Psychrilyobacter atlanticus gen. nov., sp. nov., a marine member of the phylum *Fusobacteria* that produces H_2 and degrades nitramine explosives under low temperature conditions

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A Gram-negative and obligately anaerobic marine bacterium, strain HAW-EB21^T, was isolated in a previous study from marine sediment from the Atlantic Ocean, near Halifax Harbor, Canada, and found to have the potential to degrade both hexahydro-1,3,5-trinitro-1,3,5-triazine and octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine. In the present study, phylogenetic analyses showed that strain HAW-EB21^T was only distantly related to the genera *Propionigenium* and *Ilyobacter* with 6.6–7.5 % and 8.2–10.5 % dissimilarity as measured by 16S rRNA and 23S rRNA gene sequence analyses, respectively. Strain HAW-EB21^T displayed unique properties in being psychrotrophic (18.5 °C optimum) and unable to utilize any of the carbon substrates (succinate, L-tartrate, 3-hydroxybutyrate, quinate or shikimate) used for isolating members of the genera *Propionigenium* and *Ilyobacter*. Strain HAW-EB21^T utilized glucose, fructose, maltose, *N*-acetyl-D-glucosamine, citrate, pyruvate, fumarate and Casitone as carbon sources and produced H_2 and acetate as the major fermentation products. Cells grown at 10 °C produced $C_{15:1}$ (30 %), $C_{16:1}\omega7$ (15 %) and $C_{16:0}$ (16 %) as major membrane fatty acids. The novel strain had a genomic DNA G+C content of 28.1 mol%, lower than the values of the genera *Ilyobacter* and *Propionigenium*. Based on the present results, the novel isolate is suggested to be a member of a new genus for which the name *Psychrilyobacter atlanticus* gen. nov., sp. nov. is proposed. The type strain of the type species is HAW-EB21^T (=DSM 19335^T=JCM 14977^T).

Previously we isolated an anaerobic bacterium, strain HAW-EB21^T, from marine sediment sampled from the Emerald Basin (215 m below sea level) near Halifax harbour, Nova Scotia, Canada, for its ability to metabolize both hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX) and octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine (HMX) (Zhao *et al.*, 2004a, b, c). In the present study, we further characterized the biochemical, physiological and genetic properties of the novel strain and found that it represents a new genus and a novel species in the phylum *Fusobacteria*.

As described previously (Zhao *et al.*, 2004b), strain HAW-EB21^T is a Gram-negative, short rod-shaped (0.5 μm in diameter and 0.5–1 μm in length), non-spore-forming, obligately anaerobic bacterium. This bacterium grows well on Brewer anaerobic agar (Becton Dickinson) supplemented with 2 % NaCl or 4 % sea salts (w/v, Sigma) and in marine broth 20 (MB 20) composed of 1.6 % bacto-peptone, 0.4 % yeast extract and 4 % sea salts.

Strain HAW-EB21^T grew in MB 20 medium (initial OD_{600}, 0.09–0.1) at temperatures ranging from 4 to 27.5 °C. The maximal biomass (OD_{600}) obtained after 24–30 h of growth and the growth rate (increase in OD h^{-1} at the exponential growth phase) at different temperatures (°C) (maximal biomass/growth rate/temperature) are as follows: 0.59/0.017/5; 0.81/0.020/10; 0.94/0.076/17; 1.07/0.080/18.5; 0.93/0.080/20.3; 0.880/0.071/22.5; 0.42/0.049/27.5; 0.1/0.0/30. Based on the growth rate (Ratkowsky *et al.*, 1983) and the values for the final biomass, the optimal growth temperature for strain HAW-EB21^T was found to be 18.5 °C. According to the definition proposed by Morita (1975), strain HAW-EB21^T is thus considered to be a psychrotrophic bacterium.

To determine the phylogenetic affiliation of this bacterium, we isolated and purified its genomic DNA (1–5 mg) using

Abbreviations: HMX, octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine; RDX, hexahydro-1,3,5-trinitro-1,3,5-triazine.

The GenBank/EMBL/DDBJ accession numbers for the 16S and 23S rRNA gene sequences of strain HAW-EB21^T are AY579753 and EF426680, respectively.

Marmur's method (Johnson, 1994) and amplified the 16S rRNA gene using protocols as described by Sambrook & Russell (2001). The forward and reverse primers for amplification of the 16S rRNA gene were AAGCCAC-GGCTAACTACG and GTGTGTACAAGNCCCGGGAA, respectively. The 16S rRNA gene product obtained was sequenced (1266 bases), compared with published sequences using BLAST and aligned with closely related species using CLUSTAL_X (1.81). The neighbour-joining method (Saitou & Nei, 1987) included in the MEGA3 package (Kumar *et al.*, 2004) based on the pairwise nucleotide distance of the Kimura two-parameter (complete deletion) model (Kimura, 1980), was used to build a phylogenetic tree (Fig. 1). The number of bootstrap repetitions was 4000.

Phylogenetic analyses of the 16S rRNA gene sequences of strain HAW-EB21^T and related bacteria demonstrated the affiliation of the novel strain to the phylum *Fusobacteria* with 87–93 % gene sequence similarity (Fig. 1). The genera *Ilyobacter* and *Propionigenium* were most closely related to strain HAW-EB21^T (92.5–93.4 % gene sequence similarity, Fig. 1). The 16S rRNA gene sequence of strain HAW-EB21^T was also close (95–99.5 %) to two other marine strains, strain FQ50 from the Arctic (99.4 % similarity, GenBank accession no. AJ877255) and strain Ko711 from Kolbeinsey Ridge, Iceland (99.5 % similarity, GenBank accession no. AF550592), and to six deep-sea environmental sequences (Alain *et al.*, 2002, 2004; Goffredi *et al.*, 2005) obtained from clone libraries (Fig. 1). These sequences formed a highly distinct clade with strain HAW-EB21^T supported by a 100 % bootstrap value. The clade was separated distantly from those of the genera *Ilyobacter*, *Propionigenium* and *Fusobacterium* (Fig. 1). This sequence data suggests that strain HAW-EB21^T represents a new genus of the phylum *Fusobacteria* that appears to have widespread distribution in the marine environment.

The 23S rRNA gene of strain HAW-EB21^T was amplified using the protocol as described by Ludwig *et al.* (1995) using CGTTGAAAAGYBSGGGGATG and CAYGGGGTC-TTTCCGTCCT as the forward and reverse primer, respectively. The 23S rRNA gene product was sequenced (1262 bases) and compared with known sequences in GenBank. Phylogenetic analysis of the 23S rRNA gene sequences of strain HAW-EB21^T and those of related bacteria also showed that strain HAW-EB21^T had a high dissimilarity to those of members of the genera *Propionigenium* and *Ilyobacter* (9.1–11.5 %) and from those of the genus *Fusobacterium* (14–16 %) (Fig. 2). These results are consistent with the finding based on the 16S

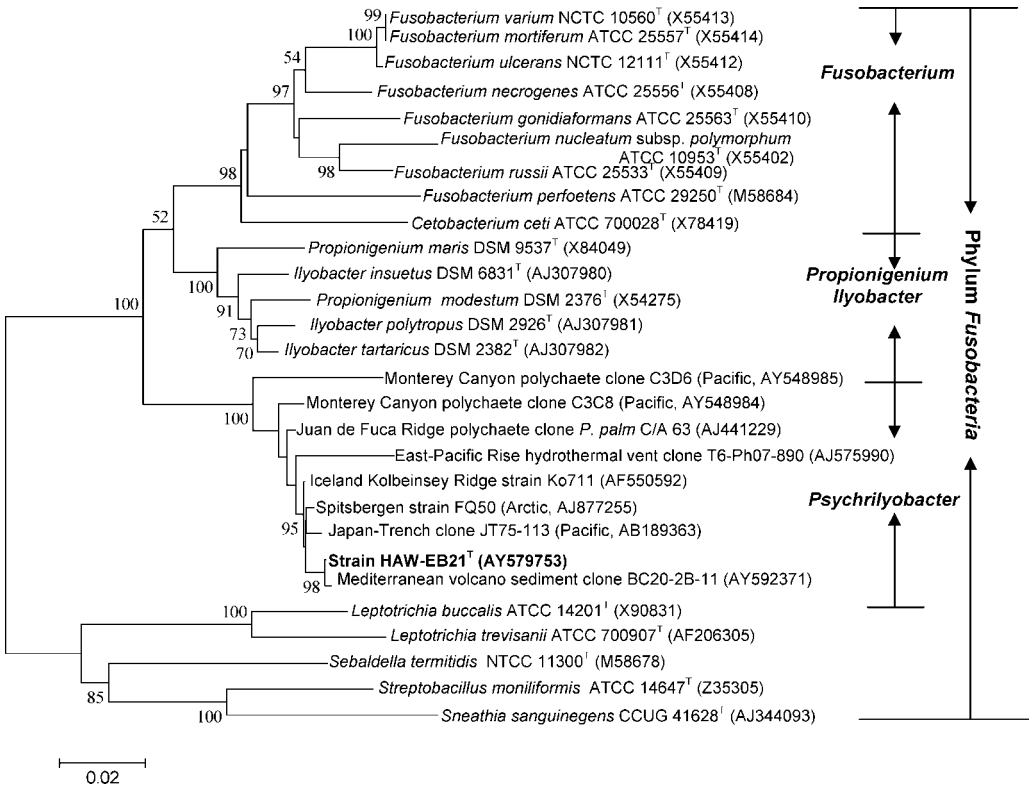

Fig. 1. The phylogenetic position of strain HAW-EB21^T within the phylum *Fusobacteria* based on 16S rRNA gene sequence analysis. The phylogenetic tree was generated based on pairwise nucleotide distance of Kimura two-parameter using the neighbour-joining method (complete deletion) included in the MEGA3 software package (Kumar *et al.*, 2004). Bar, 2 substitutions per 100 nucleotides.

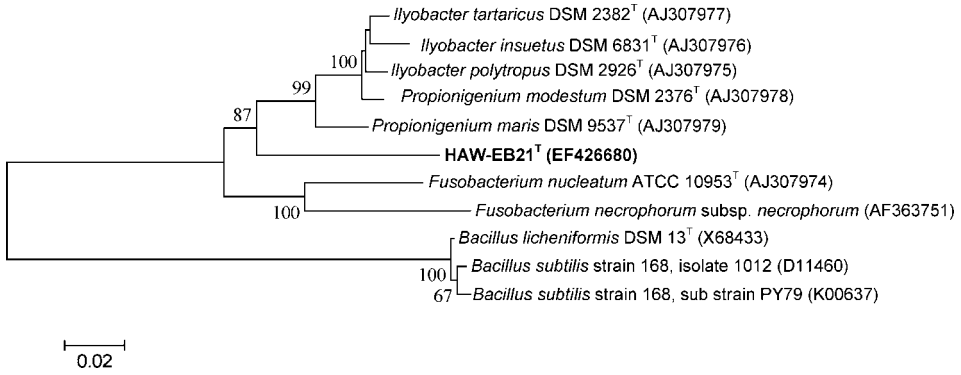

Fig. 2. The phylogenetic position of strain HAW-EB21^T within the phylum *Fusobacteria* based on 23S rRNA gene sequence analysis. The phylogenetic tree was generated by the same method as in Fig. 1. Bar, 2 substitutions per 100 nucleotides.

rRNA gene sequence and further demonstrate that strain HAW-EB21^T is not a member of any recognized genus belonging to the phylum *Fusobacteria*.

The genomic DNA G+C content of strain HAW-EB21^T was determined from its thermal denaturation profile obtained as described by Sly *et al.* (1986). The genomic DNA of *Escherichia coli* ATCC 11775^T, with a known DNA G+C content of 51.7 mol%, was used as an internal reference standard. Strain HAW-EB21^T was found to have a DNA G+C content of 28.1 mol%, lower than values for members of the genera *Ilyobacter* (32.2–35.7 mol%) and *Propionigenium* (33.9–40 mol%) (Table 1).

To compare the phenotypic properties of strain HAW-EB21^T with those of related bacteria, we tested the ability of the novel strain to utilize some sugars, organic acids and amino acids (0.1 %, w/v) in basic sea salts medium containing NH_4Cl (0.04 % w/v, as nitrogen source) and yeast extract (0.01 % w/v, pH 7.0) under anaerobic conditions. The liquid medium (4.5 ml) in sealed serum bottles (20 ml) was made anaerobic by repeated degassing under a vacuum and recharging with argon. Fresh cultures (0.5 ml) of the novel strain (pre-grown in anaerobic MB 20 at 10 °C for 4 days) were inoculated to liquid medium described above at an initial OD_{600} of 0.1. Enhanced growth after 3 days incubation in the presence of carbon sources was used as an indicator for bacterial utilization of the carbon source tested. Unless otherwise noted, the concentration of the substrates remaining and the acidic products that had accumulated in the medium were

Table 1. Properties of strain HAW-EB21^T that distinguish it from the genera *Propionigenium*, *Ilyobacter* and *Fusobacterium* of the phylum *Fusobacteria*

Taxa: 1, strain HAW-EB21^T; 2, *Propionigenium* [data for *Propionigenium maris* (Janssen & Liesack, 1995; Watson *et al.*, 2000) and *Propionigenium modestum* (Schink & Pfennig, 1982)]; 3, *Ilyobacter* [*I. polytropus* (Stieb & Schink, 1984), *I. tartaricus* (Schink, 1984) and *I. insuetus* (Brune & Schink, 1992)]; 4, *Fusobacterium* (Robrish *et al.*, 1991; Hofstad, 1991; Moore *et al.*, 1984). +, Positive; −, negative; V, variable among species; NK, not known.

Characteristic	1	2	3	4
Habitat	Cold deep marine sediment	Anaerobic mud, human saliva	Anoxic sediment and sludge	Human mouth, gastrointestinal tract, animal cavity
Morphology	Short rod	Short rod or cocci	Round-ended rod or cocci	Spindle-shaped or pleomorphic rod
Growth at:				
4 °C	+	−	−	−
37 °C	−	+	NK	+
Temp. optima (°C)	18.5	33–37	28–34	37
NaCl requirement	+	+	+	−
Utilization of:				
Succinate	−	+	NK	NK
3-Hydroxybutyrate, tartrate or quinate (shikimate)	−	NK	V	NK
DNA G+C content (mol%)	28.1	33.9–40	32.2–35.7	26–34

measured using HPLC (Waters) equipped with a model 717+ injector, a model 600 pump, a model 2996 photodiode array (PDA) detector and a 2414 refractive index detector. The separation unit consisted of a ICsep ICE-ION-300 column (300 mm × 7.8 mm; Transgenomics) and an ion guard GC-801 column (Transgenomics). The mobile phase was 0.0175 M sulfuric acid with a pH of 4, flowing at a rate of 0.4 ml min^{-1}. Measurements were performed at 210 nm. Volatile fatty acids (acetate, propionate and butyrate) produced during fermentative growth were determined using a GC system (Agilent 6890) equipped with a flame ionization detector (FID) and a glass column (1 m × 2 mm Carbopack C, 60–80 mesh) coated with 0.3 % Carbowax 20 M and 0.1 % H_3PO_4. Ethanol was measured using a previously described protocol for methanol (Zhao *et al.*, 2002). All tests were performed in triplicate. Hydrogen, sampled using a gas tight syringe (500 μl) from the headspace of the culture vials, was measured by GC (Agilent 6890) connected to a Chromosorb 102 column (11 m × 2 mm; Supelco) and a thermal conductivity detector (TCD, 150 °C) with argon as the carrier gas. The initial column temperature was maintained at 35 °C for 7.5 min, increased to 100 °C at a rate of 75 °C min^{-1} and then maintained at 100 °C for 6 min. Spore formation and the activities of catalase and oxidase were tested using protocols described by Smibert & Krieg (1994). NaCl tolerance was tested using Brewer anaerobic agar (containing 0.5 % NaCl) supplemented with additional amounts of NaCl (Bowman, 2001).

Strain HAW-EB21^T displayed significant differences in growth conditions from members of the genera *Ilyobacter*, *Propionigenium* and *Fusobacterium*. When temperatures for growth were determined (Tables 1 and 2), strain HAW-EB21^T was found to be a psychrotrophic bacterium with a growth temperature optimum of 18.5 °C (Zhao *et al.*, 2004b), whereas members of the genera *Ilyobacter*, *Propionigenium* and *Fusobacterium* are all mesophilic bacteria with temperature optima of 28–37 °C (Moore *et al.*, 1984; Schink & Pfennig, 1982; Stieb & Schink, 1984; Schink, 1984; Janssen & Liesack, 1995; Brune *et al.*, 2002). As regards requirement for NaCl, strain HAW-EB21^T was moderately halophilic, showing no visible growth on Brewer anaerobic agar (Becton Dickinson) that contained only 0.5 % NaCl, but good growth if 2 % NaCl or 4 % sea salts was added. In contrast, members of the genus *Fusobacterium* did not require NaCl for growth (Table 1).

Strain HAW-EB21^T was also different from species of the genera *Propionigenium* and *Ilyobacter* in the utilization of carbon sources (Table 2). For example, strain HAW-EB21^T did not grow on succinate, the carbon source for the isolation of members of the genus *Propionigenium* (Schink & Pfennig, 1982; Janssen & Liesack, 1995; Schink, 1992), or on L-tartrate, 3-hydroxybutyrate or quinate (and shikimate) which have been used for the isolation of *Ilyobacter tartaricus* (Schink, 1984), *Ilyobacter polytropus* (Stieb & Schink, 1984) and *Ilyobacter insuetus* (Brune & Schink, 1992; Brune *et al.*, 2002), respectively.

Table 2. Properties of strain HAW-EB21^T different from the most closely related species of the genera *Ilyobacter* and *Propionigenium*

Taxa: 1, strain HAW-EB21^T; 2, *I. tartaricus* (data from Schink, 1984); 3, *I. insuetus* (Brune & Schink, 1992); 4, *I. polytropus* (Stieb & Schink, 1984); 5, *Propionigenium modestum* (Schink & Pfennig, 1982); 6, *Propionigenium maris* (Janssen & Liesack, 1995; Watson *et al.*, 2000). +, Positive; −, negative; Ac, acetate; Et, ethanol; Fo, formate; Pr, propionate; Lac, lactate; Bu, butyrate; NA, not applicable; ND, no data; v, variable among strains.

Characteristic	1	2	3	4	5	6
Cell shape	Short rod	Short rod	Coccoid	Short rod	Short rod	Round-ended rod
Size (μm, diameter × length)	0.5 × (0.5–1)	1 × (1.2–2.5)	0.8–1.5	0.7 × (1.5–3)	0.5 × (0.5–2)	1.0 × (1.2–2.5)
Optimum temp. (°C)	18.5	28	30	28–34	33	34–37
Utilization of:						
Glucose, fructose, citrate	+	+	−	+	−	v
Maltose	+	−	ND	−	−	v
Glutamate	+	ND	ND	ND	−	v
Quinate, shikimate	−	ND	+	ND	ND	ND
Tartrate	−	+	−	−	−	−
Succinate	−	ND	ND	−	+	+
3-Hydroxybutyrate	−	ND	−	−	ND	+
Fumarate	+	−	−	+	+	+
Malate	−	−	−	+	+	−
Acid produced from fermentation of:						
Glucose, fructose	Ac, Et, Bu, Fo	Fo, Ac, Et	NA	Fo, Ac, Et	NA	Lac, Fo, Ac, Et
Pyruvate	Ac, Bu, Et	Ac, Fo	NA	Fo, Ac	Ac, Pr	Pro, Bu
Fumarate	Ac, Bu, Fo	NA	NA	Fo, Ac, Pr	Pro, Ac	v
Quinate	NA	NA	Ac, Bu	NA	NA	NA

Like other members of the phylum *Fusobacteria*, the novel strain was able to grow by fermenting carbon substrates (Table 3). Strain HAW-EB21^T utilized glucose, fructose and maltose which are not utilized by *I. insuetus* (Brune & Schink, 1992) and most species of the genus *Fusobacterium* (Robrish *et al.*, 1991; Hofstad, 1991; Moore *et al.*, 1984). Most species of the genus *Fusobacterium* only use peptone and amino acids as carbon sources (Hofstad, 1991; Moore *et al.*, 1984).

Table 3 summarizes the fermentation products of various substrates utilized by strain HAW-EB21^T as detected in the present study. The fermentation pathways derived from the data in Table 3 are shown in Fig. 3. The results clearly show that strain HAW-EB21^T fermented all tested substrates to acetate, with butyrate and ethanol as minor products (Table 3, Fig. 3). Butyrate was a significant fermentation product from fumarate, aspartate, lysine and Casitone. Of all substrates tested, only threonine was fermented to propionate. Some species of the genus *Fusobacterium* such as *Fusobacterium necrophorum, Fusobacterium gonidiaformans*, *Fusobacterium mortiferum* and *Fusobacterium varium* (Hofstad, 1991; Moore *et al.*, 1984) are also known for their ability to ferment threonine to propionate. For strain HAW-EB21^T, formate was a significant product in the fermentation of fumarate and citrate, but was only a minor product in the fermentation of glucose and *N*-acetyl-D-glucosamine. Unlike *Propionigenium modestum* (Schink & Pfennig, 1982), strain HAW-EB21^T did not ferment pyruvate to propionate. The novel strain did not ferment fructose and pyruvate to formate, in contrast to *I. tartaricus* (Schink, 1984) and *I. polytropus* (Stieb & Schink, 1984).

Strain HAW-EB21^T also fermented carbohydrates (glucose, fructose and maltose) and acids to produce H_2 (Table 3). The H_2/acetate molar ratio (1.5–2.2) was high in the fermentation of glucose, fructose or maltose, but was very low (0.1–0.38) for the fermentation of C_3–C_5 acids (pyruvate, fumarate and citrate) and amino acids (glutamate). Little H_2 was produced in the fermentation of lysine or aspartate. Members of the phylum *Fusobacteria* such as *I. insuetus* (Brune & Schink, 1992), *Propionigenium maris* (Janssen & Liesack, 1995; Watson *et al.*, 2000) and the species of the genus *Fusobacterium* (Hofstad, 1991; Moore *et al.*, 1984) are also known to produce H_2. However, none of these bacteria have been reported to produce H_2 at low temperatures (4–10 °C). Strain HAW-EB21^T may thus be suitable for further investigation of optimized production of biohydrogen under low temperature conditions.

Strain HAW-EB21^T was also able to degrade RDX and HMX (Zhao *et al.*, 2004b, c) as reported previously. So far, no other strains of the phylum *Fusobacteria* are known to metabolize RDX or HMX.

The fatty acid composition of membrane lipids of the novel strain was determined using a protocol similar to that described by Bowman (2001) and Fay & Richli (1991) with experimental details as described previously by Zhao *et al.* (2005). As previously found for species of the genus *Fusobacterium*, strain HAW-EB21^T contained fatty acids $C_{16:0}$ (16 %) and $C_{16:1}\omega7$ (15 %). The major membrane acid in the novel strain was $C_{15:1}$ (30 %). However, this fatty acid is absent from the cell membranes of members of the genus *Fusobacterium*, which instead possess $C_{14:0}$ as the major membrane acid (Calhoon *et al.*, 1983; Tuner *et al.*, 1992). The major fatty acids of cells of strain HAW-EB21^T grown at 10°C in MB 2216 were (%): $C_{11:0}$ (1), $C_{12:0}$ (1), $C_{13:0}$ (2), $C_{14:0}$ (6), $C_{14:1}$ (4), $C_{15:0}$ (9), $C_{15:1}$ (30), $C_{16:0}$ (16), $C_{16:1}\omega7$ (15), $C_{17:0}$ (8), $C_{17:1}$ (5), $C_{18:0}$ (1) and $C_{18:1}\omega7$ (1).

Table 3. Growth of strain HAW-EB21^T on substrates (3 days) and fermentation products

NAG, *N*-acetyl-D-glucosamine; Ac, Acetate; Bu, butyrate; Et, ethanol; Pr, propionate; Fo, formate; −, no data. The subtotal column is calculated as the sum of Ac + Bu + Pr + Fo + Et.

Substrate	Amount (μmol)		Biomass increase (OD_{600})	Fermentation products (μmol)						
	Initial	Removal		Ac	Bu	Pr	Fo	Et	Subtotal	H_2
Glucose	113	40	0.34	9.1	1.1	0.0	3.0	5.1	18.3	20.0
Maltose	55	12	0.44	15.9	3.4	0.0	0.0	5.9	25.2	25.2
Fructose	110	70	0.21	17.1	4.3	0.0	0.0	4.0	25.4	24.5
NAG	90	14	0.31	31.8	2.3	0.0	5.4	7.7	47.2	27.5
Fumarate	138	136	0.08	94.7	19.6	0.2	26.0	0.4	140.9	35.0
Citrate	70	70	0.22	131.4	0.5	0.0	35.2	0.6	167.7	16.7
Pyruvate	183	183	0.19	130.2	5.5	0.0	0.0	2.2	137.9	49.5
Aspartate	131	−	0	86.3	16.9	0.3	0.0	0.2	103.7	0.0
Threonine	168	−	0.07	0.0	1.3	144.2	0.0	0.3	145.8	17.8
Glutamate	128	−	0.07	172.1	4.4	0.3	0.0	2.2	179.0	18.2
Lysine	109	−	0.07	21.7	28.9	0.1	0.0	0.2	50.9	0.0
Casitone	1*	−	0.27	14.5	13.6	3.3	0.0	1.1	32.5	13.0

*Amount shown in g l^{-1}.

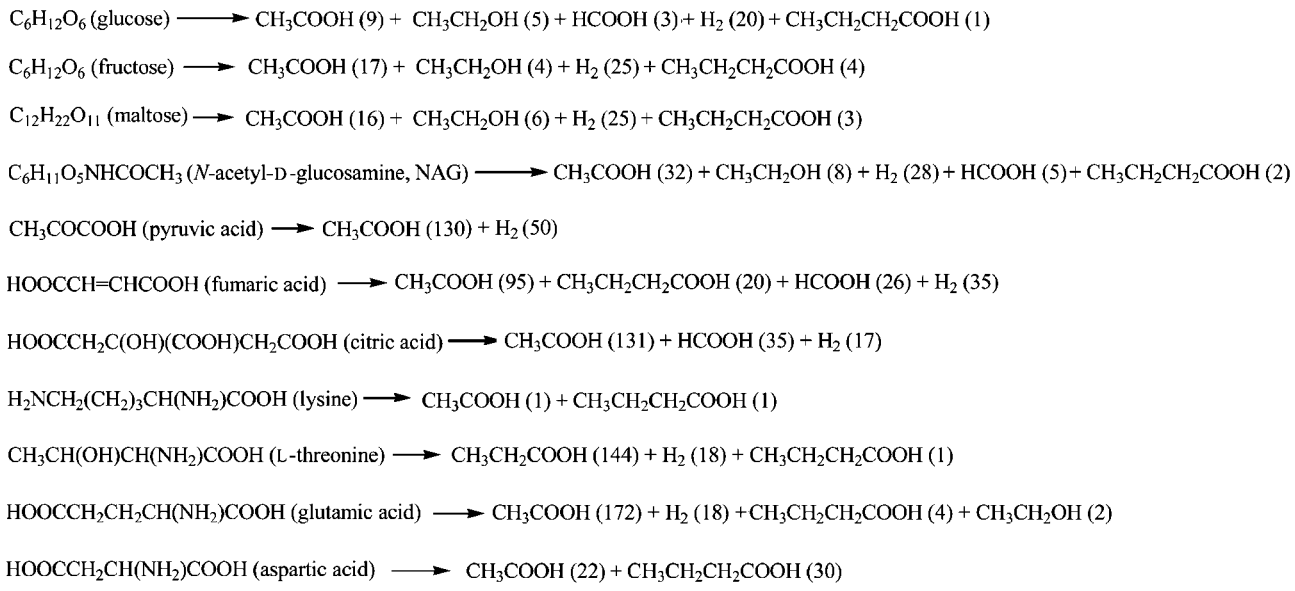

Fig. 3. Fermentation pathways of sugar or acids as derived from data in Table 3. Values in parentheses are the approximate amount (mol) of product relative to those of other products.

The phenotypic properties of the two deep-sea strains, Ko711 and FQ50, which were shown to be closely related to strain HAW-EB21^T by their 16S rRNA gene sequences, have not been determined. Based on the description in GenBank (http://www.ncbi.nlm.nih.gov/) and the fact that they were also isolated from the deep and cold Kolbeinsey Ridge, Iceland, and Spitsbergen in the Arctic Ocean, they are assumed to be similar to strain HAW-EB21^T in their capacity to grow at low temperatures and in marine media.

Based on the above genotypic and phenotypic characteristics of strain HAW-EB21^T, the new isolate is suggested to represent a new genus and novel species, for which the name *Psychrilyobacter atlanticus* gen. nov., sp. nov. is proposed.

Description of *Psychrilyobacter* gen. nov.

Psychrilyobacter (psy.chri.lyo′bac.ter. Gr. adj. *psychros* cold; N.L. masc. n. *Ilyobacter* a bacterial genus name; N.L. masc. n. *Psychrilyobacter* a psychrotrophic bacterium related to the genus *Ilyobacter*).

Obligately anaerobic. Short rod-shaped, Gram-negative bacteria. No spore formation. Grow at low temperatures. Require NaCl for growth and are slightly halophilic. Positive in tests for the utilization of sugars, organic and amino acids and peptone. Fermentative metabolism. H_2 and acetate are major fermentation products. No utilization of succinate, L-tartrate, 3-hydroxybutyrate or quinate (and shikimate). The type species is *Psychrilyobacter atlanticus*.

Description of *Psychrilyobacter atlanticus* sp. nov.

Psychrilyobacter atlanticus (at.lan′ti.cus. L. masc. adj. *atlanticus* of or pertaining to the Atlantic Ocean).

Displays the following characteristics in addition to those given in the genus description. Cells are 0.5 μm in diameter and 0.5–1 μm in length. Colonies are round and colourless. Non-motile. Catalase- and oxidase-negative. Grows at temperatures of between 4 and 27.5 °C; optimum at 18.5 °C. Slightly halophilic (2 % NaCl). No growth occurs at NaCl concentrations of <0.5 % (w/v). Chemo-organotrophic and fermentative growth. Positive in tests for the utilization of glucose, fructose, maltose, *N*-acetyl-D-glucosamine, citrate, pyruvate, Casitone, fumarate, glutamate, lysine, threonine and aspartate. Does not utilize tartrate, glycerol, 3-hydroxybutyrate, quinate, shikimate or malate. Does not degrade chitin, starch or cellulose. H_2 and acetate are major fermentation products; ethanol and butyrate are minor products. Ferments threonine to propionate and H_2. Ferments lysine and aspartate to acetate and butyrate. No H_2 production from lysine or aspartate. RDX and HMX are degraded to nitroso derivatives, methylenedinitramine (MEDINA), formaldehyde (HCHO) and N_2O. The major fatty acids are $C_{15:1}$, $C_{16:0}$, $C_{16:1}\omega7$, $C_{15:0}$, $C_{17:0}$ and $C_{14:0}$.

The type strain, HAW-EB21^T (=DSM 19335^T=JCM 14977^T), was isolated from a site 215 m deep and 50 nautical miles from Halifax harbour, in the Atlantic Ocean. The genomic DNA G+C content of the type strain is 28.1 mol%.

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