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Vibrio coralliilyticus sp. nov., a temperature-dependent pathogen of the coral *Pocillopora damicornis*

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Vibrio sp. YB1^T (=ATCC BAA-450^T =LMG 20984^T), the aetiological agent of tissue lysis of the coral *Pocillopora damicornis*, was characterized as a novel *Vibrio* species on the basis of 16S rDNA sequence, DNA–DNA hybridization data (G+C content is 45·6 mol%), AFLP and GTG_5-PCR genomic fingerprinting patterns and phenotypic properties, including the cellular fatty acid profile. The predominant fatty acids were 16 : 0 and 18 : 1ω7*c*. The name *Vibrio coralliilyticus* sp. nov. is proposed for the novel coral-pathogenic species. In addition to strain YB1^T, which was isolated from the Indian Ocean, five additional strains of *V. coralliilyticus* have been isolated, three from diseased *P. damicornis* in the Red Sea, one from diseased oyster larvae (Kent, UK) and one from bivalve larvae (Brazil). The six *V. coralliilyticus* strains showed high genotypic and phenotypic similarities and all were pathogenic to *P. damicornis*. The closest phylogenetic neighbours to *V. coralliilyticus* are *Vibrio tubiashii*, *Vibrio nereis* and *Vibrio shilonii*.

INTRODUCTION

During the last two decades, there has been a large increase in the frequency and distribution of coral diseases (Hoegh-Guldberg, 1999; Peters, 1997; Richardson, 1998). These diseases have altered both total abundance and species diversity (Loya *et al.*, 2001). The causative agents of only a few of these diseases have been reported: bleaching of *Oculina patagonica* by *Vibrio shilonii* (Kushmaro *et al.*, 1996, 1997), black band disease by a microbial consortium (Carlton & Richardson, 1995), sea fan disease by *Aspergillus sydowii* (Smith *et al.*, 1996; Geiser *et al.*, 1998) and coral white plague possibly by a *Sphingomonas* sp. (Richardson *et al.*, 1998). The aetiological agents of most coral diseases are presently unknown. What is known is that most, if not all, of the diseases occur at higher-than-normal sea-water temperatures (Rosenberg & Ben-Haim, 2002; Hoegh-Guldberg, 1999). Since temperatures are expected to rise considerably during this century, it is likely that coral disease will become even more prevalent. Thus, there is an increasing need to identify and characterize coral pathogens.

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The GenBank/EMBL/DDBJ accession numbers for the 16S rDNA sequences of strains LMG 10953, LMG 21349 and YB1^T are AJ316167, AJ440004 and AJ440005.

Recently, we isolated a novel temperature-dependent pathogen of the coral *Pocillopora damicornis*, tentatively named *Vibrio* sp. YB1. At water temperatures above 26 °C, a pure culture of this pathogenic strain caused rapid destruction of the coral tissue within 2 weeks (Ben-Haim & Rosenberg, 2002). The present study was carried out in order to characterize *Vibrio* sp. YB1 and related strains using a polyphasic approach.

METHODS

Micro-organisms, media and growth conditions. Strain YB1^T (=ATCC BAA-450^T =LMG 20984^T) was isolated from a diseased coral of the species *P. damicornis* in the Indian Ocean near Zanzibar, as described previously (Ben-Haim & Rosenberg, 2002). Strains LMG 21348, LMG 21349 and LMG 21350 were isolated from three different diseased *P. damicornis* colonies on the Eilat coral reef, Red Sea. The bacteria were isolated from the crushed tissues of the corals, as described previously (Ben-Haim & Rosenberg, 2002). *V. shilonii* ATCC BAA-91^T (=LMG 19703^T =DSM 13774^T) was isolated from a bleached coral of the species *O. patagonica* (Kushmaro *et al.*, 1996, 2001). The other *Vibrio* strains used in this study were obtained from the BCCM/LMG Bacteria Collection (Table 1). Strains were grown routinely at 30 °C in MBT medium [1·8 % Marine broth (MB) 2216 (Difco), 0·9 % NaCl, 0·5 % tryptone (Difco)], on MB 2216 agar (1·8 % MB 2216, 0·9 % NaCl, 1·8 % agar), tryptone soy agar (TSA; Oxoid) supplemented with 2 % NaCl or on TCBS (thiosulphate-citrate-bile salts-sucrose; Difco) agar adjusted to 3 % NaCl. Liquid cultures were prepared in 125 ml

Table 1. Strains used in this study

Strain(s)	Source and date of isolation
V. coralliilyticus sp. nov.	
YB1^T (=ATCC BAA-450^T =LMG 20984^T)	Diseased coral (*P. damicornis*), Zanzibar, Tanzania, 1999
LMG 21348 (=YB2), LMG 21349 (=YB3), LMG 21350 (=YB4)	Diseased coral (*P. damicornis*), Eilat, Israel, 2001
LMG 10953 (=NCIMB 2165)	Diseased oyster (*C. gigas*) larvae, Kent, UK, 1980s
LMG 20538 (=INCO 83)	Bivalve (*N. nodosus*) larvae, LCMM Florianópolis, Brazil, 1998
V. nereis LMG 3895^T (=ATCC 25917^T)	Sea water, Hawaii, USA
V. tubiashii LMG 10936^T (=ATCC 19109^T)	Hard clam, Milford, CT, USA
V. mediterranei LMG 11258^T (=CIP 10320^T)	Coastal sea water, Valencia, Spain
V. shilonii LMG 19703^T (=ATCC BAA-91^T =DSM 13774^T)	Bleached coral, Tel Aviv, Israel, 1995

flasks containing 10 ml MBT, incubated with shaking (160 r.p.m.) for 24–48 h. Cultures were stored either at −70 °C in 15 % glycerol or as lyophilized cells.

DNA isolation and genomic fingerprinting (FAFLP, BOX-PCR, GTG_5-PCR). Bacterial DNA was extracted following the technique of Pitcher *et al.* (1989). Fluorescent amplified fragment length polymorphism (FAFLP) patterns were generated and analysed as described previously (Thompson *et al.*, 2001). Briefly, 1 μg high-molecular-mass DNA was digested with *Taq*I and *Hin*dIII, followed by ligation of restriction-half-site-specific adaptors to all restricted fragments. Subsequently, two PCR amplifications were applied using primers H00/T00 and H01-6FAM/T03. Separation of the PCR products was done on 36 cm denaturing polyacrylamide gels on an ABI Prism 377 DNA sequencer (Applied Biosystems). Tracking and normalization of the lanes were performed by the GeneScan 3.1 software (Applied Biosystems). Normalized tables of peaks, containing fragments of 50–536 bp, were analysed with BioNumerics 2.0 software (Applied Maths). Similarity among band patterns was calculated using the Dice similarity coefficient and dendrograms were built using the Ward algorithm (Sneath & Sokal, 1973).

Rep-PCR fingerprinting using GTG_5 and BOX primers was performed as described previously (Rademaker *et al.*, 1998). Briefly, PCRs consisted of 1 μl template DNA (50 ng μl^{-1}), 5 μl 5 × Gitschier buffer, 0·4 μl BSA (10 mg ml^{-1}), 2·5 μl DMSO (10 mg ml^{-1}), 1·25 μl of a dNTP mixture (100 mM of each), 1 μl GTG_5 primer or 1 μl BOX primer (both 0·3 μg μl^{-1}; Amersham Pharmacia Biotech) and 0·4 μl *Taq* DNA polymerase (5 U μl^{-1}; Goldstar Red). The PCR products generated with the GTG_5 primer were electrophoresed in a 1·5 % agarose gel (w/v) and 1 × TAE buffer at a constant 55 V for 900 min at 4 °C. The PCR products generated with the BOX primer were electrophoresed in a 2 % agarose gel (w/v) and 1 × TBE buffer at a constant 130 V for 222 min at 4 °C. After staining with ethidium bromide, the digitized patterns were normalized and analysed numerically using the software BioNumerics 2.0. Similarity among patterns was calculated based on Pearson's similarity coefficient and dendrograms were built using UPGMA (Sneath & Sokal, 1973).

16S rDNA sequencing. Almost-complete 16S rDNA sequences were obtained following the methodology described previously (Thompson *et al.*, 2001). Briefly, fragments of the 16S rDNA were amplified by PCR using the conserved primers pA (16F27) and pH (16R1522) or MH1 (16F27) and MH2 (16R1485). Subsequently, purified products were used as templates for sequencing amplification using the ABI Prism BigDye Terminator Ready Reaction mix and eight primers (16F358, 16F536, 16F926, 16F1112, 16F1241, 16R339, 16R519 and 16R1093). Purified sequencing products were run on 48 cm denaturing polyacrylamide gels on an ABI Prism 377 DNA sequencer. Sequences were assembled with the AutoAssembler software (Applied Biosystems). The consensus sequences were transferred into BioNumerics 2.0 software, where a phylogenetic tree was constructed based on the neighbour-joining method (Saitou & Nei, 1987). The phylogenetic positions of the consensus sequences were obtained using the program FASTA (Pearson & Lipman, 1988) and compared to known *Vibrio* 16S rDNA sequences (Dorsch *et al.*, 1992; Ruimy *et al.*, 1994; Mellado *et al.*, 1996; Denner *et al.*, 2002).

DNA–DNA hybridization experiments and G+C content determination. DNA–DNA hybridization was performed under stringent conditions using the microplate technique (Ezaki *et al.*, 1989) with photobiotin-labelled DNA at 39 °C for 3 h as described previously (Willems *et al.*, 2001). Hybridization values are means of the reciprocal and non-reciprocal values, each of which was performed in four replicates. The G+C content of DNA was determined by HPLC (Tamaoka & Komagata, 1984).

Phenotypic characterization. Colony morphology was examined using a stereoscopic microscope. Cell morphology was examined by scanning electron microscopy (JEOL 840A). Exponentially growing bacteria in MBT medium were adhered to a carbon-coated grid and negatively stained with 1 % uranyl acetate. Classical phenotypic tests were performed by standard methods (Farmer & Hickman-Brenner, 1992). Biochemical tests were performed using the API 20NE system (micromethod tests for the identification of non-enteric Gram-negative rods; bioMérieux). The standard API 20NE protocol was used except that media were adjusted to 3 % NaCl. NaCl tolerance was determined in MBT medium containing varying concentrations (1–15 %) of NaCl. Sensitivity to the vibriostatic compound O/129 (2,4-diamino-6,7-diisopropylpteridine; Sigma) was determined after incubation for 48 h at 30 °C on MB agar containing 30 μg of the compound on a disc.

Sensitivity to antibiotics was examined using either the minimal inhibition concentration (MIC) method, for erythromycin (24 μg ml^{-1}), tetracycline (20 μg ml^{-1}), chloramphenicol (6 μg ml^{-1}) and kanamycin (50 μg ml^{-1}), or by the paper disc method, for penicillin (10 μg disc^{-1}) and ampicillin (10 μg disc^{-1}). Growth was also tested on MB agar containing 200 μg gentamicin ml^{-1}.

Carbon utilization tests were carried out using Biolog GN2 MicroPlates. Pure cultures (12–18 h growth) were harvested from MB agar plates and suspended in 20 ml GN/GP inoculating fluid (Biolog), adjusted to 3 % NaCl, to an OD_{600} of 0·130–0·143. The suspension was then distributed into Biolog GN2 microwell plates. Each plate contained 96 microwells with one of 95 different carbon sources in each and tetrazolium violet as an indicator of metabolic activity. The plates were incubated for 48 h at 30 °C. Wells that changed to purple were marked as positive for metabolic utilization. Fatty acid analysis was performed following the protocol of the

Microbial Identification System (Microbial ID Inc.) as described previously (Huys *et al.*, 1994). Strains for analysis were grown on TSA for 48 h at 28 °C.

The ability of each strain to infect the coral *P. damicornis* in controlled aquarium experiments was examined as described previously (Ben-Haim & Rosenberg, 2002).

RESULTS

Strain YB1^T is a Gram-negative, motile, rod-shaped bacterium (1·2–1·5 × 0·8 μm) that has a single polar, sheathed flagellum (Fig. 1). These properties, together with its ability to form yellow colonies on TCBS agar and its sensitivity to the vibriostatic compound O/129, suggest that YB1^T represents a species of the genus *Vibrio* (Farmer & Hickman-Brenner, 1992). The 16S rDNA sequences of strains YB1^T (1465 bp; accession no. AJ440005), LMG 21349 (1468 bp; AJ440004) and LMG 10953 (1468 bp; AJ316167) were allocated to the *γ-Proteobacteria* using the FASTA program. These *Vibrio* strains formed a tight cluster, with more than 99 % 16S rDNA similarity (Fig. 2). Their closest phylogenetic neighbours were *Vibrio tubiashii* (97·2 %), *Vibrio nereis* (96·8 %) and *V. shilonii* (96·6 %).

AFLP, BOX-PCR and GTG_5-PCR analysis revealed that the strains possess typical genomes, respectively consisting of 102 ± 7, 19 ± 3 and 23 ± 2 bands (Fig. 3). The inner AFLP and GTG_5-PCR pattern similarities were higher than 64 %. It has recently been shown that AFLP and rep-PCR similarities around 65 % represent more than 70 % DNA–DNA similarity (Rademaker *et al.*, 2000). It was clearly demonstrated by the three fingerprinting methodologies that the six *Vibrio* isolates form a tight genomic group that is distinguishable from all other closely related *Vibrio* species. Surprisingly, strains YB1^T and LMG 21350 showed very similar patterns by AFLP and GTG_5-PCR, although YB1^T originated from the Indian Ocean and LMG 21350 from the Red Sea. Overall, the strains have less than 40 % pattern similarity towards their closest phylogenetic neighbours analysed by the three fingerprinting methodologies.

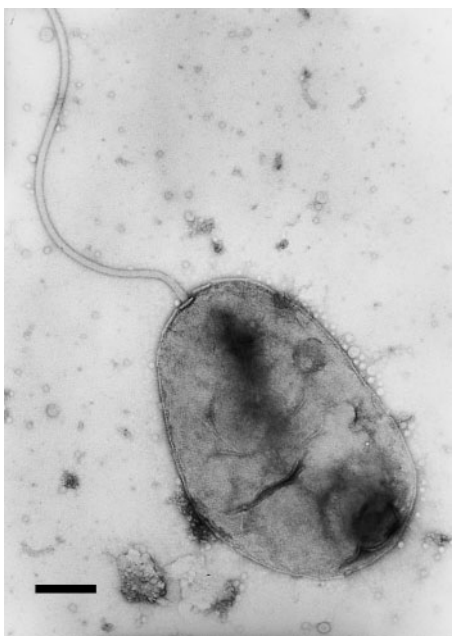

Fig. 1. Electron micrograph of a negatively stained cell of strain YB1^T. Bar, 0·2 μm.

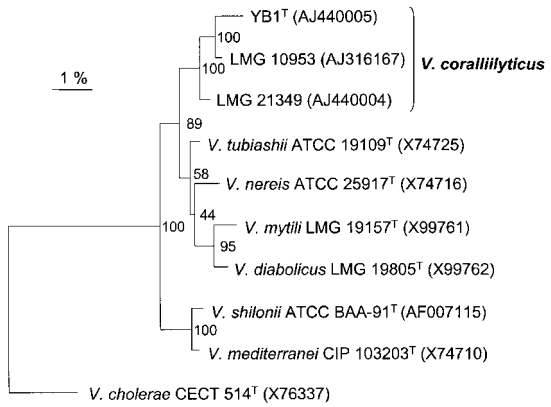

Fig. 2. Phylogenetic tree showing the relationships between *V. coralliilyticus* sp. nov. and other representative type strains of the genus *Vibrio*, based on the neighbour-joining method with almost-complete 16S rDNA sequences. Bootstrap percentages after 1000 simulations are shown. Bar, 1 % estimated sequence divergence.

DNA–DNA hybridization data (Table 2) confirmed that the six isolates form a single genomic group, with DNA–DNA similarities higher than 85 %. DNA–DNA similarity to *V. nereis* and *V. tubiashii* was respectively only 31 and 27 %. DNA–DNA similarities to *V. shilonii*, *Vibrio diabolicus* and *Vibrio mytili* were lower than 24 %. The G+C content of their DNA ranged from 44·9 to 45·6 mol%.

The novel species conforms to the description of the family *Vibrionaceae* and of the genus *Vibrio* (Farmer & Hickman-Brenner, 1992). The cellular fatty acid compositions of the six novel strains are presented in Table 3. The novel strains had similar overall fatty acid profiles, congruent with those known for the family *Vibrionaceae* (Bertone *et al.*, 1996; Urdaci *et al.*, 1990). Differences in biochemical tests and carbon compound utilization between the six isolates are shown in Table 4. Of the 117 tests performed, the strains gave the same qualitative result in 110 cases. Strains YB1^T and LMG 21350 gave identical results in all 117 phenotypic tests, although YB1^T was isolated from the Indian Ocean and LMG 21350 from the Red Sea. The other strains differed from YB1^T in only one to five of the 117 tests performed.

Strains of the novel species can be differentiated easily from their closest phylogenetic neighbours (Table 5). For

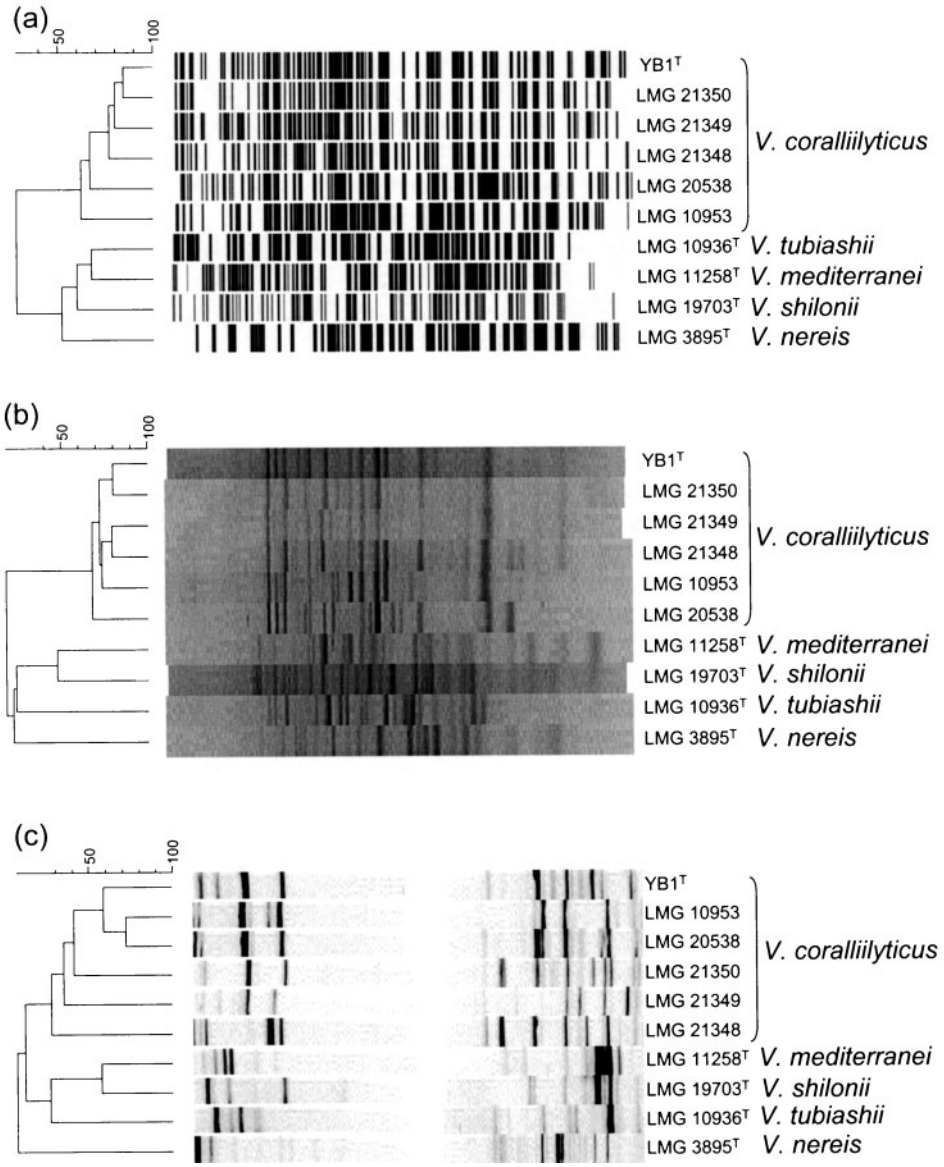

Fig. 3. (a) AFLP, (b) BOX and (c) GTG_5 clustering analysis of *V. coralliilyticus* sp. nov. ($n=6$) and its closest phylogenetic neighbours.

Table 2. DNA similarity among strains of *V. coralliilyticus* sp. nov., *V. nereis* and *V. tubiashii*

Values for DNA similarity are means ± SD of reciprocal DNA–DNA hybridization experiments.

Strain	G+C content (mol%)	DNA–DNA hybridization (%) with:					
		1	2	3	4	5	6
V. coralliilyticus sp. nov.							
1. YB1^T (=LMG 20984^T)	45·6	100					
2. LMG 20538	45·1	86±6	100				
3. LMG 10953	44·9	98±13	97±5	100			
4. LMG 21349	45·6	87±1	88±3	89±1	100		
5. *V. nereis* LMG 3895^T	45·6	27±5	31±2	30±4	30±6	100	
6. *V. tubiashii* LMG 10936^T	45·7	26±3	27±3	26±3	26±5	25±1	100

Table 3. Fatty acid profiles of strains of *V. coralliilyticus* sp. nov.

Values are percentages of total fatty acids.

Fatty acid	YB1^T	LMG 21348	LMG 21349	LMG 21350	LMG 10953	LMG 20538
12 : 0	2·2	3·4	3·8	3·8	2·8	3·6
12 : 0 3-OH	2·7	3·1	3·9	3·7	2·0	2·8
13 : 0 iso	2·6	2·3	2·6	2·4	2·8	1·7
14 : 0	6·5	8·1	7·8	8·3	6·9	8·5
14 : 0 iso	0·5	0·2	0·3	0·2	0·7	0·8
15 : 0	1·3	1·8	2·0	1·9	1·8	2·8
15 : 0 iso	2·8	1·1	1·3	1·5	2·3	1·0
15 : 0 iso 3-OH	1·7	1·3	1·5	1·4	1·1	0·6
16 : 0	14·1	15·2	14·0	15·5	14·6	16·5
17 : 0	2·0	2·6	3·0	2·4	v2·5	2·8
17 : 0 iso	3·2	1·7	1·8	2·0	2·4	0·9
17 : 1ω8*c*	1·3	1·7	2·1	1·7	1·6	2·1
18 : 1ω7*c*	18·2	20·6	18·0	18·6	19·1	14·5

instance, the isolates utilized D-serine, while *V. tubiashii*, *V. nereis* and *V. shilonii* do not. Furthermore, the isolates utilized *m*-inositol, methyl β-D-glucoside, α-ketobutyric acid and alaninamide, whereas *V. tubiashii* and *V. nereis* do not. In contrast to the novel species, *V. shilonii* is negative for utilization of D-gluconic acid, L-leucine and D-serine. Moreover, the novel species possesses larger amounts of the fatty acids 17 : 0 and 17 : 0 iso than do *V. tubiashii* and *V. nereis* and larger amounts of 17 : 0 and 18 : 1ω7*c* than does *V. shilonii*.

Because of the high genotypic and phenotypic similarities between the six novel *Vibrio* isolates, we examined the ability of each isolate to infect the coral *P. damicornis* in controlled aquarium experiments. All six strains were pathogenic, causing similar tissue damage within 2 weeks at 29 °C, whereas *P. damicornis* corals infected with *V. shilonii* under similar experimental conditions remained healthy.

DISCUSSION

Vibrio sp. YB1^T has been reported to be the aetiological agent of tissue damage to the coral *P. damicornis* (Ben-Haim & Rosenberg, 2002). From the data presented here, the strain clearly represents a novel *Vibrio* species. During the course of this investigation, three additional pathogenic *Vibrio* strains (LMG 21348, LMG 21349, LMG 21350) were isolated from diseased *P. damicornis* in the Red Sea and two other

Table 4. Differences in biochemical tests and carbon compound utilization between strains of *V. coralliilyticus* sp. nov.

All strains show the following properties: oxidase- and catalase-positive; acid is produced from glucose; nitrate is reduced to nitrite; indole is produced; positive reactions for β-glucosidase, β-galactosidase and gelatinase; urease-negative; utilize D- and L-alanine, L-histidine, hydroxyproline, L-ornithine, L-proline, D-serine, thymidine, D-mannitol, L-aspartic acid, L-histidine, α-aminobutyric acid, D-galactose, methyl succinate, D-gluconic acid, succinic acid, bromosuccinic acid, alaninamide, L-serine, L-threonine, uridine, glycerol, glucose 6-phosphate, dextrin, glycogen, DL-α-glycerol phosphate, Tweens 40 and 80, *N*-acetylglucosamine, D-fructose, maltose, D-mannose, D-psicose, sucrose, L-alanyl glycine, L-asparagine, L-glutamic acid, glycyl L-aspartic acid and glycyl L-glutamic acid; susceptible to O/129, erythromycin, tetracycline, chloramphenicol and gentamicin; resistant to kanamycin, ampicillin and penicillin; growth in 1–7 % NaCl; no growth in the absence of NaCl or in the presence of ⩾8 % NaCl. Except for the tests included in the table, all six strains gave the same qualitative results with the API-20 NE system and Biolog GN2 MicroPlate tests.

Characteristic	YB1^T	LMG 10953	LMG 20538	LMG 21348	LMG 21349	LMG 21350
Arginine dehydrolase	+	−	−	−	−	+
Utilization of:						
Citrate	+	+	+	+	−	+
Methyl β-D-glucoside	−	−	−	+	+	−
Propionic acid	+	+	+	+	−	+
Methyl pyruvate	+	+	+	−	+	+
Glucose 1-phosphate	+	+	+	+	−	+
D-Mannitol	+	−	+	+	+	+

Table 5. Differentiating phenotypic features of *V. coralliilyticus* sp. nov. and the most closely related species

Fatty acid methyl ester (FAME) analyses included six strains of *V. coralliilyticus* sp. nov., four of *V. tubiashii*, two of *V. nereis* and five of *V. shilonii*. Values are means ± SD expressed as percentages of total fatty acids. Phenotypic data for *V. nereis* and *V. tubiashii* were obtained from Holt *et al.* (1994). V, Variable.

Feature	*V. coralliilyticus*	*V. tubiashii*	*V. nereis*	*V. shilonii*
Utilization of:				
Cellobiose	−	+	−	+
D-Galactose	+	+	−	+
m-Inositol	+	−	−	V
Methyl β-D-glucoside	+	−	−	+
Acetic acid	+	−	V	+
cis-Aconitic acid	+	−	+	+
D-Gluconic acid	+	+	+	−
β-Hydroxybutyric acid	−	V	+	V
α-Ketobutyric acid	+	−	−	V
Alaninamide	+	−	−	+
L-Histidine	+	−	V	+
Hydroxy-L-proline	+	−	+	+
L-Leucine	+	−	+	−
D-Serine	+	−	−	−
Fermentation of:				
Amygdalin	−	+	+	+
Melibiose	−	V	−	V
Growth on 8 % NaCl	−	V	+	V
FAME composition:				
17 : 0	2·6 ± 0·3	0·5 ± 0·6	1·2 ± 1·1	0·2 ± 0·0
17 : 0 iso	2·0 ± 0·8	0·0	0·4 ± 0·5	4·2 ± 2·1
17 : 1ω8*c*	1·8 ± 0·3	1·0 ± 1·4	2·7 ± 2·8	0·6 ± 0·6
18 : 1ω7*c*	18·2 ± 2·0	21·6 ± 5·2	21·6 ± 1·5	12·1 ± 1·9

strains, LMG 20538 and LMG 10953, present in the BCCM/LMG Bacteria Collection, were also shown to be closely related to *Vibrio* sp. YB1^T. These six strains form a tight cluster, based on genotypic and phenotypic properties, that is significantly different from other *Vibrio* species. Based on these results, we propose the name *Vibrio coralliilyticus* sp. nov. for this novel pathogenic species.

One of the most interesting findings of this study was the demonstration of the power of bacterial taxonomy to uncover novel coral-pathogenic strains. *Vibrio* strains LMG 10953 and LMG 20538, respectively isolated from diseased larvae of *Crassostrea gigas* and *Nodipecten nodosus*, were shown to be pathogenic to the coral *P. damicornis*. Strain LMG 10953 was previously identified as *V. tubiashii* (Hada *et al.*, 1984). Our results clearly demonstrate that this strain belongs to *V. coralliilyticus* sp. nov.

The fact that coral-pathogenic strains of *V. coralliilyticus* are widely distributed is important in understanding the source of coral disease and attempts to prevent their spread. *V. coralliilyticus* sp. nov. infects and causes tissue damage to its host coral only at water temperatures above 25 °C. It is likely that during the winter, when temperatures are lower, the bacterium is present in different hosts, possibly bivalve larvae. Clearly, the taxonomic studies described here open new and unexpected avenues of investigation regarding the transmission of coral diseases.

Description of *Vibrio coralliilyticus* sp. nov.

Vibrio coralliilyticus (co.ral.li.i.ly′ti.cus. L. n. *corallium* coral; Gr. adj. *lytikos* dissolving; N.L. adj. *coralliilyticus* coral-dissolving).

Conforms to the descriptions of the family *Vibrionaceae* and of the genus *Vibrio* (Farmer & Hickman-Brenner, 1992). Cells are Gram-negative, non-spore-forming rods (1·2–1·5 × 0·8 μm) that are motile by a single polar, sheathed flagellum when grown on solid or liquid medium. Colonies are cream-coloured on marine agar and yellow on TCBS agar after 48 h incubation at 30 °C, have smooth edges and do not luminesce. The predominant cellular fatty acids are 16 : 0 and 18 : 1ω7*c*. Biochemical tests and carbon compound utilization data are summarized in Table 4. The type strain, strain YB1^T (= LMG 20984^T = ATCC BAA-450^T), was isolated from a diseased coral of the species *Pocillopora damicornis* in the Indian Ocean. Further strains have been isolated from diseased *P. damicornis* in the Red Sea and from larvae of oysters in the Atlantic Ocean. All six strains described here are pathogenic to the coral

P. damicornis. The G+C content of the type strain is 45·6 mol%.

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