ ID NOs: 2 to 10 respectively. The relationship between the entire nucleotide sequence (SEQ ID NO: 1) and ORFs 2 to 10 (SEQ ID NOs: 2 to 10, respectively) is shown in Table 2.

Table 2

SEQ ID NO	Length of Sequence	Positions on SEQ ID NO: 1
2	1983	6121-8103
3	831	8186-9016*
4	2910	9681-12590
5	864	13040-13903
6	8268	13906-22173
7	2340	22176-24515
8	6012	24518-30529
9	1629	30730-32358
10	1575	32753-34327

* Reversed sequence extending from the position No. 9016 to the position No. 8186 in the nucleotide sequence shown in SEQ ID NO: 1.

A restriction enzyme map of the above-mentioned genomic DNA insert and positions of the ORFs 2 to 10 on the map are shown in Fig. 2. In Fig. 2, the symbols Δ show the sites at which mRNA forms a hair pin structure (stem and roop structure). The pEPA was cleaved with different restriction enzymes to remove a



part of the sequence of SEQ ID NO: 1, and the cleaved pEPA was religated, and used to transform *E. coli*. Next, the transformants were tested for the productivity of eicosapentaenoic acid. As a result, in the case where the parts shown by hatching in Fig. 2 were deleted, the production of eicosapentaenoic acid was not observed. Accordingly, it is considered that for the production of eicosapentaenoic acid in *E. coli*, at least one of ORFs 2, 3 and 4, and at least one of ORFs 9 and 10 is essential.

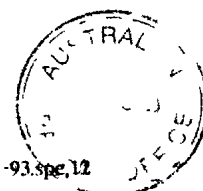
By comparison of the amino acid sequences of various ORFs with known amino acid sequences, it was found that five regions in the ORF 6, and 2 regions in the ORF 8 have homology to some extent with amino acid sequences of enzymes participating in fatty acid synthesis. The results are shown in Table 3.

Table 3

SEQ ID NO (ORF No.)	Position of amino acid sequence	Similar enzyme and position	References
6	668(Leu)-930(Leu)	MalonylCoA-ACP transferase 56(Leu)-309(Leu)	(1)
6	189(Phe)-424(His)	Fatty acid synthetase 120(Phe)-350(His)	(2)
6	200(Ser)-483(Leu)	Fatty acid synthetase (3-ketoacyl-ACP synthetase domain) 137(Ala)-406(Asp)	(3)
6	204(Ser)-488(Gln)	3-Ketoacyl-ACP synthetase 137(Ala)-406(Asp)	(4)
6	2261(Phe)-2392(Gly)	2-Oxoacylreductase 1470(Leu)-1604(Gly)	(5)
8	205(Ala)-442(Lys)	3-Ketoacyl-ACP synthetase 187(Ala)-416(Asn)	(6)
8	1373(Thr)-1547(Val)	3-Hydroxydecanoyl-ACP dehydratase 29(Leu)-163(Val)	(7)

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10 Accordingly, the present invention provides genes coding for amino acid sequences shown in SEQ ID NOs 2 to 10, and further provides DNAs coding for amino acid sequence region having a homology with amino acid sequences of known enzymes or amino acid sequences containing the same. The present invention further includes nucleotide sequences which can hybridize with one of the
15 nucleotide sequences shown in SEQ ID NOs: 1 to 10 or a part thereof and coding for a useful activity, and polypeptides encoded by one of these nucleotide sequences and having a useful activity.

Example 2 Production of EPA by Transformant GA-1/pEPA

20 Transformant AG-1/pEPA was inoculated in 100 ml of LB medium containing 50 µl/ml ampicillin, and cultured at 25 °C for 48 hours. The cells were obtained by centrifugation, washed once and suspended in 2 ml of pure water, and the suspension was extracted three times with 12 ml of a solvent composed of chloroform and methanol 2:1. The solvent layer was dried, and the residue was dissolved in 1.5 ml of methanol saturated with hydrogen chloride, and the solution
25 was sealed and incubated at 80 °C for an hour to methyl-esterify fatty acids. After allowing the solution to cool, it was extracted three times with 2 ml of hexane, and after the hexane layer was dried, the residue was dissolved in 20 µl of methanol. A part of the solution was analyzed by gas chromatography. As a result, a peak of EPA was observed, and a ratio of



EPA relating to total fatty acid esters was calculated as about 1.36% from the area of peaks. The amount of EPA per culture volume was about 0.5 mg/l. The ester mixture thus obtained was spotted on a silver nitrate silica gel plate, which was then developed by a solvent composed of hexane and ether at a ratio of 3:1. The plate was colored with fluorescein and ultraviolet light, the spot of the ester of the polyunsaturated fatty acid was scraped off, 1.8 ml of methanol and 0.2 ml of 10% NaCl were added thereto, and the mixture was shaken at room temperature for 30 minutes. The mixture was extracted three times with 2 ml of hexane, the hexane layer was dried, the residue was dissolved in 40 μ l of hexane, and GC-MS analysis was carried out. As a result, molecular weight of the substance of the desired peak on the chromatography was 316, and the peak of the fragment conformed to that of an authentic sample, and the substance was identified as EPA. Note that MS fragment peaks were as follow:

Mass: 316(M^+), 287, 273, 262, 247, 234, 220, 201, 180, 161, 148, 133, 119, 108, 93, 79, 67, 55, 41, 28.

Example 3 Production of EPA by Transformant JM109/pEPA

According to a conventional procedure the cosmid pEPA was used to transform E. coli K12/JM109. JM109/pEPA (FERM BP-4257) was obtained by selection using an LB agar medium containing 50 μ l/ml ampicillin. According to the same procedure as described in Example 2, extraction of lipid from the cells, methyl-esterification and analysis by gas chromatography were carried out and a peak of EPA was detected. A ratio of EPA relating to a total of esters of fatty acids was calculated as about 1.43% on the basis of the area of peaks. An amount of EPA per culture volume was about 0.6 mg/l.

Reference to deposited microorganisms and the



depository authority under the Rule 13-2

Depository authority: National Institute of
Bioscience and Human-Technology Agency of Industrial
Science and Technology

5 Deposition Number and Deposition Date:
 May 14, 1992 FERM BP-4257



SEQ ID NO : 1

SEQUENCE LENGTH : 37895

SEQUENCE TYPE : Nucleic acid

STRANDNESS : Double strand

TOPOLOGY : Linear

MOLECULE TYPE : Genomic DNA

ORIGINAL SOURCE : *Shewanella putrefaciens* SCRC-2874

(FERM BP-1625)

SEQUENCE

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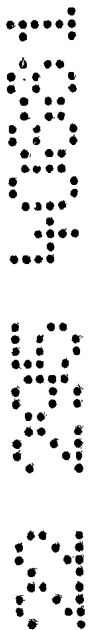
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SEQ ID NO : 2

SEQUENCE LENGTH : 1983

SEQUENCE TYPE : Nucleic acid

STRANDNESS : Double strand

TOPOLOGY : Linear

MOLECULE TYPE : Genomic DNA

ORIGINAL SOURCE : Shewanella putrefaciens SCRC-2874

(FERM BP-1625)

SEQUENCE

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Asn Ala Leu Ala Ala Gln His Glu His Asp His Ile Thr Val Asp Tyr	
20 25 30	
GAA GGG AAA GCC GCA ACA GAA CAC ACC ATA GCT CAC AAC CAA GCT GTA	144
Glu Gly Lys Ala Ala Thr Glu His Thr Ile Ala His Asn Gln Ala Val	
35 40 45	
GCT AAA ACA CTT AAC TTT GCC GAC ACG CGT GCA TTT GAG CAA TCG TCT	192
Ala Lys Thr Leu Asn Phe Ala Asp Thr Arg Ala Phe Glu Gln Ser Ser	
50 55 60	
AAA AAT CTA GTC GCC AAG TTT GAT AAA GCA ACT GCC GAT ATA TTA CGT	240
Lys Asn Leu Val Ala Lys Phe Asp Lys Ala Thr Ala Asp Ile Leu Arg	
65 70 75 80	



GCC GAA TTT GCT TTT ATT AGC GAT GAA ATC CCT GAC TCG GTT AAC CCG	288
Ala Glu Phe Ala Phe Ile Ser Asp Glu Ile Pro Asp Ser Val Asn Pro	
85 90 95	
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Ser Leu Tyr Arg Gln Ala Gln Leu Asn Met Val Pro Asn Gly Leu Tyr	
100 105 110	
AAA GTG AGC GAT GGC ATT TAC CAG GTC CGC GGT ACC GAC TTA TCT AAC	384
Lys Val Ser Asp Gly Ile Tyr Gln Val Arg Gly Thr Asp Leu Ser Asn	
115 120 125	
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Leu Thr Leu Ile Arg Ser Asp Asn Gly Trp Ile Ala Tyr Asp Val Leu	
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145 150 155 160	
CTA CCT AAA GAT GGC GAT TTA CCC GTT GTT GCG ATG ATT TAC TCC CAT	528
Leu Pro Lys Asp Gly Asp Leu Pro Val Val Ala Met Ile Tyr Ser His	
165 170 175	
AGC CAT GCG GAC CAC TTT GGC GGA GCT CGC GGT GTT CAA GAG ATG TTC	576
Ser His Ala Asp His Phe Gly Gly Ala Arg Gly Val Gln Glu Met Phe	
180 185 190	
CCT GAT GTC AAA GTC TAC GGC TCA GAT AAC ATC ACT AAA GAA ATT GTC	624
Pro Asp Val Lys Val Tyr Gly Ser Asp Asn Ile Thr Lys Glu Ile Val	
195 200 205	
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Asp Glu Asn Val Leu Ala Gly Asn Ala Met Ser Arg Arg Ala Ala Tyr	
210 215 220	
CAA TAC GGC GCA ACA CTG GGC AAA CAT GAC CAC GGT ATT GTT GAT GCT	720
Gln Tyr Gly Ala Thr Leu Gly Lys His Asp His Gly Ile Val Asp Ala	
225 230 235 240	
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Ala Leu Gly Lys Gly Leu Ser Lys Gly Glu Ile Thr Tyr Val Ala Pro	
245 250 255	



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Asp Tyr Thr Leu Asn Ser Glu Gly Lys Trp Glu Thr Leu Thr Ile Asp	
260 265 270	
GGT CTA GAG ATG GTG TTT ATG GAT GCC TCG GGC ACC GAA GCT GAG TCA	864
Gly Leu Glu Met Val Phe Met Asp Ala Ser Gly Thr Glu Ala Glu Ser	
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GAA ATG ATC ACT TAT ATT CCC TCT AAA AAA GCG CTC TGG ACG GCG GAG	912
Glu Met Ile Thr Tyr Ile Pro Ser Lys Lys Ala Leu Trp Thr Ala Glu	
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Val Arg Asp Ala Leu Lys Trp Ser Lys Asp Ile Asn Glu Met Ile Asn	
325 330 335	
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Val Trp Gly Asn Gln Ala Ile Asn Asp Phe Leu Arg Leu Gln Arg Asp	
355 360 365	
AAC TAC GGC CTA GTG CAC AAT CAA ACC TTG AGA CTT GCC AAC GAT GGT	1152
Asn Tyr Gly Leu Val His Asn Gln Thr Leu Arg Leu Ala Asn Asp Gly	
370 375 380	
GTC GGT ATA CAA GAT ATT GGC GAT GCG ATT CAA GAC ACG ATT CCA GAG	1200
Val Gly Ile Gln Asp Ile Gly Asp Ala Ile Gln Asp Thr Ile Pro Glu	
385 390 395 400	
TCT ATC TAC AAG ACG TGG CAT ACC AAT GGT TAC CAC GGC ACT TAT AGC	1248
Ser Ile Tyr Lys Thr Trp His Thr Asn Gly Tyr His Gly Thr Tyr Ser	
405 410 415	
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His Asn Ala Lys Ala Val Tyr Asn Lys Tyr Leu Gly Tyr Phe Asp Met	
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AAC CCA GCC AAC CTT AAT CCG CTG CCA ACC AAG CAA GAA TCT GCC AAG	1344
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435 440 445	
TTT GTC GAA TAC ATG GGC GGC GCA GAT GCC GCA ATT AAG CGC GCT AAA	1392
Phe Val Glu Tyr Met Gly Gly Ala Asp Ala Ala Ile Lys Arg Ala Lys	
450 455 460	
GAT GAT TAC GCT CAA GGT GAA TAC CGC TTT GTT GCA ACG GCA TTA AAT	1440
Asp Asp Tyr Ala Gln Gly Glu Tyr Arg Phe Val Ala Thr Ala Leu Asn	
465 470 475 480	
AAG GTG GTG ATG GCC GAG CCA GAA AAT GAC TCC GCT CGT CAA TTG CTA	1488
Lys Val Val Met Ala Glu Pro Glu Asn Asp Ser Ala Arg Gln Leu Leu	
485 490 495	
GCC GAT ACC TAT GAG CAA CTT GGT TAT CAA GCA GAA GGG GCT GGC TGG	1536
Ala Asp Thr Tyr Glu Gln Leu Gly Tyr Gln Ala Glu Gly Ala Gly Trp	
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Arg Asn Ile Tyr Leu Thr Gly Ala Gln Glu Leu Arg Val Gly Ile Gln	
515 520 525	
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Ala Gly Ala Pro Lys Thr Ala Ser Ala Asp Val Ile Ser Glu Met Asp	
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ATG CCG ACT CTA TTT GAC TTC CTC GCG GTG AAG ATT GAT AGT CAA CAG	1680
Met Pro Thr Leu Phe Asp Phe Leu Ala Val Lys Ile Asp Ser Gln Gln	
545 550 555 560	
GCG GCT AAG CAC GGC TTA GTT AAG ATG AAT GTT ATC ACC CCT GAT ACT	1728
Ala Ala Lys His Gly Leu Val Lys Met Asn Val Ile Thr Pro Asp Thr	
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Lys Asp Ile Leu Tyr Ile Glu Leu Ser Asn Gly Asn Leu Ser Asn Ala	
580 585 590	
GTG GTC GAC AAA GAG CAA GCA GCT GAC GCA AAC CTT ATG GTT AAT AAA	1824
Val Val Asp Lys Glu Gln Ala Ala Asp Ala Asn Leu Met Val Asn Lys	
595 600 605	



GCT GAC GTT AAC CGC ATC TTA CTT GGC CAA GTA ACC CTA AAA GCG TTA 1872
 Ala Asp Val Asn Arg Ile Leu Leu Gly Gln Val Thr Leu Lys Ala Leu
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 Leu Ala Ser Gly Asp Ala Lys Leu Thr Gly Asp Lys Thr Ala Phe Ser
 625 630 635 640
 AAA ATA GCC GAT AGC ATG GTC GAG TTT ACA CCT GAC TTC GAA ATC GTA 1968
 Lys Ile Ala Asp Ser Met Val Glu Phe Thr Pro Asp Phe Glu Ile Val
 645 650 655
 CCA ACG CCT GTT AAA 1983
 Pro Thr Pro Val Lys
 660

SEQ ID NO : 3

SEQUENCE LENGTH : 831

SEQUENCE TYPE : Nucleic acid

STRANDNESS : Double strand

TOPOLOGY : Linear

MOLECULE TYPE : Genomic DNA

ORIGINAL SOURCE : Shewanella putrefaciens SCRC-2874
 (FERM BP-1625)

SEQUENCE

ATG GTA AGA GGC TAT TTG CGC GCT TTA TTG TCA CAA CAT AGT GAA ATA 48
 Met Val Arg Gly Tyr Leu Arg Ala Leu Leu Ser Gln His Ser Glu Ile
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 Arg Pro Asn Glu Trp Arg Phe Glu Tyr Gly Asp Lys Gly Lys Pro Arg
 20 25 30
 TTG AGT GAT GCG CAA TTT GCT CAA ACC GGG GTC CAC TTT AAT GTG AGT 144
 Leu Ser Asp Ala Gln Phe Ala Gln Thr Gly Val His Phe Asn Val Ser
 35 40 45
 CAT AGT GGA GAT TGG CTA TTA GTA GGC ATT TGC ACT GCT GAT AAT AAA 192
 His Ser Gly Asp Trp Leu Leu Val Gly Ile Cys Thr Ala Asp Asn Lys
 50 55 60



GGC GCC AGT CAG GCA AGC AAG GAG GAA ACT GAC TCT GCT AGT ATT GAG	240
Gly Ala Ser Gln Ala Ser Lys Glu Glu Thr Asp Ser Ala Ser Ile Glu	
65 70 75 80	
TTT GGC GTC GAC ATT GAG CGT TGC CGT AAC AGC ACC AAT ATC CAC TCT	288
Phe Gly Val Asp Ile Glu Arg Cys Arg Asn Ser Thr Asn Ile His Ser	
85 90 95	
ATT CTT AGT CAT TAT TTC TCT GAA TCA GAA AAG CGA GCC TTG TTA GCG	336
Ile Leu Ser His Tyr Phe Ser Glu Ser Glu Lys Arg Ala Leu Leu Ala	
100 105 110	
TTA CCA GAG GCC TTG CAG CGA GAC CGC TTT TTT GAT TTG TGG GCG CTC	384
Leu Pro Glu Ala Leu Gln Arg Asp Arg Phe Phe Asp Leu Trp Ala Leu	
115 120 125	
AAG GAG TCT TAC ATT AAA GCG AAA GGA CTT GGG CTG GCA TTA TCG CTA	432
Lys Glu Ser Tyr Ile Lys Ala Lys Gly Leu Gly Leu Ala Leu Ser Leu	
130 135 140	
AAA TCT TTT GCG TTT GAC TTC TCT GCA CTG AGC GAA ACT TTT CTT GGA	480
Lys Ser Phe Ala Phe Asp Phe Ser Ala Leu Ser Glu Thr Phe Leu Gly	
145 150 155 160	
GTT AAT GCA CCT AAA AGC TTG AGC CAT TGT GTT GAT ATT TCC GAT GCT	528
Val Asn Ala Pro Lys Ser Leu Ser His Cys Val Asp Ile Ser Asp Ala	
165 170 175	
ATT GCG GAT CAC AAG GTT GAG CAT CAA CTT AAT CAG CGA CAG GTT TTG	576
Ile Ala Asp His Lys Val Glu His Gln Leu Asn Gln Arg Gln Val Leu	
180 185 190	
TTA AAA CAA GAT ATT GGT CTT GCT TTA CTA GAG TCG AGT TCT AAT AAG	624
Leu Lys Gln Asp Ile Gly Leu Ala Leu Leu Glu Ser Ser Ser Asn Lys	
195 200 205	
CCT AAC GCT GAG CCA CAA AAG TCT GGT TTA GGT TTG ATT GAG GCT AAA	672
Pro Asn Ala Glu Pro Gln Lys Ser Gly Leu Gly Leu Ile Glu Ala Lys	
210 215 220	
GAA CAG CAA ATG AAC GCT GCT GAT AAT TGG CAT TGT TTA CTG GGC CAT	720
Glu Gln Gln Met Asn Ala Ala Asp Asn Trp His Cys Leu Leu Gly His	
225 230 235 240	



CTT GAT GAT AGT TAT CGT TTT GCA CTG AGT ATT GGT CAG TGT CAG CAA 768
 Leu Asp Asp Ser Tyr Arg Phe Ala Leu Ser Ile Gly Gln Cys Gln Gln
 245 250 255
 ATA AGT ATT GCA GCA GAA GAA GTG AAT TTT AAA GCT GTT GTT CGA GCT 816
 Ile Ser Ile Ala Ala Glu Glu Val Asn Phe Lys Ala Val Val Arg Ala
 260 265 270
 TCA GCT AAG ACT AGC 831
 Ser Ala Lys Thr Ser
 275

SEQ ID NO : 4

SEQUENCE LENGTH : 2910

SEQUENCE TYPE : Nucleic acid

STRANDNESS : Double strand

TOPOLOGY : Linear

MOLECULE TYPE : Genomic DNA

ORIGINAL SOURCE : Shewanella putrefaciens SCRC-2874

(FERM BP-1625)

SEQUENCE

ATG AGT ATG TTT TTA AAT TCA AAA CTT TCG CGC TCA GTC AAA CTT GCC 48
 Met Ser Met Phe Leu Asn Ser Lys Leu Ser Arg Ser Val Lys Leu Ala
 1 5 10 15
 ATA TCC GCA GGC TTA ACA GCC TCG CTA GCT ATG CCT GTT TTT GCA GAA 96
 Ile Ser Ala Gly Leu Thr Ala Ser Leu Ala Met Pro Val Phe Ala Glu
 20 25 30
 GAA ACT GCT GCT GAA GAA CAA ATA GAA AGA GTC GCA GTG ACC GGA TCG 144
 Glu Thr Ala Ala Glu Glu Gln Ile Glu Arg Val Ala Val Thr Gly Ser
 35 40 45
 CGA ATC GCT AAA GCA GAG CTA ACT CAA CCA GCT CCA GTC GTC AGC CTT 192
 Arg Ile Ala Lys Ala Glu Leu Thr Gln Pro Ala Pro Val Val Ser Leu
 50 55 60
 TCA GCC GAA GAA CTG ACA AAA TTT GGT AAT CAA GAT TTA GGT AGC GTA 240
 Ser Ala Glu Glu Leu Thr Lys Phe Gly Asn Gln Asp Leu Gly Ser Val
 65 70 75 80



CTA GCA GAA TTA CCT GCT ATT GGT GCA ACC AAC ACT ATT ATT GGT AAT	288
Leu Ala Glu Leu Pro Ala Ile Gly Ala Thr Asn Thr Ile Ile Gly Asn	
85 90 95	
AAC AAT AGC AAC TCA AGC GCA GGT GTT AGC TCA GCA GAC TTG CGT CGT	336
Asn Asn Ser Asn Ser Ser Ala Gly Val Ser Ser Ala Asp Leu Arg Arg	
100 105 110	
CTA GGT GCT AAC AGA ACC TTA GTA TTA GTC AAC GGT AAG CGC TAC GTT	384
Leu Gly Ala Asn Arg Thr Leu Val Leu Val Asn Gly Lys Arg Tyr Val	
115 120 125	
GCC GGC CAA CCG GGC TCA GCT GAG GTA GAT TTG TCA ACT ATA CCA ACT	432
Ala Gly Gln Pro Gly Ser Ala Glu Val Asp Leu Ser Thr Ile Pro Thr	
130 135 140	
AGC ATG ATC TCG CGA GTT GAG ATT GTA ACC GGC GGT GCT TCA GCA ATT	480
Ser Met Ile Ser Arg Val Glu Ile Val Thr Gly Gly Ala Ser Ala Ile	
145 150 155 160	
TAT GGT TCG GAC GCT GTA TCA GGT GTT ATC AAC GTT ATC CTT AAA GAA	528
Tyr Gly Ser Asp Ala Val Ser Gly Val Ile Asn Val Ile Leu Lys Glu	
165 170 175	
GAC TTT GAA GGC TTT GAG TTT AAC GCA CGT ACT AGC GGT TCT ACT GAA	576
Asp Phe Glu Gly Phe Glu Phe Asn Ala Arg Thr Ser Gly Ser Thr Glu	
180 185 190	
AGT GTA GGC ACT CAA GAG CAC TCT TTT GAC ATT TTG GGT GGT GCA AAC	624
Ser Val Gly Thr Gln Glu His Ser Phe Asp Ile Leu Gly Gly Ala Asn	
195 200 205	
GTT GCA GAT GGA CGT GGT AAT GTA ACC TTC TAC GCA GGT TAT GAA CGT	672
Val Ala Asp Gly Arg Gly Asn Val Thr Phe Tyr Ala Gly Tyr Glu Arg	
210 215 220	
ACA AAA GAA GTC ATG GCT ACC GAC ATT CGC CAA TTC GAT GCT TGG GGA	720
Thr Lys Glu Val Met Ala Thr Asp Ile Arg Gln Phe Asp Ala Trp Gly	
225 230 235 240	
ACA ATT AAA AAC GAA GCC GAT GGT GGT GAA GAT GAT GGT ATT CCA GAC	768
Thr Ile Lys Asn Glu Ala Asp Gly Gly Glu Asp Asp Gly Ile Pro Asp	
245 250 255	



AGA CTA CGT GTA CCA CGA GTT TAT TCT GAA ATG ATT AAT GCT ACC GGT	816
Arg Leu Arg Val Pro Arg Val Tyr Ser Glu Met Ile Asn Ala Thr Gly	
260 265 270	
GTT ATC AAT GCA TTT GGT GGT GGA ATT GGT CGC TCA ACC TTT GAC AGT	864
Val Ile Asn Ala Phe Gly Gly Gly Ile Gly Arg Ser Thr Phe Asp Ser	
275 280 285	
AAC GGC AAT CCT ATT GCA CAA CAA GAA CGT GAT GGG ACT AAC AGC TTT	912
Asn Gly Asn Pro Ile Ala Gln Gln Glu Arg Asp Gly Thr Asn Ser Phe	
290 295 300	
GCA TTT GGT TCA TTC CCT AAT GGC TGT GAC ACA TGT TTC AAC ACT GAA	960
Ala Phe Gly Ser Phe Pro Asn Gly Cys Asp Thr Cys Phe Asn Thr Glu	
305 310 315 320	
GCA TAC GAA AAC TAT ATT CCA GGG GTA GAA AGA ATA AAC GTT GGC TCA	1008
Ala Tyr Glu Asn Tyr Ile Pro Gly Val Glu Arg Ile Asn Val Gly Ser	
325 330 335	
TCA TTC AAC TTT GAT TTT ACC GAT AAC ATT CAA TTT TAC ACT GAC TTC	1056
Ser Phe Asn Phe Asp Phe Thr Asp Asn Ile Gln Phe Tyr Thr Asp Phe	
340 345 350	
AGA TAT GTA AAG TCA GAT ATT CAG CAA CAA TTT CAG CCT TCA TTC CGT	1104
Arg Tyr Val Lys Ser Asp Ile Gln Gln Gln Phe Gln Pro Ser Phe Arg	
355 360 365	
TTT GGT AAC ATT AAT ATC AAT GTT GAA GAT AAC GCC TTT TTG AAT GAC	1152
Phe Gly Asn Ile Asn Ile Asn Val Glu Asp Asn Ala Phe Leu Asn Asp	
370 375 380	
GAC TTG CGT CAG CAA ATG CTC GAT GCG GGT CAA ACC AAT GCT AGT TTT	1200
Asp Leu Arg Gln Gln Met Leu Asp Ala Gly Gln Thr Asn Ala Ser Phe	
385 390 395 400	
GCC AAG TTT TTT GAT GAA TTA GGA AAT CGC TCA GCA GAA AAT AAA CGC	1248
Ala Lys Phe Phe Asp Glu Leu Gly Asn Arg Ser Ala Glu Asn Lys Arg	
405 410 415	
GAA CTT TTC CGT TAC GTA GGT GGC TTT AAA GGT GGC TTT GAT ATT AGC	1296
Glu Leu Phe Arg Tyr Val Gly Gly Phe Lys Gly Gly Phe Asp Ile Ser	
420 425 430	



GAA ACC ATA TTT GAT TAC GAC CTT TAC TAT GTT TAT GGC GAG ACT AAT	1344
Glu Thr Ile Phe Asp Tyr Asp Leu Tyr Tyr Val Tyr Gly Glu Thr Asn	
435 440 445	
AAC CGT CGT AAA ACC CTT AAT GAC CTA ATT CCT GAT AAC TTT GTC GCA	1392
Asn Arg Arg Lys Thr Leu Asn Asp Leu Ile Pro Asp Asn Phe Val Ala	
450 455 460	
GCT GTC GAC TCT GTT ATT GAT CCT GAT ACT GGC TTA GCA GCG TGT CGC	1440
Ala Val Asp Ser Val Ile Asp Pro Asp Thr Gly Leu Ala Ala Cys Arg	
465 470 475 480	
TCA CAA GTA GCA AGC GCT CAA GGC GAT GAC TAT ACA GAT CCC GCG TCT	1488
Ser Gln Val Ala Ser Ala Gln Gly Asp Asp Tyr Thr Asp Pro Ala Ser	
485 490 495	
GTA AAT GGT AGC GAC TGT GTT GCT TAT AAC CCA TTT GGC ATG GGT CAA	1536
Val Asn Gly Ser Asp Cys Val Ala Tyr Asn Pro Phe Gly Met Gly Gln	
500 505 510	
GCT TCA GCA GAA GCC CGC GAC TGG GTT TCT GCT GAT GTG ACT CGT GAA	1584
Ala Ser Ala Glu Ala Arg Asp Trp Val Ser Ala Asp Val Thr Arg Glu	
515 520 525	
GAC AAA ATA ACT CAA CAA GTG ATT GGT GGT ACT CTC GGT ACC GAT TCT	1632
Asp Lys Ile Thr Gln Gln Val Ile Gly Gly Thr Leu Gly Thr Asp Ser	
530 535 540	
GAA GAA CTA TTT GAG CTT CAA GGT GGT GCA ATC GCT ATG GTT GTT GGT	1680
Glu Glu Leu Phe Glu Leu Gln Gly Gly Ala Ile Ala Met Val Val Gly	
545 550 555 560	
TTT GAA TAC CGT GAA GAA ACG TCT GGT TCA ACA ACC GAT GAA TTT ACT	1728
Phe Glu Tyr Arg Glu Glu Thr Ser Gly Ser Thr Thr Asp Glu Phe Thr	
565 570 575	
AAA GCA GGT TTC TTG ACA AGC GCT GCA ACG CCA GAT TCT TAT GGC GAA	1776
Lys Ala Gly Phe Leu Thr Ser Ala Ala Thr Pro Asp Ser Tyr Gly Glu	
580 585 590	
TAC GAC GTG ACT GAG TAT TTT GTT GAG GTG AAC ATC CCA GTA CTA AAA	1824
Tyr Asp Val Thr Glu Tyr Phe Val Glu Val Asn Ile Pro Val Leu Lys	
595 600 605	



GAA TTA CCT TTT GCA CAT GAG TTG AGC TTT GAC GGT GCA TAC CGT AAT	1872
Glu Leu Pro Phe Ala His Glu Leu Ser Phe Asp Gly Ala Tyr Arg Asn	
610 615 620	
GCT GAT TAC TCA CAT GCC GGT AAG ACT GAA GCA TGG AAA GCT GGT ATG	1920
Ala Asp Tyr Ser His Ala Gly Lys Thr Glu Ala Trp Lys Ala Gly Met	
625 630 635 640	
TTC TAC TCA CCA TTA GAG CAA CTT GCA TTA CGT GGT ACG GTA GGT GAA	1968
Phe Tyr Ser Pro Leu Glu Gln Leu Ala Leu Arg Gly Thr Val Gly Glu	
645 650 655	
GCA GTA CGA GCA CCA AAC ATT GCA GAA GCC TTT AGT CCA CGC TCT CCT	2016
Ala Val Arg Ala Pro Asn Ile Ala Glu Ala Phe Ser Pro Arg Ser Pro	
660 665 670	
GGT TTT GGC CGC GTT TCA GAT CCA TGT GAT GCA GAT AAC ATT AAT GAC	2064
Gly Phe Gly Arg Val Ser Asp Pro Cys Asp Ala Asp Asn Ile Asn Asp	
675 680 685	
GAT CCG GAT CGC GTG TCA AAC TGT GCA GCA TTG GGG ATC CCT CCA GGA	2112
Asp Pro Asp Arg Val Ser Asn Cys Ala Ala Leu Gly Ile Pro Pro Gly	
690 695 700	
TTC CAA GCT AAT GAT AAC GTC AGT GTA GAT ACC TTA TCT GGT GGT AAC	2160
Phe Gln Ala Asn Asp Asn Val Ser Val Asp Thr Leu Ser Gly Gly Asn	
705 710 715 720	
CCA GAT CTA AAA CCT GAA ACA TCA ACA TCC TTT ACA GGT GGT CTT GTT	2208
Pro Asp Leu Lys Pro Glu Thr Ser Thr Ser Phe Thr Gly Gly Leu Val	
725 730 735	
TGG ACA CCA ACG TTT GCT GAC AAT CTA TCA TTC ACT GTC GAT TAT TAT	2256
Trp Thr Pro Thr Phe Ala Asp Asn Leu Ser Phe Thr Val Asp Tyr Tyr	
740 745 750	
GAT ATT CAA ATT GAG GAT GCT ATT TTG TCA GTA GCC ACC CAG ACT GTG	2304
Asp Ile Gln Ile Glu Asp Ala Ile Leu Ser Val Ala Thr Gln Thr Val	
755 760 765	
GCT GAT AAC TGT GTT GAC TCA ACT GGC GGA CCT GAC ACC GAC TTC TGT	2352
Ala Asp Asn Cys Val Asp Ser Thr Gly Gly Pro Asp Thr Asp Phe Cys	
770 775 780	





CGT GCA TTC CTA GGT ATT AAG GTA ATG ATG
Arg Ala Phe Leu Gly Ile Lys Val Met Met
965 970

2910

SEQ ID NO : 5

SEQUENCE LENGTH : 864

SEQUENCE TYPE : Nucleic acid

STRANDNESS : Double strand

TOPOLOGY : Linear

MOLECULE TYPE : Genomic DNA

ORIGINAL SOURCE : Shewanella putrefaciens SCRC-2874
(FERM BP-1625)

SEQUENCE

ATG GCA AAA ATA AAT AGT GAA CAC TTG GAT GAA GCT ACT ATT ACT TCG	48
Met Ala Lys Ile Asn Ser Glu His Leu Asp Glu Ala Thr Ile Thr Ser	
1 5 10 15	
AAT AAG TGT ACG CAA ACA GAG ACT GAG GCT CGG CAT AGA AAT GCC ACT	96
Asn Lys Cys Thr Gln Thr Glu Thr Glu Ala Arg His Arg Asn Ala Thr	
20 25 30	
ACA ACA CCT GAG ATG CGC CGA TTC ATA CAA GAG TCG GAT CTC AGT GTT	144
Thr Thr Pro Glu Met Arg Arg Phe Ile Gln Glu Ser Asp Leu Ser Val	
35 40 45	
AGC CAA CTG TCT AAA ATA TTA AAT ATC AGT GAA GCT ACC GTA CGT AAG	192
Ser Gln Leu Ser Lys Ile Leu Asn Ile Ser Glu Ala Thr Val Arg Lys	
50 55 60	
TGG CGC AAG CGT GAC TCT GTC GAA AAC TGT CCT AAT ACC CCG CAC CAT	240
Trp Arg Lys Arg Asp Ser Val Glu Asn Cys Pro Asn Thr Pro His His	
65 70 75 80	
CTC AAT ACC ACG CTA ACC CCT TTG CAA GAA TAT GTG GTT GTG GGC CTG	288
Leu Asn Thr Thr Leu Thr Pro Leu Gln Glu Tyr Val Val Val Gly Leu	
85 90 95	
CGT TAT CAA TTG AAA ATG CCA TTA GAC AGA TTG CTC AAA GCA ACC CAA	336
Arg Tyr Gln Leu Lys Met Pro Leu Asp Arg Leu Leu Lys Ala Thr Gln	
100 105 110	



GAG TTT ATC AAT CCA AAC GTG TCG CGC TCA GGT TTA GCA AGA TGT TTG	384
Glu Phe Ile Asn Pro Asn Val Ser Arg Ser Gly Leu Ala Arg Cys Leu	
115 120 125	
AAG CGT TAT GGC GTT TCA CGG GTG AGT GAT ATC CAA AGC CCA CAC GTA	432
Lys Arg Tyr Gly Val Ser Arg Val Ser Asp Ile Gln Ser Pro His Val	
130 135 140	
CCA ATG CGC TAC TTT AAT CAA ATT CCA GTC ACT CAA GGC AGC GAT GTG	480
Pro Met Arg Tyr Phe Asn Gln Ile Pro Val Thr Gln Gly Ser Asp Val	
145 150 155 160	
CAA ACC TAC ACC CTG CAC TAT GAA ACG CTG GCA AAA ACC TTA GCC TTA	528
Gln Thr Tyr Thr Leu His Tyr Glu Thr Leu Ala Lys Thr Leu Ala Leu	
165 170 175	
CCT AGT ACC GAT GGT GAC AAT GTG GTG CAA GTG GTG TCT CTC ACC ATT	576
Pro Ser Thr Asp Gly Asp Asn Val Val Gln Val Val Ser Leu Thr Ile	
180 185 190	
CCA CCA AAG TTA ACC GAA GAA GCA CCC AGT TCA ATT TTG CTC GGC ATT	624
Pro Pro Lys Leu Thr Glu Glu Ala Pro Ser Ser Ile Leu Leu Gly Ile	
195 200 205	
GAT CCT CAT AGC GAC TGG ATC TAT CTC GAC ATA TAC CAA GAT GGC AAT	672
Asp Pro His Ser Asp Trp Ile Tyr Leu Asp Ile Tyr Gln Asp Gly Asn	
210 215 220	
ACA CAA GCC ACG AAT AGA TAT ATG GCT TAT GTG CTA AAA CAC GGG CCA	720
Thr Gln Ala Thr Asn Arg Tyr Met Ala Tyr Val Leu Lys His Gly Pro	
225 230 235 240	
TTC CAT TTA CGA AAG TTA CTC GTG CGT AAC TAT CAC ACC TTT TTA CAG	768
Phe His Leu Arg Lys Leu Leu Val Arg Asn Tyr His Thr Phe Leu Gln	
245 250 255	
CGC TTT CCT GGA GCG ACG CAA AAT CGC CGC CCC TCT AAA GAT ATG CCT	816
Arg Phe Pro Gly Ala Thr Gln Asn Arg Arg Pro Ser Lys Asp Met Pro	
260 265 270	
GAA ACA ATC AAC AAG ACG CCT GAA ACA CAG GCA CCC AGT GGA GAC TCA	864
Glu Thr Ile Asn Lys Thr Pro Glu Thr Gln Ala Pro Ser Gly Asp Ser	
275 280 285	

SEQ ID NO : 6

SEQUENCE LENGTH : 8268



SEQUENCE TYPE : Nucleic acid

STRANDNESS : Double strand

TOPOLOGY : Linear

MOLECULE TYPE : Genomic DNA

ORIGINAL SOURCE : *Shewanella putrefaciens* SCRC-2874

(FERM BP-1625)

SEQUENCE

ATG AGC CAG ACC TCT AAA CCT ACA AAC TCA GCA ACT GAG CAA GCA CAA	48
Met Ser Gln Thr Ser Lys Pro Thr Asn Ser Ala Thr Glu Gln Ala Gln	
1 5 10 15	
GAC TCA CAA GCT GAC TCT CGT TTA AAT AAA CGA CTA AAA GAT ATG CCA	96
Asp Ser Gln Ala Asp Ser Arg Leu Asn Lys Arg Leu Lys Asp Met Pro	
20 25 30	
ATT GCT ATT GTT GGC ATG GCG AGT ATT TTT GCA AAC TCT CGC TAT TTG	144
Ile Ala Ile Val Gly Met Ala Ser Ile Phe Ala Asn Ser Arg Tyr Leu	
35 40 45	
AAT AAG TTT TGG GAC TTA ATC AGC GAA AAA ATT GAT GCG ATT ACT GAA	192
Asn Lys Phe Trp Asp Leu Ile Ser Glu Lys Ile Asp Ala Ile Thr Glu	
50 55 60	
TTA CCA TCA ACT CAC TGG CAG CCT GAA GAA TAT TAC GAC GCA GAT AAA	240
Leu Pro Ser Thr His Trp Gln Pro Glu Glu Tyr Tyr Asp Ala Asp Lys	
65 70 75 80	
ACC GCA GCA GAC AAA AGC TAC TGT AAA CGT GGT GGC TTT TTG CCA GAT	288
Thr Ala Ala Asp Lys Ser Tyr Cys Lys Arg Gly Gly Phe Leu Pro Asp	
85 90 95	
GTA GAC TTC AAC CCA ATG GAG TTT GGC CTG CCG CCA AAC ATT TTG GAA	336
Val Asp Phe Asn Pro Met Glu Phe Gly Leu Pro Pro Asn Ile Leu Glu	
100 105 110	
CTG ACC GAT TCA TCG CAA CTA TTA TCA CTC ATC GTT GCT AAA GAA GTG	384
Leu Thr Asp Ser Ser Gln Leu Leu Ser Leu Ile Val Ala Lys Glu Val	
115 120 125	
TTG GCT GAT GCT AAC TTA CCT GAG AAT TAC GAC CGC GAT AAA ATT GGT	432
Leu Ala Asp Ala Asn Leu Pro Glu Asn Tyr Asp Arg Asp Lys Ile Gly	
130 135 140	



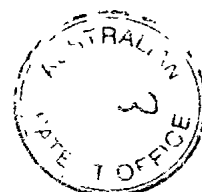
ATC ACC TTA GGT GTC GGC GGT GGT CAA AAA ATT AGC CAC AGC CTA ACA	480
Ile Thr Leu Gly Val Gly Gly Gly Gln Lys Ile Ser His Ser Leu Thr	
145 150 155 160	
GCG CGT CTG CAA TAC CCA GTA TTG AAG AAA GTA TTC GCC AAT AGC GGC	528
Ala Arg Leu Gln Tyr Pro Val Leu Lys Lys Val Phe Ala Asn Ser Gly	
165 170 175	
ATT AGT GAC ACC GAC AGC GAA ATG CTT ATC AAG AAA TTC CAA GAC CAA	576
Ile Ser Asp Thr Asp Ser Glu Met Leu Ile Lys Lys Phe Gln Asp Gln	
180 185 190	
TAT GTA CAC TGG GAA GAA AAC TCG TTC CCA GGT TCA CTT GGT AAC GTT	624
Tyr Val His Trp Glu Glu Asn Ser Phe Pro Gly Ser Leu Gly Asn Val	
195 200 205	
ATT GCG GGC CGT ATC GCC AAC CGC TTC GAT TTT GGC GGC ATG AAC TGT	672
Ile Ala Gly Arg Ile Ala Asn Arg Phe Asp Phe Gly Gly Met Asn Cys	
210 215 220	
GTC GTT GAT GCT GCC TGT GCT GGA TCA CTT GCT GCT ATG CGT ATG GCG	720
Val Val Asp Ala Ala Cys Ala Gly Ser Leu Ala Ala Met Arg Met Ala	
225 230 235 240	
CTA ACA GAG CTA ACT GAA GGT CGC TCT GAA ATG ATG ATC ACC GGT GGT	768
Leu Thr Glu Leu Thr Glu Gly Arg Ser Glu Met Met Ile Thr Gly Gly	
245 250 255	
GTG TGT ACT GAT AAC TCA CCC TCT ATG TAT ATG AGC TTT TCA AAA ACG	816
Val Cys Thr Asp Asn Ser Pro Ser Met Tyr Met Ser Phe Ser Lys Thr	
260 265 270	
CCC GCC TTT ACC ACT AAC GAA ACC ATT CAG CCA TTT GAT ATC GAC TCA	864
Pro Ala Phe Thr Thr Asn Glu Thr Ile Gln Pro Phe Asp Ile Asp Ser	
275 280 285	
AAA GGC ATG ATG ATT GGT GAA GGT ATT GGC ATG GTG GCG CTA AAG CGT	912
Lys Gly Met Met Ile Gly Glu Gly Ile Gly Met Val Ala Leu Lys Arg	
290 295 300	
CTT GAA GAT GCA GAG CGC GAT GGC GAC CGC ATT TAC TCT GTA ATT AAA	960
Leu Glu Asp Ala Glu Arg Asp Gly Asp Arg Ile Tyr Ser Val Ile Lys	
305 310 315 320	



GGT GTG GGT GCA TCA TCT GAC GGT AAG TTT AAA TCA ATC TAT GCC CCT	1008
Gly Val Gly Ala Ser Ser Asp Gly Lys Phe Lys Ser Ile Tyr Ala Pro	
325 330 335	
CGC CCA TCA GGC CAA GCT AAA GCA CTT AAC CGT GCC TAT GAT GAC GCA	1056
Arg Pro Ser Gly Gln Ala Lys Ala Leu Asn Arg Ala Tyr Asp Asp Ala	
340 345 350	
GGT TTT GCG CCG CAT ACC TTA GGT CTA ATT GAA GCT CAC GGA ACA GGT	1104
Gly Phe Ala Pro His Thr Leu Gly Leu Ile Glu Ala His Gly Thr Gly	
355 360 365	
ACT GCA GCA GGT GAC GCG GCA GAG TTT GCC GGC CTT TGC TCA GTA TTT	1152
Thr Ala Ala Gly Asp Ala Ala Glu Phe Ala Gly Leu Cys Ser Val Phe	
370 375 380	
GCT GAA GGC AAC GAT ACC AAG CAA CAC ATT GCG CTA GGT TCA GTT AAA	1200
Ala Glu Gly Asn Asp Thr Lys Gln His Ile Ala Leu Gly Ser Val Lys	
385 390 395 400	
TCA CAA ATT GGT CAT ACT AAA TCA ACT GCA GGT ACA GCA GGT TTA ATT	1248
Ser Gln Ile Gly His Thr Lys Ser Thr Ala Gly Thr Ala Gly Leu Ile	
405 410 415	
AAA GCT GCT CTT GCT TTG CAT CAC AAG GTA CTG CCG CCG ACC ATT AAC	1296
Lys Ala Ala Leu Ala Leu His His Lys Val Leu Pro Pro Thr Ile Asn	
420 425 430	
GTT AGT CAG CCA AGC CCT AAA CTT GAT ATC GAA AAC TCA CCG TTT TAT	1344
Val Ser Gln Pro Ser Pro Lys Leu Asp Ile Glu Asn Ser Pro Phe Tyr	
435 440 445	
CTA AAC ACT GAG ACT CGT CCA TGG TTA CCA CGT GTT GAT GGT ACG CCG	1392
Leu Asn Thr Glu Thr Arg Pro Trp Leu Pro Arg Val Asp Gly Thr Pro	
450 455 460	
CGC CGC GCG GGT ATT AGC TCA TTT GGT TTT GGT GGC ACT AAC TTC CAT	1440
Arg Arg Ala Gly Ile Ser Ser Phe Gly Phe Gly Gly Thr Asn Phe His	
465 470 475 480	
TTT GTA CTA GAA GAG TAC AAC CAA GAA CAC AGC CGT ACT GAT AGC GAA	1488
Phe Val Leu Glu Glu Tyr Asn Gln Glu His Ser Arg Thr Asp Ser Glu	
485 490 495	



AAA GCT AAG TAT CGT CAA CGC CAA GTG GCG CAA AGC TTC CTT GTT AGC	1536
Lys Ala Lys Tyr Arg Gln Arg Gln Val Ala Gln Ser Phe Leu Val Ser	
500 505 510	
GCA AGC GAT AAA GCA TCG CTA ATT AAC GAG TTA AAC GTA CTA GCA GCA	1584
Ala Ser Asp Lys Ala Ser Leu Ile Asn Glu Leu Asn Val Leu Ala Ala	
515 520 525	
TCT GCA AGC CAA GCT GAG TTT ATC CTC AAA GAT GCA GCA GCA AAC TAT	1632
Ser Ala Ser Gln Ala Glu Phe Ile Leu Lys Asp Ala Ala Ala Asn Tyr	
530 535 540	
GGC GTA CGT GAG CTT GAT AAA AAT GCA CCA CGG ATC GGT TTA GTT GCA	1680
Gly Val Arg Glu Leu Asp Lys Asn Ala Pro Arg Ile Gly Leu Val Ala	
545 550 555 560	
AAC ACA GCT GAA GAG TTA GCA GGC CTA ATT AAG CAA GCA CTT GCC AAA	1728
Asn Thr Ala Glu Glu Leu Ala Gly Leu Ile Lys Gln Ala Leu Ala Lys	
565 570 575	
CTA GCA GCT AGC GAT GAT AAC GCA TGG CAG CTA CCT GGT GGC ACT AGC	1776
Leu Ala Ala Ser Asp Asp Asn Ala Trp Gln Leu Pro Gly Gly Thr Ser	
580 585 590	
TAC CGC GCC GCT GCA GTA GAA GGT AAA GTT GCC GCA CTG TTT GCT GGC	1824
Tyr Arg Ala Ala Ala Val Glu Gly Lys Val Ala Ala Leu Phe Ala Gly	
595 600 605	
CAA GGT TCA CAA TAT CTC AAT ATG GGC CGT GAC CTT ACT TGT TAT TAC	1872
Gln Gly Ser Gln Tyr Leu Asn Met Gly Arg Asp Leu Thr Cys Tyr Tyr	
610 615 620	
CCA GAG ATG CGT CAG CAA TTT GTA ACT GCA GAT AAA GTA TTT GCC GCA	1920
Pro Glu Met Arg Gln Gln Phe Val Thr Ala Asp Lys Val Phe Ala Ala	
625 630 635 640	
AAT GAT AAA ACG CCG TTA TCG CAA ACT CTG TAT CCA AAG CCT GTA TTT	1968
Asn Asp Lys Thr Pro Leu Ser Gln Thr Leu Tyr Pro Lys Pro Val Phe	
645 650 655	
AAT AAA GAT GAA TTA AAG GCT CAA GAA GCC ATT TTG ACC AAT ACC GCC	2016
Asn Lys Asp Glu Leu Lys Ala Gln Glu Ala Ile Leu Thr Asn Thr Ala	
660 665 670	



AAT GCC CAA AGC GCA ATT GGT GCG ATT TCA ATG GGT CAA TAC GAT TTG	2064
Asn Ala Gln Ser Ala Ile Gly Ala Ile Ser Met Gly Gln Tyr Asp Leu	
675 680 685	
TTT ACT GCG GCT GGC TTT AAT GCC GAC ATG GTT GCA GGC CAT AGC TTT	2112
Phe Thr Ala Ala Gly Phe Asn Ala Asp Met Val Ala Gly His Ser Phe	
690 695 700	
GGT GAG CTA AGT GCA CTG TGT GCT GCA GGT GTT ATT TCA GCT GAT GAC	2160
Gly Glu Leu Ser Ala Leu Cys Ala Ala Gly Val Ile Ser Ala Asp Asp	
705 710 715 720	
TAC TAC AAG CTG GCT TTT GCT CGT GGT GAG GCT ATG GCA ACA AAA GCA	2208
Tyr Tyr Lys Leu Ala Phe Ala Arg Gly Glu Ala Met Ala Thr Lys Ala	
725 730 735	
CCG GCT AAA GAC GGC GTT GAA GCA GAT GCA GGA GCA ATG TTT GCA ATC	2256
Pro Ala Lys Asp Gly Val Glu Ala Asp Ala Gly Ala Met Phe Ala Ile	
740 745 750	
ATA ACC AAG AGT GCT GCA GAC CTT GAA ACC GTT GAA GCC ACC ATC GCT	2304
Ile Thr Lys Ser Ala Ala Asp Leu Glu Thr Val Glu Ala Thr Ile Ala	
755 760 765	
AAA TTT GAT GGG GTG AAA GTC GCT AAC TAT AAC GCG CCA ACG CAA TCA	2352
Lys Phe Asp Gly Val Lys Val Ala Asn Tyr Asn Ala Pro Thr Gln Ser	
770 775 780	
GTA ATT GCA GGC CCA ACA GCA ACT ACC GCT GAT GCG GCT AAA GCG CTA	2400
Val Ile Ala Gly Pro Thr Ala Thr Thr Ala Asp Ala Ala Lys Ala Leu	
785 790 795 800	
ACT GAG CTT GGT TAC AAA GCG ATT AAC CTG CCA GTA TCA GGT GCA TTC	2448
Thr Glu Leu Gly Tyr Lys Ala Ile Asn Leu Pro Val Ser Gly Ala Phe	
805 810 815	
CAC ACT GAA CTT GTT GGT CAC GCT CAA GCG CCA TTT GCT AAA GCG ATT	2496
His Thr Glu Leu Val Gly His Ala Gln Ala Pro Phe Ala Lys Ala Ile	
820 825 830	
GAC GCA GCC AAA TTT ACT AAA ACA AGC CGA GCA CTT TAC TCA AAT GCA	2544
Asp Ala Ala Lys Phe Thr Lys Thr Ser Arg Ala Leu Tyr Ser Asn Ala	
835 840 845	



ACT GGC GGA CTT TAT GAA AGC ACT GCT GCA AAG ATT AAA GCC TCG TTT	2592
Thr Gly Gly Leu Tyr Glu Ser Thr Ala Ala Lys Ile Lys Ala Ser Phe	
850 855 860	
AAG AAA CAT ATG CTT CAA TCA GTG CGC TTT ACT AGC CAG CTA GAA GCC	2640
Lys Lys His Met Leu Gln Ser Val Arg Phe Thr Ser Gln Leu Glu Ala	
865 870 875 880	
ATG TAC AAC GAC GGC GCC CGT GTA TTT GTT GAA TTT GGT CCA AAG AAC	2688
Met Tyr Asn Asp Gly Ala Arg Val Phe Val Glu Phe Gly Pro Lys Asn	
885 890 895	
ATC TTA CAA AAA TTA GTT CAA GGC ACG CTT GTC AAC ACT GAA AAT GAA	2736
Ile Leu Gln Lys Leu Val Gln Gly Thr Leu Val Asn Thr Glu Asn Glu	
900 905 910	
GTT TGC ACT ATC TCT ATC AAC CCT AAT CCT AAA GTT GAT AGT GAT CTG	2784
Val Cys Thr Ile Ser Ile Asn Pro Asn Pro Lys Val Asp Ser Asp Leu	
915 920 925	
CAG CTT AAG CAA GCA GCA ATG CAG CTA GCG GTT ACT GGT GTG GTA CTC	2832
Gln Leu Lys Gln Ala Ala Met Gln Leu Ala Val Thr Gly Val Val Leu	
930 935 940	
AGT GAA ATT GAC CCA TAC CAA GCC GAT ATT GCC GCA CCA GCG AAA AAG	2880
Ser Glu Ile Asp Pro Tyr Gln Ala Asp Ile Ala Ala Pro Ala Lys Lys	
945 950 955 960	
TCG CCA ATG AGC ATT TCG CTT AAT GCT GCT AAC CAT ATC AGC AAA GCA	2928
Ser Pro Met Ser Ile Ser Leu Asn Ala Ala Asn His Ile Ser Lys Ala	
965 970 975	
ACT CGC GCT AAG ATG GCC AAG TCT TTA GAG ACA GGT ATC GTC ACC TCG	2976
Thr Arg Ala Lys Met Ala Lys Ser Leu Glu Thr Gly Ile Val Thr Ser	
980 985 990	
CAA ATA GAA CAT GTT ATT GAA GAA AAA ATC GTT GAA GTT GAG AAA CTG	3024
Gln Ile Glu His Val Ile Glu Glu Lys Ile Val Glu Val Glu Lys Leu	
995 1000 1005	
GTT GAA GTC GAA AAG ATC GTC GAA AAA GTG GTT GAA GTA GAG AAA GTT	3072
Val Glu Val Glu Lys Ile Val Glu Lys Val Val Glu Val Glu Lys Val	
1010 1015 1020	



GTT GAG GTT GAA GCT CCT GTT AAT TCA GTG CAA GCC AAT GCA ATT CAA	3120
Val Glu Val Glu Ala Pro Val Asn Ser Val Gln Ala Asn Ala Ile Gln	
1025 1030 1035 1040	
ACC CGT TCA GTT GTC GCT CCA GTA ATA GAG AAC CAA GTC GTG TCT AAA	3168
Thr Arg Ser Val Val Ala Pro Val Ile Glu Asn Gln Val Val Ser Lys	
1045 1050 1055	
AAC AGT AAG CCA GCA GTC CAG AGC ATT AGT GGT GAT GCA CTC AGC AAC	3216
Asn Ser Lys Pro Ala Val Gln Ser Ile Ser Gly Asp Ala Leu Ser Asn	
1060 1065 1070	
TTT TTT GCT GCA CAG CAG CAA ACC GCA CAG TTG CAT CAG CAG TTC TTA	3264
Phe Phe Ala Ala Gln Gln Gln Thr Ala Gln Leu His Gln Gln Phe Leu	
1075 1080 1085	
GCT ATT CCG CAG CAA TAT GGT GAG ACG TTC ACT ACG CTG ATG ACC GAG	3312
Ala Ile Pro Gln Gln Tyr Gly Glu Thr Phe Thr Thr Leu Met Thr Glu	
1090 1095 1100	
CAA GCT AAA CTG GCA AGT TCT GGT GTT GCA ATT CCA GAG AGT CTG CAA	3360
Gln Ala Lys Leu Ala Ser Ser Gly Val Ala Ile Pro Glu Ser Leu Gln	
1105 1110 1115 1120	
CGC TCA ATG GAG CAA TTC CAC CAA CTA CAA GCG CAA ACA CTA CAA AGC	3408
Arg Ser Met Glu Gln Phe His Gln Leu Gln Ala Gln Thr Leu Gln Ser	
1125 1130 1135	
CAC ACC CAG TTC CTT GAG ATG CAA GCG GGT AGC AAC ATT GCA GCG TTA	3456
His Thr Gln Phe Leu Glu Met Gln Ala Gly Ser Asn Ile Ala Ala Leu	
1140 1145 1150	
AAC CTA CTC AAT AGC AGC CAA GCA ACT TAC GCT CCA GCC ATT CAC AAT	3504
Asn Leu Leu Asn Ser Ser Gln Ala Thr Tyr Ala Pro Ala Ile His Asn	
1155 1160 1165	
GAA GCG ATT CAA AGC CAA GTG GTT CAA AGC CAA ACT GCA GTC CAG CCA	3552
Glu Ala Ile Gln Ser Gln Val Val Gln Ser Gln Thr Ala Val Gln Pro	
1170 1175 1180	
GTA ATT TCA ACA CAA GTT AAC CAT GTG TCA GAG CAG CCA ACT CAA GCT	3600
Val Ile Ser Thr Gln Val Asn His Val Ser Glu Gln Pro Thr Gln Ala	
1185 1190 1195 1200	



CCA GCT CCA AAA GCG CAG CCA GCA CCT GTG ACA ACT GCA GTT CAA ACT	3648
Pro Ala Pro Lys Ala Gln Pro Ala Pro Val Thr Thr Ala Val Gln Thr	
1205 1210 1215	
GCT CCG GCA CAA GTT GTT CGT CAA GCC GCA CCA GTT CAA GCC GCT ATT	3696
Ala Pro Ala Gln Val Val Arg Gln Ala Ala Pro Val Gln Ala Ala Ile	
1220 1225 1230	
GAA CCG ATT AAT ACA AGT GTT GCG ACT ACA ACG CCT TCA GCC TTC AGC	3744
Glu Pro Ile Asn Thr Ser Val Ala Thr Thr Thr Pro Ser Ala Phe Ser	
1235 1240 1245	
GCC GAA ACA GCC CTG AGC GCA ACA AAA GTC CAA GCC ACT ATG CTT GAA	3792
Ala Glu Thr Ala Leu Ser Ala Thr Lys Val Gln Ala Thr Met Leu Glu	
1250 1255 1260	
GTG GTT GCT GAG AAA ACC GGT TAC CCA ACT GAA ATG CTA GAG CTT GAA	3840
Val Val Ala Glu Lys Thr Gly Tyr Pro Thr Glu Met Leu Glu Leu Glu	
1265 1270 1275 1280	
ATG GAT ATG GAA GCC GAT TTA GGC ATC GAT TCT ATC AAG CGT GTA GAA	3888
Met Asp Met Glu Ala Asp Leu Gly Ile Asp Ser Ile Lys Arg Val Glu	
1285 1290 1295	
ATT CTT GGC ACA GTA CAA GAT GAG CTA CCG GGT CTA CCT GAG CTT AGC	3936
Ile Leu Gly Thr Val Gln Asp Glu Leu Pro Gly Leu Pro Glu Leu Ser	
1300 1305 1310	
CCT GAA GAT CTA GCT GAG TGT CGA ACG CTA GGC GAA ATC GTT GAC TAT	3984
Pro Glu Asp Leu Ala Glu Cys Arg Thr Leu Gly Glu Ile Val Asp Tyr	
1315 1320 1325	
ATG GGC AGT AAA CTG CCG GCT GAA GGC TCT ATG AAT TCT CAG CTG TCT	4032
Met Gly Ser Lys Leu Pro Ala Glu Gly Ser Met Asn Ser Gln Leu Ser	
1330 1335 1340	
ACA GGT TCC GCA GCT GCG ACT CCT GCA GCG AAT GGT CTT TCT GCG GAG	4080
Thr Gly Ser Ala Ala Ala Thr Pro Ala Ala Asn Gly Leu Ser Ala Glu	
1345 1350 1355 1360	
AAA GTT CAA GCG ACT ATG ATG TCT GTG GTT GCC GAA AAG ACT GGC TAC	4128
Lys Val Gln Ala Thr Met Met Ser Val Val Ala Glu Lys Thr Gly Tyr	
1365 1370 1375	



CCA ACT GAA ATG CTA GAG CTT GAA ATG GAT ATG GAA GCC GAT TTA GGC	4176
Pro Thr Glu Met Leu Glu Leu Glu Met Asp Met Glu Ala Asp Leu Gly	
1380 1385 1390	
ATA GAT TCT ATC AAG CGC GTT GAA ATT CTT GGC ACA GTA CAA GAT GAG	4224
Ile Asp Ser Ile Lys Arg Val Glu Ile Leu Gly Thr Val Gln Asp Glu	
1395 1400 1405	
CTA CCG GGT CTA CCT GAG CTT AGC CCT GAA GAT CTA GCT GAG TGT CGT	4272
Leu Pro Gly Leu Pro Glu Leu Ser Pro Glu Asp Leu Ala Glu Cys Arg	
1410 1415 1420	
ACT CTA GGC GAA ATC GTT GAC TAT ATG AAC TCT AAA CTC GCT GAC GGC	4320
Thr Leu Gly Glu Ile Val Asp Tyr Met Asn Ser Lys Leu Ala Asp Gly	
1425 1430 1435 1440	
TCT AAG CTG CCG GCT GAA GGC TCT ATG AAT TCT CAG CTG TCT ACA AGT	4368
Ser Lys Leu Pro Ala Glu Gly Ser Met Asn Ser Gln Leu Ser Thr Ser	
1445 1450 1455	
GCC GCA GCT GCG ACT CCT GCA GCG AAT GGT CTC TCT GCG GAG AAA GTT	4416
Ala Ala Ala Ala Thr Pro Ala Ala Asn Gly Leu Ser Ala Glu Lys Val	
1460 1465 1470	
CAA GCG ACT ATG ATG TCT GTG GTT GCC GAA AAG ACT GGC TAC CCA ACT	4464
Gln Ala Thr Met Met Ser Val Val Ala Glu Lys Thr Gly Tyr Pro Thr	
1475 1480 1485	
GAA ATG CTA GAA CTT GAA ATG GAT ATG GAA GCT GAC CTT GGC ATC GAT	4512
Glu Met Leu Glu Leu Glu Met Asp Met Glu Ala Asp Leu Gly Ile Asp	
1490 1495 1500	
TCA ATC AAG CGC GTT GAA ATT CTT GGC ACA GTA CAA GAT GAG CTA CCG	4560
Ser Ile Lys Arg Val Glu Ile Leu Gly Thr Val Gln Asp Glu Leu Pro	
1505 1510 1515 1520	
GGT TTA CCT GAG CTA AAT CCA GAA GAT TTG GCA GAG TGT CGT ACT CTT	4608
Gly Leu Pro Glu Leu Asn Pro Glu Asp Leu Ala Glu Cys Arg Thr Leu	
1525 1530 1535	
GGC GAA ATC GTG ACT TAT ATG AAC TCT AAA CTC GCT GAC GGC TCT AAG	4656
Gly Glu Ile Val Thr Tyr Met Asn Ser Lys Leu Ala Asp Gly Ser Lys	
1540 1545 1550	



CTG CCA GCT GAA GGC TCT ATG CAC TAT CAG CTG TCT ACA AGT ACC GCT	4704
Leu Pro Ala Glu Gly Ser Met His Tyr Gln Leu Ser Thr Ser Thr Ala	
1555 1560 1565	
GCT GCG ACT CCT GTA GCG AAT GGT CTC TCT GCA GAA AAA GTT CAA GCG	4752
Ala Ala Thr Pro Val Ala Asn Gly Leu Ser Ala Glu Lys Val Gln Ala	
1570 1575 1580	
ACC ATG ATG TCT GTA GTT GCA GAT AAA ACT GGC TAC CCA ACT GAA ATG	4800
Thr Met Met Ser Val Val Ala Asp Lys Thr Gly Tyr Pro Thr Glu Met	
1585 1590 1595 1600	
CTT GAA CTT GAA ATG GAT ATG GAA GCC GAT TTA GGT ATC GAT TCT ATC	4848
Leu Glu Leu Glu Met Asp Met Glu Ala Asp Leu Gly Ile Asp Ser Ile	
1605 1610 1615	
AAG CGC GTT GAA ATT CTT GGC ACA GTA CAA GAT GAG CTA CCG GGT TTA	4896
Lys Arg Val Glu Ile Leu Gly Thr Val Gln Asp Glu Leu Pro Gly Leu	
1620 1625 1630	
CCT GAG CTA AAT CCA GAA GAT CTA GCA GAG TGT CGC ACC CTA GGC GAA	4944
Pro Glu Leu Asn Pro Glu Asp Leu Ala Glu Cys Arg Thr Leu Gly Glu	
1635 1640 1645	
ATC GTT GAC TAT ATG GGC AGT AAA CTG CCG GCT GAA GGC TCT GCT AAT	4992
Ile Val Asp Tyr Met Gly Ser Lys Leu Pro Ala Glu Gly Ser Ala Asn	
1650 1655 1660	
ACA AGT GCC GCT GCG TCT CTT AAT GTT AGT GCC GTT GCG GCG CCT CAA	5040
Thr Ser Ala Ala Ala Ser Leu Asn Val Ser Ala Val Ala Ala Pro Gln	
1665 1670 1675 1680	
GCT GCT GCG ACT CCT GTA TCG AAC GGT CTC TCT GCA GAG AAA GTG CAA	5088
Ala Ala Ala Thr Pro Val Ser Asn Gly Leu Ser Ala Glu Lys Val Gln	
1685 1690 1695	
AGC ACT ATG ATG TCA GTA GTT GCA GAA AAG ACC GGC TAC CCA ACT GAA	5136
Ser Thr Met Met Ser Val Val Ala Glu Lys Thr Gly Tyr Pro Thr Glu	
1700 1705	
ATG CTA GAA CTT GGC ATG GAT ATG GAA GCC GAT TTA GGT ATC ATC TCA	5184
Met Leu Glu Leu Gly Met Asp Met Glu Ala Asp Leu Gly Ile Asp Ser	
1715 1720 1725	



ATT AAA CGC GTT GAG ATT CTT GGC ACA GTA CAA GAT GAG CTA CCG GGT	5232
Ile Lys Arg Val Glu Ile Leu Gly Thr Val Gln Asp Glu Leu Pro Gly	
1730 1735 1740	
CTA CCA GAG CTT AAT CCT GAA GAT TTA GCT GAG TGC CGT ACG CTG GGC	5280
Leu Pro Glu Leu Asn Pro Glu Asp Leu Ala Glu Cys Arg Thr Leu Gly	
1745 1750 1755 1760	
GAA ATC GTT GAC TAT ATG AAC TCT AAG CTG GCT GAC GGC TCT AAG CTT	5328
Glu Ile Val Asp Tyr Met Asn Ser Lys Leu Ala Asp Gly Ser Lys Leu	
1765 1770 1775	
CCA GCT GAA GGC TCT GCT AAT ACA AGT GCC ACT GCT GCG ACT CCT GCA	5376
Pro Ala Glu Gly Ser Ala Asn Thr Ser Ala Thr Ala Ala Thr Pro Ala	
1780 1785 1790	
GTG AAT GGT CTT TCT GCT GAC AAG GTA CAG GCG ACT ATG ATG TCT GTA	5424
Val Asn Gly Leu Ser Ala Asp Lys Val Gln Ala Thr Met Met Ser Val	
1795 1800 1805	
GTT GCT GAA AAG ACC GGC TAC CCA ACT GAA ATG CTA GAA CTT GGC ATG	5472
Val Ala Glu Lys Thr Gly Tyr Pro Thr Glu Met Leu Glu Leu Gly Met	
1810 1815 1820	
GAT ATG GAA GCA GAC CTT GGT ATT GAT TCT ATT AAG CGC GTT GAA ATT	5520
Asp Met Glu Ala Asp Leu Gly Ile Asp Ser Ile Lys Arg Val Glu Ile	
1825 1830 1835 1840	
CTT GGC ACA GTA CAA GAT GAG CTC CCA GGT TTA CCT GAG CTT AAT CCT	5568
Leu Gly Thr Val Gln Asp Glu Leu Pro Gly Leu Pro Glu Leu Asn Pro	
1845 1850 1855	
GAA GAT CTC GCT GAG TGC CGC ACG CTT GGC GAA ATC GTT AGC TAT ATG	5616
Glu Asp Leu Ala Glu Cys Arg Thr Leu Gly Glu Ile Val Ser Tyr Met	
1860 1865 1870	
AAC TCT CAA CTG GCT GAT GGC TCT AAA CTT TCT ACA AGT GCG GCT GAA	5664
Asn Ser Gln Leu Ala Asp Gly Ser Lys Leu Ser Thr Ser Ala Ala Glu	
1875 1880 1885	
GGC TCT GCT GAT ACA AGT GCT GCA AAT GCT GCA AAG CCG GCA GCA ATT	5712
Gly Ser Ala Asp Thr Ser Ala Ala Asn Ala Ala Lys Pro Ala Ala Ile	
1890 1895 1900	





CTA AAA GAT GCT GAG CTA AAC CAA GCA GCA TTA GCT GGT TTA ACT AAA	6288
Leu Lys Asp Ala Glu Leu Asn Gln Ala Ala Leu Ala Gly Leu Thr Lys	
2085 2090 2095	
ACC TTA AGC CAT GAA TGG CCA CAA GTG TTC TGT CGC GCG CTA GAT ATT	6336
Thr Leu Ser His Glu Trp Pro Gln Val Phe Cys Arg Ala Leu Asp Ile	
2100 2105 2110	
GCA ACA GAT GTT GAT GCA ACC CAT CTT GCT GAT GCA ATC ACC AGT GAA	6384
Ala Thr Asp Val Asp Ala Thr His Leu Ala Asp Ala Ile Thr Ser Glu	
2115 2120 2125	
CTA TTT GAT AGC CAA GCT CAG CTA CCT GAA GTG GGC TTA AGC TTA ATT	6432
Leu Phe Asp Ser Gln Ala Gln Leu Pro Glu Val Gly Leu Ser Leu Ile	
2130 2135 2140	
GAT GGC AAA GTT AAC CGC GTA ACT CTA GTT GCT GCT GAA GCT GCA GAT	6480
Asp Gly Lys Val Asn Arg Val Thr Leu Val Ala Ala Glu Ala Ala Asp	
2145 2150 2155 2160	
AAA ACA GCA AAA GCA GAG CTT AAC AGC ACA GAT AAA ATC TTA GTG ACT	6528
Lys Thr Ala Lys Ala Glu Leu Asn Ser Thr Asp Lys Ile Leu Val Thr	
2165 2170 2175	
GGT GGG GCA AAA GGG GTG ACA TTT GAA TGT GCA CTG GCA TTA GCA TCT	6576
Gly Gly Ala Lys Gly Val Thr Phe Glu Cys Ala Leu Ala Leu Ala Ser	
2180 2185 2190	
CGC AGC CAG TCT CAC TTT ATC TTA GCT GGG CGC AGT GAA TTA CAA GCT	6624
Arg Ser Gln Ser His Phe Ile Leu Ala Gly Arg Ser Glu Leu Gln Ala	
2195 2200 2205	
TTA CCA AGC TGG GCT GAG GGT AAG CAA ACT AGC GAG CTA AAA TCA GCT	6672
Leu Pro Ser Trp Ala Glu Gly Lys Gln Thr Ser Glu Leu Lys Ser Ala	
2210 2215 2220	
GCA ATC GCA CAT ATT ATT TCT ACT GGT CAA AAG CCA ACG CCT AAG CAA	6720
Ala Ile Ala His Ile Ile Ser Thr Gly Gln Lys Pro Thr Pro Lys Gln	
2225 2230 2235 2240	
GTT GAA GCC GCT GTG TGG CCA GTG CAA AGC AGC ATT GAA ATT AAT GCC	6768
Val Glu Ala Ala Val Trp Pro Val Gln Ser Ser Ile Glu Ile Asn Ala	
2245 2250 2255	



GCC CTA GCC GCC TTT AAC AAA GTT GGC GCC TCA GCT GAA TAC GTC AGC	6816
Ala Leu Ala Ala Phe Asn Lys Val Gly Ala Ser Ala Glu Tyr Val Ser	
2260 2265 2270	
ATG GAT GTT ACC GAT AGC GCC GCA ATC ACA GCA GCA CTT AAT GGT CGC	6864
Met Asp Val Thr Asp Ser Ala Ala Ile Thr Ala Ala Leu Asn Gly Arg	
2275 2280 2285	
TCA AAT GAG ATC ACC GGT CTT ATT CAT GGC GCA GGT GTA CTA GCC GAC	6912
Ser Asn Glu Ile Thr Gly Leu Ile His Gly Ala Gly Val Leu Ala Asp	
2290 2295 2300	
AAG CAT ATT CAA GAC AAG ACT CTT GCT GAA CTT GCT AAA GTT TAT GGC	6960
Lys His Ile Gln Asp Lys Thr Leu Ala Glu Leu Ala Lys Val Tyr Gly	
2305 2310 2315 2320	
ACT AAA GTC AAC GGC CTA AAA GCG CTG CTC GCG GCA CTT GAG CCA AGC	7008
Thr Lys Val Asn Gly Leu Lys Ala Leu Leu Ala Ala Leu Glu Pro Ser	
2325 2330 2335	
AAA ATT AAA TTA CTT GCT ATG TTC TCA TCT GCA GCA GGT TTT TAC GGT	7056
Lys Ile Lys Leu Leu Ala Met Phe Ser Ser Ala Ala Gly Phe Tyr Gly	
2340 2345 2350	
AAT ATC GGC CAA AGC GAT TAC GCG ATG TCG AAC GAT ATT CTT AAC AAG	7104
Asn Ile Gly Gln Ser Asp Tyr Ala Met Ser Asn Asp Ile Leu Asn Lys	
2355 2360 2365	
GCA GCG CTG CAG TTC ACC GCT CGC AAC CCA CAA GCT AAA GTC ATG AGC	7152
Ala Ala Leu Gln Phe Thr Ala Arg Asn Pro Gln Ala Lys Val Met Ser	
2370 2375 2380	
TTT AAC TGG GGT CCT TGG GAT GGC GGC ATG GTT AAC CCA GCG CTT AAA	7200
Phe Asn Trp Gly Pro Trp Asp Gly Gly Met Val Asn Pro Ala Leu Lys	
2385 2390 2395 2400	
AAG ATG TTT ACC GAG CGT GGT GTG TAC GTT ATT CCA CTA AAA GCA GGT	7248
Lys Met Phe Thr Glu Arg Gly Val Tyr Val Ile Pro Leu Lys Ala Gly	
2405 2410 2415	
GCA GAG CTA TTT GCC ACT CAG CTA TTG GCT GAA ACT GGC GTG CAG TTG	7296
Ala Glu Leu Phe Ala Thr Gln Leu Leu Ala Glu Thr Gly Val Gln Leu	
2420 2425 2430	



CTC ATT GGT ACG TCA ATG CAA GGT GGC AGC GAC ACT AAA GCA ACT GAG	7344
Leu Ile Gly Thr Ser Met Gln Gly Gly Ser Asp Thr Lys Ala Thr Glu	
2435 2440 2445	
ACT GCT TCT GTA AAA AAG CTT AAT GCG GGT GAG GTG CTA AGT GCA TCG	7392
Thr Ala Ser Val Lys Lys Leu Asn Ala Gly Glu Val Leu Ser Ala Ser	
2450 2455 2460	
CAT CCG CGT GCT GGT GCA CAA AAA ACA CCA CTA CAA GCT GTC ACT GCA	7440
His Pro Arg Ala Gly Ala Gln Lys Thr Pro Leu Gln Ala Val Thr Ala	
2465 2470 2475 2480	
ACG CGT CTG TTA ACC CCA AGT GCC ATG GTC TTC ATT GAA GAT CAC CGC	7488
Thr Arg Leu Leu Thr Pro Ser Ala Met Val Phe Ile Glu Asp His Arg	
2485 2490 2495	
ATT GGC GGT AAC AGT GTG TTG CCA ACG GTA TGC GCC ATC GAC TGG ATG	7536
Ile Gly Gly Asn Ser Val Leu Pro Thr Val Cys Ala Ile Asp Trp Met	
2500 2505 2510	
CGT GAA GCG GCA AGC GAC ATG CTT GGC GCT CAA GTT AAG GTA CTT GAT	7584
Arg Glu Ala Ala Ser Asp Met Leu Gly Ala Gln Val Lys Val Leu Asp	
2515 2520 2525	
TAC AAG CTA TTA AAA GGC ATT GTA TTT GAG ACT GAT GAG CCG CAA GAG	7632
Tyr Lys Leu Leu Lys Gly Ile Val Phe Glu Thr Asp Glu Pro Gln Glu	
2530 2535 2540	
TTA ACA CTT GAG CTA ACG CCA GAC GAT TCA GAC GAA GCT ACG CTA CAA	7680
Leu Thr Leu Glu Leu Thr Pro Asp Asp Ser Asp Glu Ala Thr Leu Gln	
2545 2550 2555 2560	
GCA TTA ATC AGC TGT AAT GGG CGT CCG CAA TAC AAG GCG ACG CTT ATC	7728
Ala Leu Ile Ser Cys Asn Gly Arg Pro Gln Tyr Lys Ala Thr Leu Ile	
2565 2570 2575	
AGT GAT AAT GCC GAT ATT AAG CAA CTT AAC AAG CAG TTT GAT TTA AGC	7776
Ser Asp Asn Ala Asp Ile Lys Gln Leu Asn Lys Gln Phe Asp Leu Ser	
2580 2585 2590	
GCT AAG GCG ATT ACC ACA GCA AAA GAG CTT TAT AGC AAC GGC ACC TTG	7824
Ala Lys Ala Ile Thr Thr Ala Lys Glu Leu Tyr Ser Asn Gly Thr Leu	
2595 2600 2605	



TTC CAC GGT CCG CGT CTA CAA GGG ATC CAA TCT GTA GTG CAG TTC GAT	7872
Phe His Gly Pro Arg Leu Gln Gly Ile Gln Ser Val Val Gln Phe Asp	
2610 2615 2620	
GAT CAA GGC TTA ATT GCT AAA GTC GCT CTG CCT AAG GTT GAA CTT AGC	7920
Asp Gln Gly Leu Ile Ala Lys Val Ala Leu Pro Lys Val Glu Leu Ser	
2625 2630 2635 2640	
GAT TGT GGT GAG TTC TTG CCG CAA ACC CAC ATG GGT GGC AGT CAA CCT	7968
Asp Cys Gly Glu Phe Leu Pro Gln Thr His Met Gly Gly Ser Gln Pro	
2645 2650 2655	
TTT GCT GAG GAC TTG CTA TTA CAA GCT ATG CTG GTT TGG GCT CGC CTT	8016
Phe Ala Glu Asp Leu Leu Leu Gln Ala Met Leu Val Trp Ala Arg Leu	
2660 2665 2670	
AAA ACT GGC TCG GCA AGT TTG CCA TCA AGC ATT GGT GAG TTT ACC TCA	8064
Lys Thr Gly Ser Ala Ser Leu Pro Ser Ser Ile Gly Glu Phe Thr Ser	
2675 2680 2685	
TAC CAA CCA ATG GCC TTT GGT GAA ACT GGT ACC ATA GAG CTT GAA GTG	8112
Tyr Gln Pro Met Ala Phe Gly Glu Thr Gly Thr Ile Glu Leu Glu Val	
2690 2695 2700	
ATT AAG CAC AAC AAA CGC TCA CTT GAA GCG AAT GTT GCG CTA TAT CGT	8160
Ile Lys His Asn Lys Arg Ser Leu Glu Ala Asn Val Ala Leu Tyr Arg	
2705 2710 2715 2720	
GAC AAC GGC GAG TTA AGT GCC ATG TTT AAG TCA GCT AAA ATC ACC ATT	8208
Asp Asn Gly Glu Leu Ser Ala Met Phe Lys Ser Ala Lys Ile Thr Ile	
2725 2730 2735	
AGC AAA AGC TTA AAT TCA GCA TTT TTA CCT GCT GTC TTA GCA AAC GAC	8256
Ser Lys Ser Leu Asn Ser Ala Phe Leu Pro Ala Val Leu Ala Asn Asp	
2740 2745 2750	
AGT GAG GCG AAT	8268
Ser Glu Ala Asn	
2755	

SEQ ID NO : 7

SEQUENCE LENGTH : 2340

SEQUENCE TYPE : Nucleic acid

STRANDNESS : Double strand

TOPOLOGY : Linear



MOLECULE TYPE : Genomic DNA

ORIGINAL SOURCE : Shewanella putrefaciens SCRC-2874

(FERM BP-1625)

SEQUENCE

GTG GAA CAA ACG CCT AAA GCT AGT GCG ATG CCG CTG CGC ATC GCA CTT	48
Val Glu Gln Thr Pro Lys Ala Ser Ala Met Pro Leu Arg Ile Ala Leu	
1 5 10 15	
ATC TTA CTG CCA ACA CCG CAG TTT GAA GTT AAC TCT GTC GAC CAG TCA	96
Ile Leu Leu Pro Thr Pro Gln Phe Glu Val Asn Ser Val Asp Gln Ser	
20 25 30	
GTA TTA GCC AGC TAT CAA ACA CTG CAG CCT GAG CTA AAT GCC CTG CTT	144
Val Leu Ala Ser Tyr Gln Thr Leu Gln Pro Glu Leu Asn Ala Leu Leu	
35 40 45	
AAT AGT GCG CCG ACA CCT GAA ATG CTC AGC ATC ACT ATC TCA GAT GAT	192
Asn Ser Ala Pro Thr Pro Glu Met Leu Ser Ile Thr Ile Ser Asp Asp	
50 55 60	
AGC GAT GCA AAC AGC TTT GAG TCG CAG CTA AAT GCT GCG ACC AAC GCA	240
Ser Asp Ala Asn Ser Phe Glu Ser Gln Leu Asn Ala Ala Thr Asn Ala	
65 70 75 80	
ATT AAC AAT GGC TAT ATC GTC AAG CTT GCT ACG GCA ACT CAC GCT TTG	288
Ile Asn Asn Gly Tyr Ile Val Lys Leu Ala Thr Ala Thr His Ala Leu	
85 90 95	
TTA ATG CTG CCT GCA TTA AAA GCG GCG CAA ATG CGG ATC CAT CCT CAT	336
Leu Met Leu Pro Ala Leu Lys Ala Ala Gln Met Arg Ile His Pro His	
100 105 110	
GCG CAG CTT GCC GCT ATG CAG CAA GCT AAA TCG ACG CCA ATG AGT CAA	384
Ala Gln Leu Ala Ala Met Gln Gln Ala Lys Ser Thr Pro Met Ser Gln	
115 120 125	
GTA TCT GGT GAG CTA AAG CTT GGC GCT AAT GCG CTA AGC CTA GCT CAG	432
Val Ser Gly Glu Leu Lys Leu Gly Ala Asn Ala Leu Ser Leu Ala Gln	
130 135 140	
ACT AAT GCG CTG TCT CAT GCT TTA AGC CAA GCC AAG CGT AAC TTA ACT	480
Thr Asn Ala Leu Ser His Ala Leu Ser Gln Ala Lys Arg Asn Leu Thr	
145 150 155 160	



GAT GTC AGC GTG AAT GAG TGT TTT GAG AAC CTC AAA AGT GAA CAG CAG	528
Asp Val Ser Val Asn Glu Cys Phe Glu Asn Leu Lys Ser Glu Gln Gln	
165 170 175	
TTC ACA GAG GTT TAT TCG CTT ATT CAG CAA CTT GCT AGC CGC ACC CAT	576
Phe Thr Glu Val Tyr Ser Leu Ile Gln Gln Leu Ala Ser Arg Thr His	
180 185 190	
GTG AGA AAA GAG GTT AAT CAA GGT GTG GAA CTT GGC CCT AAA CAA GCC	624
Val Arg Lys Glu Val Asn Gln Gly Val Glu Leu Gly Pro Lys Gln Ala	
195 200 205	
AAA AGC CAC TAT TGG TTT AGC GAA TTT CAC CAA AAC CGT GTT GCT GCC	672
Lys Ser His Tyr Trp Phe Ser Glu Phe His Gln Asn Arg Val Ala Ala	
210 215 220	
ATC AAC TTT ATT AAT GGC CAA CAA GCA ACC AGC TAT GTG CTT ACT CAA	720
Ile Asn Phe Ile Asn Gly Gln Gln Ala Thr Ser Tyr Val Leu Thr Gln	
225 230 235 240	
GGT TCA GGA TTG TTA GCT GCG AAA TCA ATG CTA AAC CAG CAA AGA TTA	768
Gly Ser Gly Leu Leu Ala Ala Lys Ser Met Leu Asn Gln Gln Arg Leu	
245 250 255	
ATG TTT ATC TTG CCG GGT AAC AGT CAG CAA CAA ATA ACC GCA TCA ATA	816
Met Phe Ile Leu Pro Gly Asn Ser Gln Gln Gln Ile Thr Ala Ser Ile	
260 265 270	
ACT CAG TTA ATG CAG CAA TTA GAG CGT TTG CAG GTA ACT GAG GTT AAT	864
Thr Gln Leu Met Gln Gln Leu Glu Arg Leu Gln Val Thr Glu Val Asn	
275 280 285	
GAG CTT TCT CTA GAA TGC CAA CTA GAG CTG CTC AGC ATA ATG TAT GAC	912
Glu Leu Ser Leu Glu Cys Gln Leu Glu Leu Leu Ser Ile Met Tyr Asp	
290 295 300	
AAC TTA GTC AAC GCA GAC AAA CTC ACT ACT CGC GAT AGT AAG CCC GCT	960
Asn Leu Val Asn Ala Asp Lys Leu Thr Thr Arg Asp Ser Lys Pro Ala	
305 310 315 320	
TAT CAG GCT GTG ATT CAA GCA AGC TCT GTT AGC GCT GCA AAG CAA GAG	1008
Tyr Gln Ala Val Ile Gln Ala Ser Ser Val Ser Ala Ala Lys Gln Glu	
325 330 335	



TTA AGC GCG CTT AAC GAT GCA CTC ACA GCG CTG TTT GCT GAG CAA ACA	1056
Leu Ser Ala Leu Asn Asp Ala Leu Thr Ala Leu Phe Ala Glu Gln Thr	
340 345 350	
AAC GCC ACA TCA ACG AAT AAA GGC TTA ATC CAA TAC AAA ACA CCG GCG	1104
Asn Ala Thr Ser Thr Asn Lys Gly Leu Ile Gln Tyr Lys Thr Pro Ala	
355 360 365	
GGC AGT TAC TTA ACC CTA ACA CCG CTT GGC AGC AAC AAT GAC AAC GCC	1152
Gly Ser Tyr Leu Thr Leu Thr Pro Leu Gly Ser Asn Asn Asp Asn Ala	
370 375 380	
CAA GCG GGT CTT GCT TTT GTC TAT CCG GGT GTG GGA ACG GTT TAC GCC	1200
Gln Ala Gly Leu Ala Phe Val Tyr Pro Gly Val Gly Thr Val Tyr Ala	
385 390 395 400	
GAT ATG CTT AAT GAG CTG CAT CAG TAC TTC CCT GCG CTT TAC GCC AAA	1248
Asp Met Leu Asn Glu Leu His Gln Tyr Phe Pro Ala Leu Tyr Ala Lys	
405 410 415	
CTT GAG CGT GAA GGC GAT TTA AAG GCG ATG CTA CAA GCA GAA GAT ATC	1296
Leu Glu Arg Glu Gly Asp Leu Lys Ala Met Leu Gln Ala Glu Asp Ile	
420 425 430	
TAT CAT CTT GAC CCT AAA CAT GCT GCC CAA ATG AGC TTA GGT GAC TTA	1344
Tyr His Leu Asp Pro Lys His Ala Ala Gln Met Ser Leu Gly Asp Leu	
435 440 445	
GCC ATT GCT GGC GTG GGG AGC AGC TAC CTG TTA ACT CAG CTG CTC ACC	1392
Ala Ile Ala Gly Val Gly Ser Ser Tyr Leu Leu Thr Gln Leu Leu Thr	
450 455 460	
GAT GAG TTT AAT ATT AAG CCT AAT TTT GCA TTA GGT TAC TCA ATG GGT	1440
Asp Glu Phe Asn Ile Lys Pro Asn Phe Ala Leu Gly Tyr Ser Met Gly	
465 470 475 480	
GAA GCA TCA ATG TGG GCA AGC TTA GGC GTA TGG CAA AAC CCG CAT GCG	1488
Glu Ala Ser Met Trp Ala Ser Leu Gly Val Trp Gln Asn Pro His Ala	
485 490 495	
CTG ATC AGC AAA ACC CAA ACC GAC CCG CTA TTT ACT TCT GCT ATT TCC	1536
Leu Ile Ser Lys Thr Gln Thr Asp Pro Leu Phe Thr Ser Ala Ile Ser	
500 505 510	



GGC AAA TTG ACC GCG GTT AGA CAA GCT TGG CAG CTT GAT GAT ACC GCA	1584
Gly Lys Leu Thr Ala Val Arg Gln Ala Trp Gln Leu Asp Asp Thr Ala	
515 520 525	
GCG GAA ATC CAG TGG AAT AGC TTT GTG GTT AGA AGT GAA GCA GCG CCG	1632
Ala Glu Ile Gln Trp Asn Ser Phe Val Val Arg Ser Glu Ala Ala Pro	
530 535 540	
ATT GAA GCC TTG CTA AAA GAT TAC CCA CAC GCT TAC CTC GCG ATT ATT	1680
Ile Glu Ala Leu Leu Lys Asp Tyr Pro His Ala Tyr Leu Ala Ile Ile	
545 550 555 560	
CAA GGG GAT ACC TGC GTA ATC GCT GGC TGT GAA ATC CAA TGT AAA GCG	1728
Gln Gly Asp Thr Cys Val Ile Ala Gly Cys Glu Ile Gln Cys Lys Ala	
565 570 575	
CTA CTT GCA GCA CTG GGT AAA CGC GGT ATT GCA GCT AAT CGT GTA ACG	1776
Leu Leu Ala Ala Leu Gly Lys Arg Gly Ile Ala Ala Asn Arg Val Thr	
580 585 590	
GCG ATG CAT ACG CAG CCT GCG ATG CAA GAG CAT CAA AAT GTG ATG GAT	1824
Ala Met His Thr Gln Pro Ala Met Gln Glu His Gln Asn Val Met Asp	
595 600 605	
TTT TAT CTG CAA CCG TTA AAA GCA GAG CTT CCT AGT GAA ATA AGC TTT	1872
Phe Tyr Leu Gln Pro Leu Lys Ala Glu Leu Pro Ser Glu Ile Ser Phe	
610 615 620	
ATC AGC GCC GCT GAT TTA ACT GCC AAG CAA ACG GTG AGT GAG CAA GCA	1920
Ile Ser Ala Ala Asp Leu Thr Ala Lys Gln Thr Val Ser Glu Gln Ala	
625 630 635 640	
CTT AGC AGC CAA GTC GTT GCT CAG TCT ATT GCC GAC ACC TTC TGC CAA	1968
Leu Ser Ser Gln Val Val Ala Gln Ser Ile Ala Asp Thr Phe Cys Gln	
645 650 655	
ACC TTG GAC TTT ACC GCG CTA GTA CAT CAC GCC CAA CAT CAA GGC GCT	2016
Thr Leu Asp Phe Thr Ala Leu Val His His Ala Gln His Gln Gly Ala	
660 665 670	
AAG CTG TTT GTT GAA ATT GGC GCG GAT AGA CAA AAC TGC ACC TTG ATA	2064
Lys Leu Phe Val Glu Ile Gly Ala Asp Arg Gln Asn Cys Thr Leu Ile	
675 680 685	



GAC AAG ATT GTT AAA CAA GAT GGT GCC AGC AGT GTA CAA CAT CAA CCT	2112
Asp Lys Ile Val Lys Gln Asp Gly Ala Ser Ser Val Gln His Gln Pro	
690 695 700	
TGT TGC ACA GTG CCT ATG AAC GCA AAA GGT AGC CAA GAT ATT ACC AGC	2160
Cys Cys Thr Val Pro Met Asn Ala Lys Gly Ser Gln Asp Ile Thr Ser	
705 710 715 720	
GTG ATT AAA GCG CTT GGC CAA TTA ATT AGC CAT CAG GTG CCA TTA TCG	2208
Val Ile Lys Ala Leu Gly Gln Leu Ile Ser His Gln Val Pro Leu Ser	
725 730 735	
GTG CAA CCA TTT ATT GAT GGA CTC AAG CGC GAG CTA ACA CTT TGC CAA	2256
Val Gln Pro Phe Ile Asp Gly Leu Lys Arg Glu Leu Thr Leu Cys Gln	
740 745 750	
TTG ACC AGC CAA CAG CTG GCA GCA CAT GCA AAT GTT GAC AGC AAG TTT	2304
Leu Thr Ser Gln Gln Leu Ala Ala His Ala Asn Val Asp Ser Lys Phe	
755 760 765	
GAG TCT AAC CAA GAC CAT TTA CTT CAA GGG GAA GTC	2340
Glu Ser Asn Gln Asp His Leu Leu Gln Gly Glu Val	
770 775 780	

SEQ ID NO : 8

SEQUENCE LENGTH : 6012

SEQUENCE TYPE : Nucleic acid

STRANDNESS : Double strand

TOPOLOGY : Linear

MOLECULE TYPE : Genomic DNA

ORIGINAL SOURCE : Shewanella putrefaciens SCRC-2874

(FERM BP-1625)

SEQUENCE

ATG TCA TTA CCA GAC AAT GCT TCT AAC CAC CTT TCT GCC AAC CAG AAA	48
Met Ser Leu Pro Asp Asn Ala Ser Asn His Leu Ser Ala Asn Gln Lys	
1 5 10 15	
GCG GCA TCT CAG GCA AGT AAA ACC AGT AAG CAA AGC AAA ATC GCC ATT	96
Gly Ala Ser Gln Ala Ser Lys Thr Ser Lys Gln Ser Lys Ile Ala Ile	
20 25 30	



GTC GGT TTA GCC ACT CTG TAT CCA GAC GCT AAA ACC CCG CAA GAA TTT	144
Val Gly Leu Ala Thr Leu Tyr Pro Asp Ala Lys Thr Pro Gln Glu Phe	
35 40 45	
TGG CAG AAT TTG CTG GAT AAA CGC GAC TCT CGC AGC ACC TTA ACT AAC	192
Trp Gln Asn Leu Leu Asp Lys Arg Asp Ser Arg Ser Thr Leu Thr Asn	
50 55 60	
GAA AAA CTC GGC GCT AAC AGC CAA GAT TAT CAA GGT GTG CAA GGC CAA	240
Glu Lys Leu Gly Ala Asn Ser Gln Asp Tyr Gln Gly Val Gln Gly Gln	
65 70 75 80	
TCT GAC CGT TTT TAT TGT AAT AAA GGC GGC TAC ATT GAG AAC TTC AGC	288
Ser Asp Arg Phe Tyr Cys Asn Lys Gly Gly Tyr Ile Glu Asn Phe Ser	
85 90 95	
TTT AAT GCT GCA GGC TAC AAA TTG CCG GAG CAA AGC TTA AAT GGC TTG	336
Phe Asn Ala Ala Gly Tyr Lys Leu Pro Glu Gln Ser Leu Asn Gly Leu	
100 105 110	
GAC GAC AGC TTC CTT TGG GCG CTC GAT ACT AGC CGT AAC GCA CTA ATT	384
Asp Asp Ser Phe Leu Trp Ala Leu Asp Thr Ser Arg Asn Ala Leu Ile	
115 120 125	
GAT GCT GGT ATT GAT ATC AAC GGC GCT GAT TTA AGC CGC GCA GGT GTA	432
Asp Ala Gly Ile Asp Ile Asn Gly Ala Asp Leu Ser Arg Ala Gly Val	
130 135 140	
GTC ATG GGC GCG CTG TCG TTC CCA ACT ACC GGC TCA AAC GAT CTG TTT	480
Val Met Gly Ala Leu Ser Phe Pro Thr Thr Arg Ser Asn Asp Leu Phe	
145 150 155 160	
TTG CCA ATT TAT CAC AGC GCC GTT GAA AAA GCC CTG CAA GAT AAA CTA	528
Leu Pro Ile Tyr His Ser Ala Val Glu Lys Ala Leu Gln Asp Lys Leu	
165 170 175	
GGC GTA AAG GCA TTT AAG CTA AGC CCA ACT AAT GCT CAT ACC GCT CGC	576
Gly Val Lys Ala Phe Lys Leu Ser Pro Thr Asn Ala His Thr Ala Arg	
180 185 190	
GCG GCA AAT GAG AGC AGC CTA AAT GCA GCC AAT GGT GCC ATT GCC CAT	624
Ala Ala Asn Glu Ser Ser Leu Asn Ala Ala Asn Gly Ala Ile Ala His	
195 200 205	



AAC AGC TCA AAA GTG GTG GCC GAT GCA CTT GGC CTT GGC GGC GCA CAA	672
Asn Ser Ser Lys Val Val Ala Asp Ala Leu Gly Leu Gly Gly Ala Gln	
210 215 220	
CTA AGC CTA GAT GCT GCC TGT GCT AGT TCG GTT TAC TCA TTA AAG CTT	720
Leu Ser Leu Asp Ala Ala Cys Ala Ser Ser Val Tyr Ser Leu Lys Leu	
225 230 235 240	
GCC TGC GAT TAC CTA AGC ACT GGC AAA GCC GAT ATC ATG CTA GCA GGC	768
Ala Cys Asp Tyr Leu Ser Thr Gly Lys Ala Asp Ile Met Leu Ala Gly	
245 250 255	
GCA GTA TCT GGC GCG GAT CCT TTC TTT ATT AAT ATG GGA TTC TCA ATC	816
Ala Val Ser Gly Ala Asp Pro Phe Phe Ile Asn Met Gly Phe Ser Ile	
260 265 270	
TTC CAC GCC TAC CCA GAC CAT GGT ATC TCA GTA CCG TTT GAT GCC AGC	864
Phe His Ala Tyr Pro Asp His Gly Ile Ser Val Pro Phe Asp Ala Ser	
275 280 285	
AGT AAA GGT TTG TTT GCT GGC GAA GGC GCT GGC GTA TTA GTG CTT AAA	912
Ser Lys Gly Leu Phe Ala Gly Glu Gly Ala Gly Val Leu Val Leu Lys	
290 295 300	
CGT CTT GAA GAT GCC GAG CGC GAC AAT GAC AAA ATC TAT GCG GTT GTT	960
Arg Leu Glu Asp Ala Glu Arg Asp Asn Asp Lys Ile Tyr Ala Val Val	
305 310 315 320	
AGC GGC GTA GGT CTA TCA AAC GAC GGT AAA GGC CAG TTT GTA TTA AGC	1008
Ser Gly Val Gly Leu Ser Asn Asp Gly Lys Gly Gln Phe Val Leu Ser	
325 330 335	
CCT AAT CCA AAA GGT CAG GTG AAG GCC TTT GAA CGT GCT TAT GCT GCC	1056
Pro Asn Pro Lys Gly Gln Val Lys Ala Phe Glu Arg Ala Tyr Ala Ala	
340 345 350	
AGT GAC ATT GAG CCA AAA GAC ATT GAA GTG ATT GAG TGC CAC GCA ACA	1104
Ser Asp Ile Glu Pro Lys Asp Ile Glu Val Ile Glu Cys His Ala Thr	
355 360 365	
GGC ACA CCG CTT GGC GAT AAA ATT GAG CTC ACT TCA ATG GAA ACC TTC	1152
Gly Thr Pro Leu Gly Asp Lys Ile Glu Leu Thr Ser Met Glu Thr Phe	
370 375 380	





TTT GGC TTA GCA TCT GCG CCA AAA GGT GCT TAT GTT GAT AAC TTC GAG	1728
Phe Gly Leu Ala Ser Ala Pro Lys Gly Ala Tyr Val Asp Asn Phe Glu	
565 570 575	
CTG GAC TTT TTA CGC TTT AAA CTG CCG CCA AAC GAA GAT GAC CGT TTG	1776
Leu Asp Phe Leu Arg Phe Lys Leu Pro Pro Asn Glu Asp Asp Arg Leu	
580 585 590	
ATC TCA CAG CAG CTA ATG CTA ATG CGA GTA ACA GAC GAA GCC ATT CGT	1824
Ile Ser Gln Gln Leu Met Leu Met Arg Val Thr Asp Glu Ala Ile Arg	
595 600 605	
GAT GCC AAG CTT GAG CCG GGG CAA AAA GTA GCT GTA TTA GTG GCA ATG	1872
Asp Ala Lys Leu Glu Pro Gly Gln Lys Val Ala Val Leu Val Ala Met	
610 615 620	
GAA ACT GAG CTT GAA CTG CAT CAG TTC CGC GGC CGG GTT AAC TTG CAT	1920
Glu Thr Glu Leu Glu Leu His Gln Phe Arg Gly Arg Val Asn Leu His	
625 630 635 640	
ACT CAA TTA GCG CAA AGT CTT GCC GCC ATG GGC GTG AGT TTA TCA ACG	1968
Thr Gln Leu Ala Gln Ser Leu Ala Ala Met Gly Val Ser Leu Ser Thr	
645 650 655	
GAT GAA TAC CAA GCG CTT GAA GCC ATC GCC ATG GAC AGC GTG CTT GAT	2016
Asp Glu Tyr Gln Ala Leu Glu Ala Ile Ala Met Asp Ser Val Leu Asp	
660 665 670	
GCT GCC AAG CTC AAT CAG TAC ACC AGC TTT ATT GGT AAT ATT ATG GCG	2064
Ala Ala Lys Leu Asn Gln Tyr Thr Ser Phe Ile Gly Asn Ile Met Ala	
675 680 685	
TCA CGC GTG GCG TCA CTA TGG GAC TTT AAT GGC CCA GCC TTC ACT ATT	2112
Ser Arg Val Ala Ser Leu Trp Asp Phe Asn Gly Pro Ala Phe Thr Ile	
690 695 700	
TCA GCA GCA GAG CAA TCT GTG AGC CGC TGT ATC GAT GTG GCG CAA AAC	2160
Ser Ala Ala Glu Gln Ser Val Ser Arg Cys Ile Asp Val Ala Gln Asn	
705 710 715 720	
CTC ATC ATG GAG GAT AAC CTA GAT GCG GTG GTG ATT GCA GCG GTC GAT	2208
Leu Ile Met Glu Asp Asn Leu Asp Ala Val Val Ile Ala Ala Val Asp	
725 730 735	



CTC TCT GGT AGC TTT GAG CAA GTC ATT CTT AAA AAT GCC ATT GCA CCT	2256
Leu Ser Gly Ser Phe Glu Gln Val Ile Leu Lys Asn Ala Ile Ala Pro	
740 745 750	
GTA GCC ATT GAG CCA AAC CTC GAA GCA AGC CTT AAT CCA ACA TCA GCA	2304
Val Ala Ile Glu Pro Asn Leu Glu Ala Ser Leu Asn Pro Thr Ser Ala	
755 760 765	
AGC TGG AAT GTC GGT GAA GGT GCT GGC GCG GTC GTG CTT GTT AAA AAT	2352
Ser Trp Asn Val Gly Glu Gly Ala Gly Ala Val Val Leu Val Lys Asn	
770 775 780	
GAA GCT ACA TCG GGC TGC TCA TAC GGC CAA ATT GAT GCA CTT GGC TTT	2400
Glu Ala Thr Ser Gly Cys Ser Tyr Gly Gln Ile Asp Ala Leu Gly Phe	
785 790 795 800	
GCT AAA ACT GCC GAA ACA GCG TTG GCT ACC GAC AAG CTA CTG AGC CAA	2448
Ala Lys Thr Ala Glu Thr Ala Leu Ala Thr Asp Lys Leu Leu Ser Gln	
805 810 815	
ACT GCC ACA GAC TTT AAT AAG GTT AAA GTG ATT GAA ACT ATG GCA GCG	2496
Thr Ala Thr Asp Phe Asn Lys Val Lys Val Ile Glu Thr Met Ala Ala	
820 825 830	
CCT GCT AGC CAA ATT CAA TTA GCG CCA ATA GTT AGC TCT CAA GTG ACT	2544
Pro Ala Ser Gln Ile Gln Leu Ala Pro Ile Val Ser Ser Gln Val Thr	
835 840 845	
CAC ACT GCT GCA GAG CAG CGT GTT GGT CAC TGC TTT GCT GCA GCG GGT	2592
His Thr Ala Ala Glu Gln Arg Val Gly His Cys Phe Ala Ala Ala Gly	
850 855 860	
ATG GCA AGC CTA TTA CAC GGC TTA CTT AAC TTA AAT ACT GTA GCC CAA	2640
Met Ala Ser Leu Leu His Gly Leu Leu Asn Leu Asn Thr Val Ala Gln	
865 870 875 880	
ACC AAT AAA GCC AAT TGC GCG CTT ATC AAC AAT ATC AGT GAA AAC CAA	2688
Thr Asn Lys Ala Asn Cys Ala Leu Ile Asn Asn Ile Ser Glu Asn Gln	
885 890 895	
TTA TCA CAG CTG TTG ATT AGC CAA ACA GCG AGC GAA CAA CAA GCA TTA	2736
Leu Ser Gln Leu Leu Ile Ser Gln Thr Ala Ser Glu Gln Gln Ala Leu	
900 905 910	



ACC GCG CGT TTA AGC AAT GAG CTT AAA TCC GAT GCT AAA CAC CAA CTG	2784
Thr Ala Arg Leu Ser Asn Glu Leu Lys Ser Asp Ala Lys His Gln Leu	
915 920 925	
GTT AAG CAA GTC ACC TTA GGT GGC CGT GAT ATC TAC CAG CAT ATT GTT	2832
Val Lys Gln Val Thr Leu Gly Gly Arg Asp Ile Tyr Gln His Ile Val	
930 935 940	
GAT ACA CCG CTT GCA AGC CTT GAA AGC ATT ACT CAG AAA TTG GCG CAA	2880
Asp Thr Pro Leu Ala Ser Leu Glu Ser Ile Thr Gln Lys Leu Ala Gln	
945 950 955 960	
GCG ACA GCA TCG ACA GTG GTC AAC CAA GTT AAA CCT ATT AAG GCC GCT	2928
Ala Thr Ala Ser Thr Val Val Asn Gln Val Lys Pro Ile Lys Ala Ala	
965 970 975	
GGC TCA GTC GAA ATG GCT AAC TCA TTC GAA ACG GAA AGC TCA GCA GAG	2976
Gly Ser Val Glu Met Ala Asn Ser Phe Glu Thr Glu Ser Ser Ala Glu	
980 985 990	
CCA CAA ATA ACA ATT GCA GCA CAA CAG ACT GCA AAC ATT GGC GTC ACC	3024
Pro Gln Ile Thr Ile Ala Ala Gln Gln Thr Ala Asn Ile Gly Val Thr	
995 1000 1005	
GCT CAG GCA ACC AAA CGT GAA TTA GGT ACC CCA CCA ATG ACA ACA AAT	3072
Ala Gln Ala Thr Lys Arg Glu Leu Gly Thr Pro Pro Met Thr Thr Asn	
1010 1015 1020	
ACC ATT GCT AAT ACA GCA AAT AAT TTA GAC AAG ACT CTT GAG ACT GTT	3120
Thr Ile Ala Asn Thr Ala Asn Asn Leu Asp Lys Thr Leu Glu Thr Val	
1025 1030 1035 1040	
GCT GGC AAT ACT GTT GCT AGC AAG GTT GGC TCT GGC GAC ATA GTC AAT	3168
Ala Gly Asn Thr Val Ala Ser Lys Val Gly Ser Gly Asp Ile Val Asn	
1045 1050 1055	
TTT CAA CAG AAC CAA CAA TTG GCT CAA CAA GCT CAC CTC GCC TTT CTT	3216
Phe Gln Gln Asn Gln Gln Leu Ala Gln Gln Ala His Leu Ala Phe Leu	
1060 1065 1070	
GAA AGC CGC AGT GCG GGT ATG AAG GTG GCT GAT GCT TTA TTG AAG CAA	3264
Glu Ser Arg Ser Ala Gly Met Lys Val Ala Asp Ala Leu Leu Lys Gln	
1075 1080 1085	



CAG CTA GCT CAA GTA ACA GGC CAA ACT ATC GAT AAT CAG GCC CTC GAT	3312
Gln Leu Ala Gln Val Thr Gly Gln Thr Ile Asp Asn Gln Ala Leu Asp	
1090 1095 1100	
ACT CAA GCC GTC GAT ACT CAA ACA AGC GAG AAT GTA GCG ATT GCC GCA	3360
Thr Gln Ala Val Asp Thr Gln Thr Ser Glu Asn Val Ala Ile Ala Ala	
1105 1110 1115 1120	
GAA TCA CCA GTT CAA GTT ACA ACA CCT GTT CAA GTT ACA ACA CCT GTT	3408
Glu Ser Pro Val Gln Val Thr Thr Pro Val Gln Val Thr Thr Pro Val	
1125 1130 1135	
CAA ATC AGT GTT GTG GAG TTA AAA CCA GAT CAC GCT AAT GTG CCA CCA	3456
Gln Ile Ser Val Val Glu Leu Lys Pro Asp His Ala Asn Val Pro Pro	
1140 1145 1150	
TAC ACG CCG CCA GTG CCT GCA TTA AAG CCG TGT ATC TGG AAC TAT GCC	3504
Tyr Thr Pro Pro Val Pro Ala Leu Lys Pro Cys Ile Trp Asn Tyr Ala	
1155 1160 1165	
GAT TTA GTT GAG TAC GCA GAA GGC GAT ATC GCC AAG GTA TTT GGC AGT	3552
Asp Leu Val Glu Tyr Ala Glu Gly Asp Ile Ala Lys Val Phe Gly Ser	
1170 1175 1180	
GAT TAT GCC ATT ATC GAC AGC TAC TCG CGC CGC GTA CGT CTA CCG ACC	3600
Asp Tyr Ala Ile Ile Asp Ser Tyr Ser Arg Arg Val Arg Leu Pro Thr	
1185 1190 1195 1200	
ACT GAC TAC CTG TTG GTA TCG CGC GTG ACC AAA CTT GAT GCG ACC ATC	3648
Thr Asp Tyr Leu Leu Val Ser Arg Val Thr Lys Leu Asp Ala Thr Ile	
1205 1210 1215	
AAT CAA TTT AAG CCA TGC TCA ATG ACC ACT GAG TAC GAC ATC CCT GTT	3696
Asn Gln Phe Lys Pro Cys Ser Met Thr Thr Glu Tyr Asp Ile Pro Val	
1220 1225 1230	
GAT GCG CCG TAC TTA GTA GAC GGA CAA ATC CCT TGG GCG GTA GCA GTA	3744
Asp Ala Pro Tyr Leu Val Asp Gly Gln Ile Pro Trp Ala Val Ala Val	
1235 1240 1245	
GAA TCA GGC CAA TGT GAC TTG ATG CTT ATT AGC TAT CTC GGT ATC GAC	3792
Glu Ser Gly Gln Cys Asp Leu Met Leu Ile Ser Tyr Leu Gly Ile Asp	
1250 1255 1260	



TTT GAG AAC AAA GGC GAG CGG GTT TAT CGA CTA CTC GAT TGT ACC CTC	3840
Phe Glu Asn Lys Gly Glu Arg Val Tyr Arg Leu Leu Asp Cys Thr Leu	
1265 1270 1275 1280	
ACC TTC CTA GGC GAC TTG CCA CGT GGC GGA GAT ACC CTA CGT TAC GAC	3888
Thr Phe Leu Gly Asp Leu Pro Arg Gly Gly Asp Thr Leu Arg Tyr Asp	
1285 1290 1295	
ATT AAG ATC AAT AAC TAT GCT CGC AAC GGC GAC ACC CTG CTG TTC TTC	3936
Ile Lys Ile Asn Asn Tyr Ala Arg Asn Gly Asp Thr Leu Leu Phe Phe	
1300 1305 1310	
TTC TCG TAT GAG TGT TTT GTT GGC GAC AAG ATG ATC CTC AAG ATG GAT	3984
Phe Ser Tyr Glu Cys Phe Val Gly Asp Lys Met Ile Leu Lys Met Asp	
1315 1320 1325	
GGC GGC TGC GCT GGC TTC TTC ACT GAT GAA GAG CTT GCC GAC GGT AAA	4032
Gly Gly Cys Ala Gly Phe Phe Thr Asp Glu Glu Leu Ala Asp Gly Lys	
1330 1335 1340	
GGC GTG ATT CGC ACA GAA GAA GAG ATT AAA GCT CGC AGC CTA GTG CAA	4080
Gly Val Ile Arg Thr Glu Glu Glu Ile Lys Ala Arg Ser Leu Val Gln	
1345 1350 1355 1360	
AAG CAA CGC TTT AAT CCG TTA CTA GAT TGT CCT AAA ACC CAA TTT AGT	4128
Lys Gln Arg Phe Asn Pro Leu Leu Asp Cys Pro Lys Thr Gln Phe Ser	
1365 1370 1375	
TAT GGT GAT ATT CAT AAG CTA TTA ACT GCT GAT ATT GAG GGT TGT TTT	4176
Tyr Gly Asp Ile His Lys Leu Leu Thr Ala Asp Ile Glu Gly Cys Phe	
1380 1385 1390	
GGC CCA AGC CAC AGT GGC GTC CAC CAG CCG TCA CTT TGT TTC GCA TCT	4224
Gly Pro Ser His Ser Gly Val His Gln Pro Ser Leu Cys Phe Ala Ser	
1395 1400 1405	
GAA AAA TTC TTG ATG ATT GAA CAA GTC AGC AAG GTT GAT CGC ACT GGC	4272
Glu Lys Phe Leu Met Ile Glu Gln Val Ser Lys Val Asp Arg Thr Gly	
1410 1415 1420	
GGT ACT TGG GGA CTT GGC TTA ATT GAG GGT CAT AAG CAG CTT GAA GCA	4320
Gly Thr Trp Gly Leu Gly Leu Ile Glu Gly His Lys Gln Leu Glu Ala	
1425 1430 1435 1440	



GAC CAC TGG TAC TTC CCA TGT CAT TTC AAG GGC GAC CAA GTG ATG GCT	4368
Asp His Trp Tyr Phe Pro Cys His Phe Lys Gly Asp Gln Val Met Ala	
1445 1450 1455	
GGC TCG CTA ATG GCT GAA GGT TGT GGC CAG TTA TTG CAG TTC TAT ATG	4416
Gly Ser Leu Met Ala Glu Gly Cys Gly Gln Leu Leu Gln Phe Tyr Met	
1460 1465 1470	
CTG CAC CTT GGT ATG CAT ACC CAA ACT AAA AAT GGT CGT TTC CAA CCT	4464
Leu His Leu Gly Met His Thr Gln Thr Lys Asn Gly Arg Phe Gln Pro	
1475 1480 1485	
CTT GAA AAC GCC TCA CAG CAA GTA CGC TGT CGC GGT CAA GTG CTG CCA	4512
Leu Glu Asn Ala Ser Gln Gln Val Arg Cys Arg Gly Gln Val Leu Pro	
1490 1495 1500	
CAA TCA GGC GTG CTA ACT TAC CGT ATG GAA GTG ACT GAA ATC GGT TTC	4560
Gln Ser Gly Val Leu Thr Tyr Arg Met Glu Val Thr Glu Ile Gly Phe	
1505 1510 1515 1520	
AGT CCA CGC CCA TAT GCT AAA GCT AAC ATC GAT ATC TTG CTT AAT GGC	4608
Ser Pro Arg Pro Tyr Ala Lys Ala Asn Ile Asp Ile Leu Leu Asn Gly	
1525 1530 1535	
AAA GCG GTA GTG GAT TTC CAA AAC CTA GGG GTG ATG ATA AAA GAG GAA	4656
Lys Ala Val Val Asp Phe Gln Asn Leu Gly Val Met Ile Lys Glu Glu	
1540 1545 1550	
GAT GAG TGT ACT CGT TAT CCA CTT TTG ACT GAA TCA ACA ACG GCT AGC	4704
Asp Glu Cys Thr Arg Tyr Pro Leu Leu Thr Glu Ser Thr Thr Ala Ser	
1555 1560 1565	
ACT GCA CAA GTA AAC GCT CAA ACA AGT GCG AAA AAG GTA TAC AAG CCA	4752
Thr Ala Gln Val Asn Ala Gln Thr Ser Ala Lys Lys Val Tyr Lys Pro	
1570 1575 1580	
GCA TCA GTC AAT GCG CCA TTA ATG GCA CAA ATT CCT GAT CTG ACT AAA	4800
Ala Ser Val Asn Ala Pro Leu Met Ala Gln Ile Pro Asp Leu Thr Lys	
1585 1590 1595 1600	
GAG CCA AAC AAG GGC GTT ATT CCG ATT TCC CAT GTT GAA GCA CCA ATT	4848
Glu Pro Asn Lys Gly Val Ile Pro Ile Ser His Val Glu Ala Pro Ile	
1605 1610 1615	



ACG CCA GAC TAC CCG AAC CGT GTA CCT GAT ACA GTG CCA TTC ACG CCG	4896
Thr Pro Asp Tyr Pro Asn Arg Val Pro Asp Thr Val Pro Phe Thr Pro	
1620 1625 1630	
TAT CAC ATG TTT GAG TTT GCT ACA GGC AAT ATC GAA AAC TGT TTC GGG	4944
Tyr His Met Phe Glu Phe Ala Thr Gly Asn Ile Glu Asn Cys Phe Gly	
1635 1640 1645	
CCA GAG TTC TCA ATC TAT CGC GGC ATG ATC CCA CCA CGT ACA CCA TGC	4992
Pro Glu Phe Ser Ile Tyr Arg Gly Met Ile Pro Pro Arg Thr Pro Cys	
1650 1655 1660	
GGT GAC TTA CAA GTG ACC ACA CGT GTG ATT GAA GTT AAC GGT AAG CGT	5040
Gly Asp Leu Gln Val Thr Thr Arg Val Ile Glu Val Asn Gly Lys Arg	
1665 1670 1675 1680	
GGC GAC TTT AAA AAG CCA TCA TCG TGT ATC GCT GAA TAT GAA GTG CCT	5088
Gly Asp Phe Lys Lys Pro Ser Ser Cys Ile Ala Glu Tyr Glu Val Pro	
1685 1690 1695	
GCA GAT GCG TGG TAT TTC GAT AAA AAC AGC CAC GGC GCA GTG ATG CCA	5136
Ala Asp Ala Trp Tyr Phe Asp Lys Asn Ser His Gly Ala Val Met Pro	
1700 1705 1710	
TAT TCA ATT TTA ATG GAG ATC TCA CTG CAA CCT AAC GGC TTT ATC TCA	5184
Tyr Ser Ile Leu Met Glu Ile Ser Leu Gln Pro Asn Gly Phe Ile Ser	
1715 1720 1725	
GGT TAC ATG GGC ACA ACC CTA GGC TTC CCT GGC CTT GAG CTG TTC TTC	5232
Gly Tyr Met Gly Thr Thr Leu Gly Phe Pro Gly Leu Glu Leu Phe Phe	
1730 1735 1740	
CGT AAC TTA GAC GGT AGC GGT GAG TTA CTA CGT GAA GTA GAT TTA CGT	5280
Arg Asn Leu Asp Gly Ser Gly Glu Leu Leu Arg Glu Val Asp Leu Arg	
1745 1750 1755 1760	
GGT AAA ACC ATC CGT AAC GAC TCA CGT TTA TTA TCA ACA GTG ATG GCC	5328
Gly Lys Thr Ile Arg Asn Asp Ser Arg Leu Leu Ser Thr Val Met Ala	
1765 1770 1775	
GGC ACT AAC ATC ATC CAA AGC TTT AGC TTC GAG CTA AGC ACT GAC GGT	5376
Gly Thr Asn Ile Ile Gln Ser Phe Ser Phe Glu Leu Ser Thr Asp Gly	
1780 1785 1790	



GAG CCT TTC TAT CGC GGC ACT GCG GTA TTT GGC TAT TTT AAA GGT GAC	5424
Glu Pro Phe Tyr Arg Gly Thr Ala Val Phe Gly Tyr Phe Lys Gly Asp	
1795 1800 1805	
GCA CTT AAA GAT CAG CTA GGC CTA GAT AAC GGT AAA GTC ACT CAG CCA	5472
Ala Leu Lys Asp Gln Leu Gly Leu Asp Asn Gly Lys Val Thr Gln Pro	
1810 1815 1820	
TGG CAT GTA GCT AAC GGC GTT GCT GCA AGC ACT AAG GTG AAC CTG CTT	5520
Trp His Val Ala Asn Gly Val Ala Ala Ser Thr Lys Val Asn Leu Leu	
1825 1830 1835 1840	
GAT AAG AGC TGC CGT CAC TTT AAT GCG CCA GCT AAC CAG CCA CAC TAT	5568
Asp Lys Ser Cys Arg His Phe Asn Ala Pro Ala Asn Gln Pro His Tyr	
1845 1850 1855	
CGT CTA GCC GGT GGT CAG CTG AAC TTT ATC GAC AGT GTT GAA ATT GTT	5616
Arg Leu Ala Gly Gly Gln Leu Asn Phe Ile Asp Ser Val Glu Ile Val	
1860 1865 1870	
GAT AAT GGC GGC ACC GAA GGT TTA GGT TAC TTG TAT GCC GAG CGC ACC	5664
Asp Asn Gly Gly Thr Glu Gly Leu Gly Tyr Leu Tyr Ala Glu Arg Thr	
1875 1880 1885	
ATT GAC CCA AGT GAT TGG TTC TTC CAG TTC CAC TTC CAC CAA GAT CCG	5712
Ile Asp Pro Ser Asp Trp Phe Phe Gln Phe His Phe His Gln Asp Pro	
1890 1895 1900	
GTT ATG CCA GGC TCC TTA GGT GTT GAA GCA ATT ATT GAA ACC ATG CAA	5760
Val Met Pro Gly Ser Leu Gly Val Glu Ala Ile Ile Glu Thr Met Gln	
1905 1910 1915 1920	
GCT TAC GCT ATT AGT AAA GAC TTG GGC GCA GAT TTC AAA AAT CCT AAG	5808
Ala Tyr Ala Ile Ser Lys Asp Leu Gly Ala Asp Phe Lys Asn Pro Lys	
1925 1930 1935	
TTT GGT CAG ATT TTA TCG AAC ATC AAG TGG AAG TAT CGC GGT CAA ATC	5856
Phe Gly Gln Ile Leu Ser Asn Ile Lys Trp Lys Tyr Arg Gly Gln Ile	
1940 1945 1950	
AAT CCG CTG AAC AAG CAG ATG TCT ATG GAT GTC AGC ATT ACT TCA ATC	5904
Asn Pro Leu Asn Lys Gln Met Ser Met Asp Val Ser Ile Thr Ser Ile	
1955 1960 1965	



AAA GAT GAA GAC GGT AAG AAA GTC ATC ACA GGT AAT GCC AGC TTG AGT 5952
 Lys Asp Glu Asp Gly Lys Lys Val Ile Thr Gly Asn Ala Ser Leu Ser
 1970 1975 1980

AAA GAT GGT CTG CGC ATA TAC GAG GTC TTC GAT ATA GCT ATC AGC ATC 6000
 Lys Asp Gly Leu Arg Ile Tyr Glu Val Phe Asp Ile Ala Ile Ser Ile
 1985 1990 1995 2000

GAA GAA TCT GTA 6012
 Glu Glu Ser Val

SEQ ID NO : 9

SEQUENCE LENGTH : 1629

SEQUENCE TYPE : Nucleic acid

STRANDNESS : Double strand

TOPOLOGY : Linear

MOLECULE TYPE : Genomic DNA

ORIGINAL SOURCE : Shewanella putrefaciens SCRC-2874
 (FERM BP-1625)

SEQUENCE

ATG AAT CCT ACA GCA ACT AAC GAA ATG CTT TCT CCG TGG CCA TGG GCT 48
 Met Asn Pro Thr Ala Thr Asn Glu Met Leu Ser Pro Trp Pro Trp Ala
 1 5 10 15

GTG ACA GAG TCA AAT ATC AGT TTT GAC GTG CAA GTG ATG GAA CAA CAA 96
 Val Thr Glu Ser Asn Ile Ser Phe Asp Val Gln Val Met Glu Gln Gln
 20 25 30

CTT AAA GAT TTT AGC CGG GCA TGT TAC GTG GTC AAT CAT GCC GAC CAC 144
 Leu Lys Asp Phe Ser Arg Ala Cys Tyr Val Val Asn His Ala Asp His
 35 40 45

GGC TTT GGT ATT GCG CAA ACT GCC GAT ATC GTG ACT GAA CAA GCG GCA 192
 Gly Phe Gly Ile Ala Gln Thr Ala Asp Ile Val Thr Glu Gln Ala Ala
 50 55 60

AAC AGC ACA GAT TTA CCT GTT AGT GCT TTT ACT CCT GCA TTA GGT ACC 240
 Asn Ser Thr Asp Leu Pro Val Ser Ala Phe Thr Pro Ala Leu Gly Thr
 65 70 75 80

GAA AGC CTA GGC GAC AAT AAT TTC CGC CGC GTT CAC GGC GTT AAA TAC 288
 Glu Ser Leu Gly Asp Asn Asn Phe Arg Arg Val His Gly Val Lys Tyr
 85 90 95



GCT TAT TAC GCA GGC GCT ATG GCA AAC GGT ATT TCA TCT GAA GAG CTA	336
Ala Tyr Tyr Ala Gly Ala Met Ala Asn Gly Ile Ser Ser Glu Glu Leu	
100 105 110	
GTG ATT GCC CTA GGT CAA GCT GGC ATT TTG TGT GGT TCG TTT GGA GCA	384
Val Ile Ala Leu Gly Gln Ala Gly Ile Leu Cys Gly Ser Phe Gly Ala	
115 120 125	
GCC GGT CTT ATT CCA AGT CGC GTT GAA GCG GCA ATT AAC CGT ATT CAA	432
Ala Gly Leu Ile Pro Ser Arg Val Glu Ala Ala Ile Asn Arg Ile Gln	
130 135 140	
GCA GCG CTG CCA AAT GGC CCT TAT ATG TTT AAC CTT ATC CAT AGT CCT	480
Ala Ala Leu Pro Asn Gly Pro Tyr Met Phe Asn Leu Ile His Ser Pro	
145 150 155 160	
AGC GAG CCA GCA TTA GAG CGT GGC AGC GTA GAG CTA TTT TTA AAG CAT	528
Ser Glu Pro Ala Leu Glu Arg Gly Ser Val Glu Leu Phe Leu Lys His	
165 170 175	
AAG GTA CGC ACC GTT GAA GCA TCA GCT TTC TTA GGT CTA ACA CCA CAA	576
Lys Val Arg Thr Val Glu Ala Ser Ala Phe Leu Gly Leu Thr Pro Gln	
180 185 190	
ATC GTC TAT TAC CGT GCA GCA GGA TTG AGC CGA GAC GCA CAA GGT AAA	624
Ile Val Tyr Tyr Arg Ala Ala Gly Leu Ser Arg Asp Ala Gln Gly Lys	
195 200 205	
GTT GTG GTT GGT AAC AAG GTT ATC GCT AAA GTA AGT CGC ACC GAA GTG	672
Val Val Val Gly Asn Lys Val Ile Ala Lys Val Ser Arg Thr Glu Val	
210 215 220	
GCT GAA AAG TTT ATG ATG CCA GCG CCC GCA AAA ATG CTA CAA AAA CTA	720
Ala Glu Lys Phe Met Met Pro Ala Pro Ala Lys Met Leu Gln Lys Leu	
225 230 235 240	
GTT GAT GAC GGT TCA ATT ACC GCT GAG CAA ATG GAG CTG GCG CAA CTT	768
Val Asp Asp Gly Ser Ile Thr Ala Glu Gln Met Glu Leu Ala Gln Leu	
245 250 255	
GTA CCT ATG GCT GAC GAC ATC ACT GCA GAG GCC GAT TCA GGT GGC CAT	816
Val Pro Met Ala Asp Asp Ile Thr Ala Glu Ala Asp Ser Gly Gly His	
260 265 270	



ACT GAT AAC CGT CCA TTA GTA ACA TTG CTG CCA ACC ATT TTA GCG CTG	864
Thr Asp Asn Arg Pro Leu Val Thr Leu Leu Pro Thr Ile Leu Ala Leu	
275 280 285	
AAA GAA GAA ATT CAA GCT AAA TAC CAA TAC GAC ACT CCT ATT CGT GTC	912
Lys Glu Glu Ile Gln Ala Lys Tyr Gln Tyr Asp Thr Pro Ile Arg Val	
290 295 300	
GGT TGT GGT GGC GGT GTG GGT ACG CCT GAT GCA GCG CTG GCA ACG TTT	960
Gly Cys Gly Gly Gly Val Gly Thr Pro Asp Ala Ala Leu Ala Thr Phe	
305 310 315 320	
AAC ATG GGC GCG GCG TAT ATT GTT ACC GGC TCT ATC AAC CAA GCT TGT	1008
Asn Met Gly Ala Ala Tyr Ile Val Thr Gly Ser Ile Asn Gln Ala Cys	
325 330 335	
GTT GAA GCG GGC GCA AGT GAT CAC ACT CGT AAA TTA CTT GCC ACC ACT	1056
Val Glu Ala Gly Ala Ser Asp His Thr Arg Lys Leu Leu Ala Thr Thr	
340 345 350	
GAA ATG GCC GAT GTG ACT ATG GCA CCA GCT GCA GAT ATG TTC GAG ATG	1104
Glu Met Ala Asp Val Thr Met Ala Pro Ala Ala Asp Met Phe Glu Met	
355 360 365	
GGC GTA AAA CTG CAG GTG GTT AAG CGC GGC ACG CTA TTC CCA ATG CGC	1152
Gly Val Lys Leu Gln Val Val Lys Arg Gly Thr Leu Phe Pro Met Arg	
370 375 380	
GCT AAC AAG CTA TAT GAG ATC TAC ACC CGT TAC GAT TCA ATC GAA GCG	1200
Ala Asn Lys Leu Tyr Glu Ile Tyr Thr Arg Tyr Asp Ser Ile Glu Ala	
385 390 395 400	
ATC CCA TTA GAC GAG CGT GAA AAG CTT GAG AAA CAA GTA TTC CGC TCA	1248
Ile Pro Leu Asp Glu Arg Glu Lys Leu Glu Lys Gln Val Phe Arg Ser	
405 410 415	
AGC CTA GAT GAA ATA TGG GCA GGT ACA GTG GCG CAC TTT AAC GAG CGC	1296
Ser Leu Asp Glu Ile Trp Ala Gly Thr Val Ala His Phe Asn Glu Arg	
420 425 430	
GAC CCT AAG CAA ATC GAA CGC GCA GAG GGT AAC CCT AAG CGT AAA ATG	1344
Asp Pro Lys Gln Ile Glu Arg Ala Glu Gly Asn Pro Lys Arg Lys Met	
435 440 445	



GCA TTG ATT TTC CGT TGG TAC TTA GGT CTT TCT AGT CGC TGG TCA AAC	1392
Ala Leu Ile Phe Arg Trp Tyr Leu Gly Leu Ser Ser Arg Trp Ser Asn	
450 455 460	
TCA GGC GAA GTG GGT CGT GAA ATG GAT TAT CAA ATT TGG GCT GGC CCT	1440
Ser Gly Glu Val Gly Arg Glu Met Asp Tyr Gln Ile Trp Ala Gly Pro	
465 470 475 480	
GCT CTC GGT GCA TTT AAC CAA TGG GCA AAA GGC AGT TAC TTA GAT AAC	1488
Ala Leu Gly Ala Phe Asn Gln Trp Ala Lys Gly Ser Tyr Leu Asp Asn	
485 490 495	
TAT CAA GAC CGA AAT GCC GTC GAT TTG GCA AAG CAC TTA ATG TAC GGC	1536
Tyr Gln Asp Arg Asn Ala Val Asp Leu Ala Lys His Leu Met Tyr Gly	
500 505 510	
GCG GCT TAC TTA AAT CGT ATT AAC TCG CTA ACG GCT CAA GGC GTT AAA	1584
Ala Ala Tyr Leu Asn Arg Ile Asn Ser Leu Thr Ala Gln Gly Val Lys	
515 520 525	
GTG CCA GCA CAG TTA CTT CGC TGG AAG CCA AAC CAA AGA ATG GCC	1629
Val Pro Ala Gln Leu Leu Arg Trp Lys Pro Asn Gln Arg Met Ala	
530 535 540	

SEQ ID NO : 10

SEQUENCE LENGTH : 1575

SEQUENCE TYPE : Nucleic acid

STRANDNESS : Double strand

TOPOLOGY : Linear

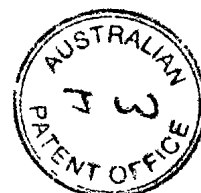
MOLECULE TYPE : Genomic DNA

ORIGINAL SOURCE : *Shewanella putrefaciens* SCRC-2874

(FERM BP-1625)

SEQUENCE

GTG CAA CTA CCA TTA ATT GAG GCC TCA TTA GTT AAA TTA TCT GAG CAA	48
Val Gln Leu Pro Leu Ile Glu Ala Ser Leu Val Lys Leu Ser Glu Gln	
1 5 10 15	
GAG CTC ACC TCT TTA AAT TAC GCT TTT CAG CAA ATG AGA AAG CCA CTA	96
Glu Leu Thr Ser Leu Asn Tyr Ala Phe Gln Gln Met Arg Lys Pro Leu	
20 25 30	



CAA ACC ATT AAT TAC GAC TAT GCG GTG TGG GAC AGA ACC TAC AGC TAT	144
Gln Thr Ile Asn Tyr Asp Tyr Ala Val Trp Asp Arg Thr Tyr Ser Tyr	
35 40 45	
ATG AAA TCA AAC TCA GCG AGC GCT AAA AGG TAC TAT GAA AAA CAT GAG	192
Met Lys Ser Asn Ser Ala Ser Ala Lys Arg Tyr Tyr Glu Lys His Glu	
50 55 60	
TAC CCA GAT GAT ACG TTC AAG AGT TTA AAA GTC GAC GGA GTA TTT ATA	240
Tyr Pro Asp Asp Thr Phe Lys Ser Leu Lys Val Asp Gly Val Phe Ile	
65 70 75 80	
TTC AAC CGT ACA AAT CAG CCA GTT TTT AGT AAA GGT TTT AAT CAT AGA	288
Phe Asn Arg Thr Asn Gln Pro Val Phe Ser Lys Gly Phe Asn His Arg	
85 90 95	
AAT GAT ATA CCG CTG GTC TTT GAA TTA ACT GAC TTT AAA CAA CAT CCA	336
Asn Asp Ile Pro Leu Val Phe Glu Leu Thr Asp Phe Lys Gln His Pro	
100 105 110	
CAA AAC ATC GCA TTA TCT CCA CAA ACC AAA CAG GCA CAC CCA CCG GCA	384
Gln Asn Ile Ala Leu Ser Pro Gln Thr Lys Gln Ala His Pro Pro Ala	
115 120 125	
AGT AAG CCG TTA GAC TCC CCT GAT GAT GTG CCT TCT ACC CAT GGG GTT	432
Ser Lys Pro Leu Asp Ser Pro Asp Asp Val Pro Ser Thr His Gly Val	
130 135 140	
ATC GCC ACA CGA TAC GGT CCA GCA ATT TAT AGC TCT ACC AGC ATT TTA	480
Ile Ala Thr Arg Tyr Gly Pro Ala Ile Tyr Ser Ser Thr Ser Ile Leu	
145 150 155 160	
AAA TCT GAT CGT AGC GGC TCC CAA CTT GGT TAT TTA GTC TTC ATT AGG	528
Lys Ser Asp Arg Ser Gly Ser Gln Leu Gly Tyr Leu Val Phe Ile Arg	
165 170 175	
TTA ATT GAT GAA TGG TTC ATC GCT GAG CTA TCG CAA TAC ACT GCC GCA	576
Leu Ile Asp Glu Trp Phe Ile Ala Glu Leu Ser Gln Tyr Thr Ala Ala	
180 185 190	
GGT GTT GAA ATC GCT ATG GCT GAT GCC GCA GAC GCA CAA TTA GCG AGA	624
Gly Val Glu Ile Ala Met Ala Asp Ala Ala Asp Ala Gln Leu Ala Arg	
195 200 205	



TTA GGC GCA AAC ACT AAG CTT AAT AAA GTA ACC GCT ACA TCC GAA CGG	672
Leu Gly Ala Asn Thr Lys Leu Asn Lys Val Thr Ala Thr Ser Glu Arg	
210 215 220	
TTA ATA ACT AAT GTC GAT GGT AAG CCT CTG TTG AAG TTA GTG CTT TAC	720
Leu Ile Thr Asn Val Asp Gly Lys Pro Leu Leu Lys Leu Val Leu Tyr	
225 230 235 240	
CAT ACC AAT AAC CAA CCG CCG CCG ATG CTA GAT TAC AGT ATA ATA ATT	768
His Thr Asn Asn Gln Pro Pro Pro Met Leu Asp Tyr Ser Ile Ile Ile	
245 250 255	
CTA TTA GTT GAG ATG TCA TTT TTA CTG ATC CTC GCT TAT TTC CTT TAC	816
Leu Leu Val Glu Met Ser Phe Leu Leu Ile Leu Ala Tyr Phe Leu Tyr	
260 265 270	
TCC TAC TTC TTA GTC AGG CCA GTT AGA AAG CTG GCT TCA GAT ATT AAA	864
Ser Tyr Phe Leu Val Arg Pro Val Arg Lys Leu Ala Ser Asp Ile Lys	
275 280 285	
AAA ATG GAT AAA AGT CGT GAA ATT AAA AAG CTA AGG TAT CAC TAC CCT	912
Lys Met Asp Lys Ser Arg Glu Ile Lys Lys Leu Arg Tyr His Tyr Pro	
290 295 300	
ATT ACT GAG CTA GTC AAA GTT GCG ACT CAC TTC AAC GCC CTA ATG GGG	960
Ile Thr Glu Leu Val Lys Val Ala Thr His Phe Asn Ala Leu Met Gly	
305 310 315 320	
ACG ATT CAG GAA CAA ACT AAA CAG CTT AAT GAA CAA GTT TTT ATT GAT	1008
Thr Ile Gln Glu Gln Thr Lys Gln Leu Asn Glu Gln Val Phe Ile Asp	
325 330 335	
AAA TTA ACC AAT ATT CCC AAT CGT CGC GCT TTT GAG CAG CGA CTT GAA	1056
Lys Leu Thr Asn Ile Pro Asn Arg Arg Ala Phe Glu Gln Arg Leu Glu	
340 345 350	
ACC TAT TGC CAA CTG CTA GCC CGG CAA CAA ATT GGC TTT ACT CTC ATC	1104
Thr Tyr Cys Gln Leu Leu Ala Arg Gln Gln Ile Gly Phe Thr Leu Ile	
355 360 365	
ATT GCC GAT GTG GAT CAT TTT AAA GAG TAC AAC GAT ACT CTT GGG CAC	1152
Ile Ala Asp Val Asp His Phe Lys Glu Tyr Asn Asp Thr Leu Gly His	
370 375 380	



CTT	GCT	GGG	GAT	GAA	GCA	TTA	ATA	AAA	GTG	GCA	CAA	ACA	CTA	TCG	CAA	1200
Leu	Ala	Gly	Asp	Glu	Ala	Leu	Ile	Lys	Val	Ala	Gln	Thr	Leu	Ser	Gln	
385					390				395					400		
CAG	TTT	TAC	CGT	GCA	GAA	GAT	ATT	TGT	GCC	CGT	TTT	GGT	GGT	GAA	GAA	1248
Gln	Phe	Tyr	Arg	Ala	Glu	Asp	Ile	Cys	Ala	Arg	Phe	Gly	Gly	Glu	Glu	
				405					410					415		
TTT	ATT	ATG	TTA	TTT	CGA	GAC	ATA	CCT	GAT	GAG	CCC	TTG	CAG	AGA	AAG	1296
Phe	Ile	Met	Leu	Phe	Arg	Asp	Ile	Pro	Asp	Glu	Pro	Leu	Gln	Arg	Lys	
			420					425					430			
CTC	GAT	GCG	ATG	CTG	CAC	TCT	TTT	GCA	GAG	CTC	AAC	CTA	CCT	CAT	CCA	1344
Leu	Asp	Ala	Met	Leu	His	Ser	Phe	Ala	Glu	Leu	Asn	Leu	Pro	His	Pro	
			435					440					445			
AAC	TCA	TCA	ACC	GCT	AAT	TAC	GTT	ACT	GTG	AGC	CTT	GGG	GTT	TGC	ACA	1392
Asn	Ser	Ser	Thr	Ala	Asn	Tyr	Val	Thr	Val	Ser	Leu	Gly	Val	Cys	Thr	
	450						455					460				
GTT	GTT	GCT	GTT	GAT	GAT	TTT	GAA	TTT	AAA	AGT	GAG	TCG	CAT	ATT	ATT	1440
Val	Val	Ala	Val	Asp	Asp	Phe	Glu	Phe	Lys	Ser	Glu	Ser	His	Ile	Ile	
465					470					475					480	
GGC	AGT	CAG	GCT	GCA	TTA	ATC	GCA	GAT	AAG	GCG	CTT	TAT	CAT	GCT	AAA	1488
Gly	Ser	Gln	Ala	Ala	Leu	Ile	Ala	Asp	Lys	Ala	Leu	Tyr	His	Ala	Lys	
				485					490					495		
GCC	TGT	GGT	CGT	AAC	CAG	TTG	TCA	AAA	ACT	ACT	ATT	ACT	GTT	GAT	GAG	1536
Ala	Cys	Gly	Arg	Asn	Gln	Leu	Ser	Lys	Thr	Thr	Ile	Thr	Val	Asp	Glu	
			500					505					510			
ATT	GAG	CAA	TTA	GAA	GCA	AAT	AAA	ATC	GGT	CAT	CAA	GCC				1575
Ile	Glu	Gln	Leu	Glu	Ala	Asn	Lys	Ile	Gly	His	Gln	Ala				
		515					520				525					



THE CLAIMS DEFINING THE INVENTION ARE AS FOLLOWS:

1. An isolated DNA encoding a group of eicosapentaenoic acid synthesis enzymes derived from a microorganism belonging to the genus *Shewanella*.

5

2. An isolated DNA encoding a group of eicosapentaenoic acid synthesis enzymes and having the nucleotide sequence shown in SEQ ID NO: 1.

3. An isolated DNA encoding a group of eicosapentaenoic acid synthesis enzymes having
10 the nucleotide sequence from nucleotide number 5661 to 34631 of SEQ ID NO: 1.

4. An isolated DNA encoding a group of eicosapentaenoic acid synthesis enzymes comprising at least any one of the amino acid sequences set forth in SEQ ID NOS: 4 or 5.

15 5. An isolated DNA encoding a group of eicosapentaenoic acid synthesis enzymes having at least any one of the nucleotide sequences shown in SEQ ID NOS: 4 or 5.

6. An isolated DNA encoding a group of eicosapentaenoic acid synthesis enzymes comprising at least any one of the amino acid sequences selected from the group of amino
20 acid sequences set forth in SEQ ID NOS: 3, 6, 7, 8, 9 and 10.

7. An isolated DNA encoding a group of eicosapentaenoic acid synthesis enzymes having at least any one of the nucleotide sequences selected from the group of nucleotide sequences set forth in SEQ ID NOS: 3, 6, 7, 8, 9 and 10.

25

8. An isolated DNA encoding an enzyme involved in eicosapentaenoic acid synthesis, said enzyme comprising an amino acid sequence selected from the amino acid sequences set forth in SEQ ID NOS: 7 and 8.



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9. An isolated DNA encoding an enzyme involved in eicosapentaenoic acid synthesis having a nucleotide sequence selected from the group of nucleotide sequences set forth in SEQ ID NOS: 7 and 8.

5 10. A plasmid comprising the isolated DNA of claim 1.

11. A plasmid comprising the isolated DNA of claim 2.

12. A plasmid comprising the isolated DNA of claim 3.

10

13. A plasmid comprising the isolated DNA of claim 4.

14. A plasmid comprising the isolated DNA of claim 5.

15 15. A plasmid comprising the isolated DNA of claim 6.

16. A plasmid comprising the isolated DNA of claim 7

17. A plasmid comprising the isolated DNA of claim 8.

20

18. A plasmid comprising the isolated DNA of claim 9.

19. A bacterium transformed with a plasmid according to claim 10.

25 20. A bacterium transformed with a plasmid according to claim 11.

21. A bacterium transformed with a plasmid according to claim 12.



22. A method of producing eicosapentaenoic acid, comprising the step of culturing a bacterium according to claim 19 for a time and under conditions sufficient for said bacterium to produce eicosapentaenoic acid.

5 23. A method of producing eicosapentaenoic acid, comprising the step of culturing a bacterium according to claim 20 for a time and under conditions sufficient for said bacterium to produce eicosapentaenoic acid.

24. A method of producing eicosapentaenoic acid, comprising the step of culturing a
10 bacterium according to claim 21 for a time and under conditions sufficient for said bacterium to produce eicosapentaenoic acid.

25. An isolated DNA encoding a group of eicosapentaenoic acid synthesis enzymes,
wherein said DNA is obtainable by the steps of:

15

(1) culturing a microorganism belonging to the genus *Shewanella* capable of producing eicosapentaenoic to obtain cultured cells;

(2) extracting chromosomal DNA from the cultured cells;

(3) partially digesting the chromosomal DNA with restriction enzyme Sau3A1 to
20 obtain DNA fragments;

(4) inserting the DNA fragments into cosmid pWE15 completely digested with restriction enzyme BamH1 to obtain recombinant expression vectors;

(5) transforming *Escherichia coli* cells with the recombinant expression vectors to obtain transformants;

25 (6) culturing the transformants to obtain cultured transformed cells;

(7) detecting eicosapentaenoic acid in the cultured transformed cells to select a positive transformed cell capable of producing eicosapentaenoic acid; and



- 90 -

(8) recovering a DNA encoding a group of eicosapentaenoic acid synthesis enzymes by cleaving the recombinant expression vector extracted from the positive transformant with restriction enzyme Sau3A1.

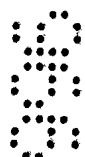
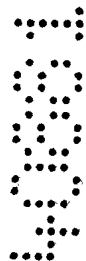
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DATED this FIFTH day of SEPTEMBER, 1996

Sagami Chemical Research Center

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Patent Attorneys for the Applicants



ABSTRACT

5 There is provided an advantageous process for
production of EPA by a gene recombinant technique wherein
genes coding for biosynthesis enzymes for
eicosapentaenoic acid (EPA) useful as pharmaceuticals,
agrochemicals, foods, feeds or the like is obtained from
microorganisms.

10 EPA is produced by obtaining genes coding for
eicosapentaenoic acid (EPA) biosynthesis enzymes,
constructing a plasmid by joining the genes to a vector,
transforming E. coli with the plasmid, and culturing the
transformed E. coli.



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Fig.1

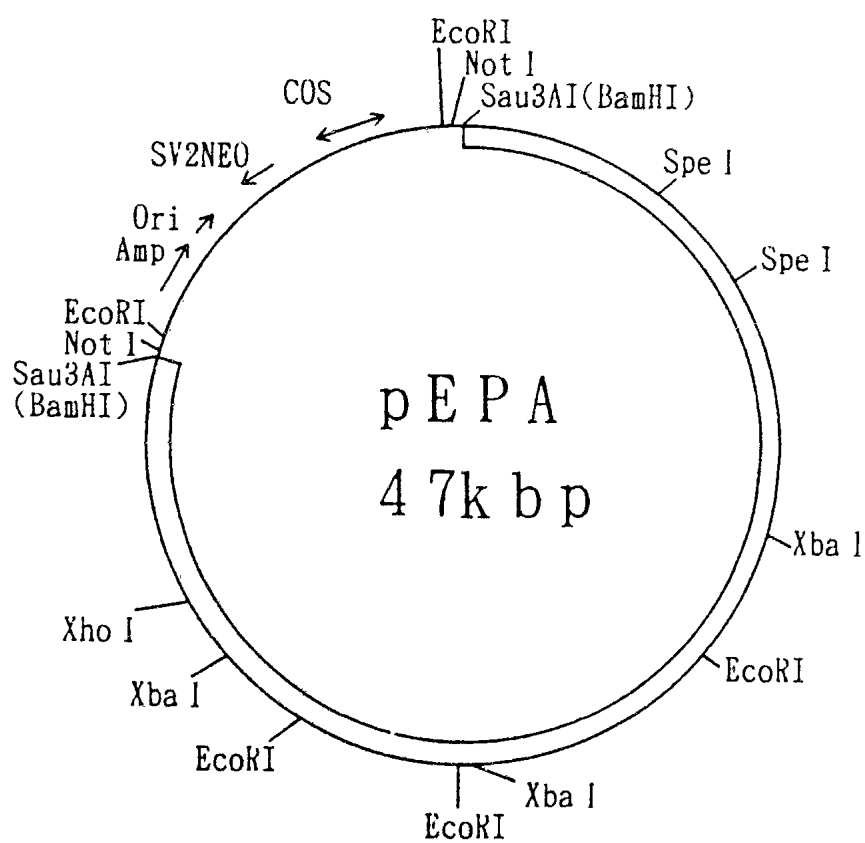
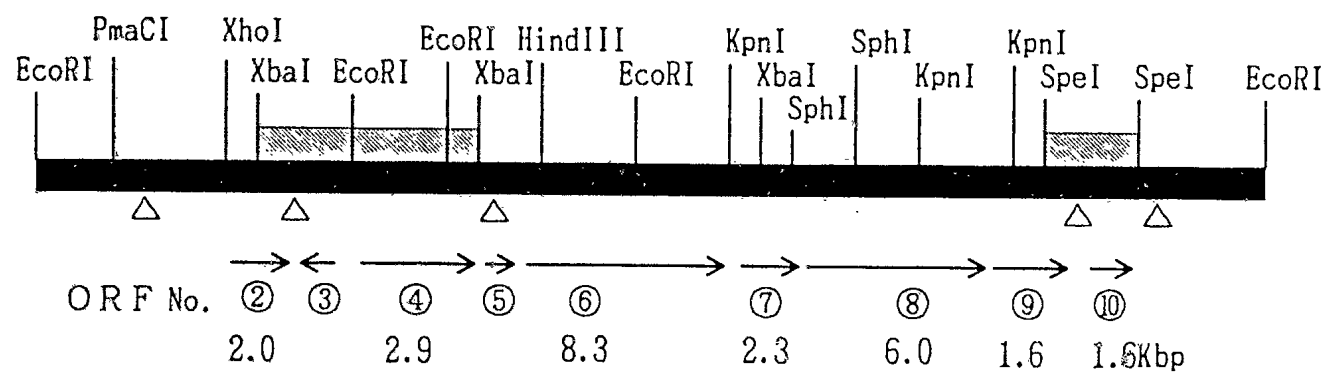


Fig. 2



△ . . . Stem & loop

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INTERNATIONAL SEARCH REPORT

International application No.

PCT/JP93/00641

A. CLASSIFICATION OF SUBJECT MATTER

Int. Cl⁵ C12N15/52

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)

Int. Cl⁵ C12N15/00, C12N9/00

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

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

BIOSIS PRE VIEWS, WPI : EPA, eicosapentaenoicacid

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	JP, A, 2-97393 (Sagami Chemical Research Center), April 9, 1990 (09. 04. 90), Claim (Family: none)	1-17
A	JP, A, 2-23877 (Sagami Chemical Research Center), January 26, 1990 (26. 01. 90), Claim (Family: none)	1-17
A	JP, A, 2-286023 (Sagami Chemical Research Center), November 26, 1990 (26. 11. 90), Claim (Family: none)	1-17

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

☐ See patent family annex.

* 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 document 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

July 27, 1993 (27. 07. 93)

Date of mailing of the international search report

August 17, 1993 (17. 08. 93)

Name and mailing address of the ISA/

Japanese Patent Office

Facsimile No.

Authorized officer

Telephone No.

A. 発明の属する分野の分類 (国際特許分類 (IPC))

Int. Cl.⁸ C12N15/52

B. 調査を行った分野

調査を行った最小限資料 (国際特許分類 (IPC))

Int. Cl.⁸ C12N15/00, C12N9/00

最小限資料以外の資料で調査を行った分野に含まれるもの

国際調査で使用了電子データベース (データベースの名称、調査に使用した用語)

BIOSIS PRE VIEWS, WPI : EPA, eicosapentaenoic acid

C. 関連すると認められる文献

引用文献の カテゴリー*	引用文献名 及び一部の箇所が関連するときは、その関連する箇所の表示	関連する 請求の範囲の番号
A	JP, A, 2-97393 (相模中央化学研究所) 9. 4月. 1990 (09. 04. 90) 特許請求の範囲, (ファミリーなし)	1-17
A	JP, A, 2-23877 (相模中央化学研究所) 26. 1月. 1990 (26. 01. 90) 特許請求の範囲, (ファミリーなし)	1-17
A	JP, A, 2-286023 (相模中央化学研究所) 26. 11月. 1990 (26. 11. 90) 特許請求の範囲, (ファミリーなし)	1-17

☐ C欄の続きにも文献が列举されている。☐ パテントファミリーに関する別紙を参照。

* 引用文献のカテゴリー

「A」特に関連のある文献ではなく、一般的技術水準を示すもの
「E」先行文献ではあるが、国際出願日以後に公表されたもの
「L」優先権主張に疑義を提起する文献又は他の文献の発行日
若しくは他の特別な理由を確立するために引用する文献
(理由を付す)
「O」口頭による開示、使用、展示等に言及する文献
「P」国際出願日前で、かつ優先権の主張の基礎となる出願の日
の後に公表された文献

「T」国際出願日又は優先日後に公表された文献であって出願と
矛盾するものではなく、発明の原理又は理論の理解のため
に引用するもの

「X」特に関連のある文献であって、当該文献のみで発明の新規
性又は進歩性がないと考えられるもの

「Y」特に関連のある文献であって、当該文献と他の1以上の文
献との、当業者にとって自明である組合せによって進歩性
がないと考えられるもの

「&」同一パテントファミリー文献

国際調査を完了した日

27. 07. 93

国際調査報告の発送日

17.08.93

名称及びあて先

日本国特許庁 (ISA/JP)

郵便番号100

東京都千代田区霞が関三丁目4番3号

特許庁審査官 (権限のある職員)

植野 浩志 ㊞

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電話番号 03-3581-1101 内線

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