Extremophiles (2015) 19:721–740
DOI 10.1007/s00792-015-0760-3

REVIEW

Microbial diversity and adaptation to high hydrostatic pressure in deep-sea hydrothermal vents prokaryotes

Mohamed Jebbar[1,2,3,8] · **Bruno Franzetti**[4,5,6] · **Eric Girard**[4,5,6] · **Philippe Oger**[7]

Received: 26 February 2015 / Accepted: 26 May 2015 / Published online: 23 June 2015

Abstract Prokaryotes inhabiting in the deep sea vent ecosystem will thus experience harsh conditions of temperature, pH, salinity or high hydrostatic pressure (HHP) stress. Among the fifty-two piezophilic and piezotolerant prokaryotes isolated so far from different deep-sea environments, only fifteen (four *Bacteria* and eleven *Archaea*) that are true hyper/thermophiles and piezophiles have been isolated from deep-sea hydrothermal vents; these belong mainly to the *Thermococcales* order. Different strategies are used by microorganisms to thrive in deep-sea hydrothermal vents in which "extreme" physico-chemical conditions prevail and where non-adapted organisms cannot live, or even survive. HHP is known to impact the structure of several cellular components and functions, such as membrane fluidity, protein activity and structure. Physically the impact of pressure resembles a lowering of temperature, since it reinforces the structure of certain molecules, such as membrane lipids, and an increase in temperature, since it will also destabilize other structures, such as proteins. However, universal molecular signatures of HHP adaptation are not yet known and are still to be deciphered.

Keywords Deep biosphere · Diversity · High hydrostatic pressure · Enzymatic function · Molecular adaptation

Introduction

Hydrostatic pressure increases with depth at an approximate rate of 10 MPa (~100 atmospheres/bars) per km in the water column and 30 MPa per km underground (Oger and Jebbar 2010). The definition of the deep biosphere is conveniently and arbitrarily defined as applying to water depths of 1000 m or more. Consequently, all environments above 10 MPa qualify as high hydrostatic pressure (HHP) biotopes. HHP waters account for 88 % of the volume of the oceans, which have an average depth of 3800 m (and thus an average hydrostatic pressure ca. 38 MPa) but reach 110 MPa in the trenches. The average temperature in the deep ocean is 2°–3°, except for hydrothermal vents. In

Communicated by S. Albers.

✉ Mohamed Jebbar
mohamed.jebbar@univ-brest.fr

[1] Université de Bretagne Occidentale, UMR 6197-Laboratoire de Microbiologie des Environnements Extrêmes (LM2E), Institut Universitaire Européen de la Mer (IUEM), Rue Dumont d'Urville, 29280 Plouzané, France

[2] CNRS, UMR 6197-Laboratoire de Microbiologie des Environnements Extrêmes (LM2E), Institut Universitaire Européen de la Mer (IUEM), Rue Dumont d'Urville, 29280 Plouzané, France

[3] Ifremer, UMR 6197-Laboratoire de Microbiologie des Environnements Extrêmes (LM2E), Technopôle Brest-Iroise, BP70, 29 280 Plouzané, France

[4] Centre National de la Recherche Scientifique, IBS, 38027 Grenoble, France

[5] Université Grenoble Alpes, Institut de Biologie Structurale (IBS), 38027 Grenoble, France

[6] Commissariat à l'Energie Atomique et aux Energies Alternatives, Direction des Sciences du Vivant, IBS, 38027 Grenoble, France

[7] CNRS, UMR 5276, Ecole Normale Supérieure de Lyon, Lyon, France

[8] Laboratoire de Microbiologie des Environnements Extrêmes (UMR 6197), Institut Universitaire Européen de la Mer (IUEM), Technopole Brest-Iroise, Rue Dumont d'Urville, 29280 Plouzané, France

contrast, the average geothermal gradient in the continental system is ca. 25 °C km^{-1}. The currently known temperature limit for life, 122 °C (Takai et al. 2008), would thus place the "deep" limit for the putative continental biosphere at ca. 5 km below ground on average, under maximal pressures of 150 MPa (Zeng et al. 2009; Oger and Jebbar 2010).

HHP affects chemical equilibria and reaction rates, depending on the reaction (ΔV) and activation ($\Delta V\neq$) volumes involved. The behavior of all systems under HHP is governed by Le Châtelier's principle, which states that the application of pressure shifts equilibrium towards the state that occupies the smallest volume. It accelerates a process whose transition state has a smaller volume than that of the ground state. For example, if the volume of a protein is smaller in its unfolded form, then this protein will be denatured by the application of HHP. Several cellular processes such as RNA synthesis, membrane fluidity, motility, cell division, nutrient uptake, membrane protein function, protein synthesis and replication are also impaired by HHP. HHP greater than 200 MPa can kill most microorganisms and is used as a means to preserve foodstuffs.

The discovery of deep-sea hydrothermal vent ecosystems is rather recent in the history of biological sciences (Corliss and Ballard 1977; Paull et al. 1984; Jannasch and Mottl 1985; Eder et al. 1999). The most significant microbial process taking place at these sites is "bacterial chemosynthesis", which contrasts with the well-known process of photosynthesis. Both processes involve the biosynthesis of organic carbon compounds from CO_2, with the source of energy being either chemical oxidations or light, respectively (Jannasch and Mottl 1985). Chemoautotrophic prokaryotes will assimilate CO_2 and is coupled in some prokaryotes with chemolithotrophy, which enables them to reduce some inorganic compounds as energy sources. Due to the mixture between the hot reduced hydrothermal fluids enriched in dissolved gas (H_2S, H_2, CH_4, CO/CO_2) and metals (Fe, Mn) and the cold oxidized sea water containing sulfates and nitrates, a wide variety of electron donors and acceptors are available to supply different microbial metabolisms (Table 1).

Most of the Earth's prokaryotes live in deep biosphere environments under HHP. From global estimates of volume, the upper 200 m of the ocean contains a total of 3.6×10^{28} prokaryotic cells of which 2.9×10^{27} cells are autotrophs; whereas the ocean water below 200 m contains 6.5×10^{28} prokaryotic cells (Whitman et al. 1998). A recent study has estimated the total cell abundance in subseafloor sediment at 2.9×10^{29}, which is 92 % lower than the previous standard estimate (35.5×10^{29}) (Whitman et al. 1998; Kallmeyer et al. 2012). Thus, even though the maximal productivity of the high-pressure continental or marine biosphere is orders of magnitude lower than that of surface biotopes due to their extremely large volume, these high-pressure biotopes contribute significantly to the production and recycling of organic carbon.

In less than 30 years, microbiologists have isolated and described many new microbial species involved in most major biogeochemical cycles and some of which are able to grow at more than 110 °C and 150 MPa. Psychrophiles, mesophiles, hyper/thermophiles, acidophiles, piezophiles and even moderate halophiles were isolated from samples originating from deep-sea hydrothermal vents constituting cultivable representatives of at least 20 phyla, 89 genus and 175 species. This represents a little more than 1 % of ~12,391 prokaryotic cultured type species (451 *Archaea* and 11,940 *Bacteria*) (http://www.bacterio.net/-number.html#notea) whose taxonomy has been well described (Euzéby 2013). These species belong to 35 different phyla (30 bacterial phyla and 5 archaeal phyla). The latest molecular biology techniques allow us to see that the cultured and described species represent only ~1–2 % of the Earth's *Bacteria* and *Archaea* according to the SILVA database (http://www.arb-silva.de), which contains 534,968 of non-redundant 16S rRNA sequences distributed as follows: *Bacteria* (86.9 %), *Archaea* (3.5 %) and *Eukarya* (9.6 %).

This review aims to provide an overview of the diversity of microorganisms thriving in deep-sea hydrothermal vents, their metabolic characteristics and the adaptive mechanisms they have evolved to cope with HHP.

Deep-sea hydrothermal vent ecosystems

Hydrothermalism

Deep-sea hydrothermal vent sites (700–5000 m below the ocean surface) are located in areas of high tectonic activity (Fig. 1) such as areas of accretion along mid-ocean ridges (e.g. the mid-Atlantic Ridge [MAR] or East Pacific Rise [EPR]), subduction zones, the back-arc basins (e.g. the North Fiji or Lau Basins) and finally hot spot volcanism (e.g. Loihi Seamount near Hawaii) (Van Dover et al. 2002). According to the InterRidge Global Database, about 600 active or ancient Submarine Hydrothermal Vent Fields have been recorded in more than 200 locations throughout the Pacific, Atlantic, and Indian Oceans confirmed or inferred (Fig. 1), including 46 locations in Indian Ocean (depth from 1500 to 4200 m), 64 locations in the Atlantic Ocean (depth from 800 to 4530 m) and 100 locations in Pacific Ocean (depth from 850 to 5000 m). The activity of the tectonic plates generates seafloor spreading centers (rifts), regions where hot basalt and magma near the sea floor cause the floor to slowly drift apart. Seawater seeping into these cracked regions mixes with hot minerals (Edmond et al. 1982) and, is emitted from the springs; these underwater hot springs are known as hydrothermal vents. The mineral rich

Table 1 List of known cultured piezotolerant and piezophilic prokaryotes

Isolate	T_{opt} (°C)	P_{opt} (MPa)	Maximal growth rate (h^{-1})	Pressure range (MPa)	Isolation source (depth [m])	GenBank genome sequence accession number	Metabolism	Reference(s)
Domain: *Bacteria*								
Phylum: *Actinobacteria*								
Class *Actinobacteria*								
Order *Actinomycetales*								
Family *Dermacoccaceae*								
Dermacoccus abyssi MT1.1^T	28	40	nd	nd	Mariana Trench, sediment (10,898)		Chemoorganoheterotroph	Pathom-aree et al. 2006
Phylum: *Firmicutes*								
Class: *Bacilli*								
Order *Lactobacillales*								
Family *Carnobacteriaceae*								
Carnobacterium sp. strain AT12	2	20		0.1–60	Alaska, Aleoutiennes Trench (2550)		Chemoorganoheterotroph	Lauro et al. 2007
Carnobacterium sp. strain AT7	20	20	0.75	0.1–60	Central Indian Ridge, black smoker fluid (2415–2460)	ABHH01000000	Chemoorganoheterotroph	Lauro et al. 2007
Class: *Clostridia*								
Order to be established								
Novel family to be established								
Anoxybacter fermentans DY22613^T	60–62	20	2.22	0.1–60	Deep-sea hydrothermal sulfide deposit at the East Pacific Rise (2891)		Chemoorganoheterotroph	Zeng et al. 2015
Phylum: *Proteobacteria*								
Class: *Alphaproteobacteria*								
Alphaproteobacterium without standing nomenclature								
Piezobacter thermophilus 108	50	35	0.46	0.1–65	Mid-Atlantic Ridge, black smoker chimney (3626)		Facultative chemoautotroph	Takai et al. 2009
Order *Rhodobacterales*								
Family *Rhodobacteraceae*								
Rhodobacterales bacterium PRT1	10	80	0.019	20–100	Puerto Rico Trench, seawater (8350)		Chemooligoorganotroph	Eloe et al. 2011
Class: *Deltaproteobacteria*								
Order *Desulfovibrionales*								
Family *Desulfovibrionaceae*								

Table 1 continued

Isolate	T_{opt} (°C)	P_{opt} (MPa)	Maximal growth rate (h^{-1})	Pressure range (MPa)	Isolation source (depth [m])	GenBank genome sequence accession number	Metabolism	Reference(s)
Desulfovibrio profundus 500-1^T	25	10–40	nd	0.1–40	Japan Sea, sediment core 518 mbsfb (900)		Chemoorganoheterotroph	Bale et al. 1997
Desulfovibrio hydrothermalis AM13^T	35	26	0.05	nd	East Pacific Rise, hydrothermal vent chimney (2600)	FO203522	Chemoorganoheterotroph	Alazard et al. 2003
Desulfovibrio piezophilus C1TLV30^T	30	10	0.01	0.1–30	Wood falls in the Mediterranean Sea (1693)	FO203427	Chemoorganoheterotroph	Khelaifia et al. 2011
Class: *Gammaproteobacteria*								
Order *Alteromonadales*								
Family *Colwelliaceae*								
Colwellia piezophila Y223G^T	10	60	0.14	0.1–80	Japan Trench, sediment (6278)	NZ_ARKQ00000000	Chemoorganoheterotroph	Nogi et al. 2004
Colwellia piezophila Y251E^T	10	60	nd	0.1–80	Japan Trench, sediment (6278)		Chemoorganoheterotroph	Nogi et al. 2004
Colwellia hadaliensis BNL-1^T	10	90	0.12	37–102	Puerto Rico Trench (7410)		Chemoorganoheterotroph	Deming et al. 1988
Colwellia sp. strain MT41	8	69	0.07	38–103	Mariana Trench, decaying amphipod (10,476)	GCA_000712155.1	Chemoorganoheterotroph	Yayanos et al. 1981
Family *Psychromonadaceae*								
Psychromonas profunda 2825^T	10	25	0.15	0.1–50	Atlantic Ocean sediment (2770)		Chemoorganoheterotroph	Xu et al. 2003
Psychromonas kaikoae JT7304^T	10	50	0.15	0.1–70	Japan Trench, cold-seep sediment (7434)		Chemoorganoheterotroph	Nogi et al. 2002
Psychromonas sp. strain CNPT3	12	52	0.19	0.1–85	Central North Pacific, decaying amphipod (5800)	CP004404	Chemoorganoheterotroph	Yayanos et al. 1979
Psychromonas hadalis K41G^T	6	60	0.14	0.1–90	Japan Trench, sediment (7542)	NZ_ATUO00000000	Chemoorganoheterotroph	Nogi et al. 2007
Family *Moritellaceae*								
Moritella profunda 2674^T	6	30	0.17	0.1–50	Atlantic Ocean, sediment (2815)		Chemoorganoheterotroph	Xu et al. 2003
Moritella abyssi 2693^T	10	30	0.20	0.1–50	Atlantic Ocean, sediment (2815)		Chemoorganoheterotroph	Xu et al. 2003
Moritella sp. strain PE36	10	41	0.28	0.1–70	Pacific Ocean, amphipod trap water (3584)	ABCQ00000000	Chemoorganoheterotroph	Yayanos et al. 1986

Table 1 continued

Isolate	T_{opt} (°C)	P_{opt} (MPa)	Maximal growth rate (h^{-1})	Pressure range (MPa)	Isolation source (depth [m])	GenBank genome sequence accession number	Metabolism	Reference(s)
Moritella japonica DSK1	15	50	0.4	0.1–70	Japan Trench, sediment (6356)		Chemoorganoheterotroph	Kato et al. 1995
Moritella yayanosii DB21MT-5	10	80	0.2	60–100	Mariana Trench, sediment (10,898)		Chemoorganoheterotroph	Nogi et Kato 1999
Family *Shewanellaceae*								
Shewanella piezotolerans WP2	15–20	15	0.5	0.1–50	West Pacific, sediment (1914)	NC_011566	Chemoorganoheterotroph	Xiao et al. 2007
Shewanella piezotolerans WP3	15–20	20	0.5	0.1–50	West Pacific, sediment (1914)	NC_011566	Chemoorganoheterotroph	Xiao et al. 2007
Shewanella profunda LT13a	25–30	10	1.33	0.1–50	Pacific Ocean Nankai Trough, sediment (4790)		Chemoorganoheterotroph	Toffin et al. 2004
Shewanella violacea DSS12	10	30	0.28	0.1–70	Ryukyu Trench, sediment (5110)	NC_014012	Chemoorganoheterotroph	Kato et al. 1995
Shewanella benthica F1A	8	30	0.15	0.1–70	Atlantic Ocean, water column (4900)		Chemoorganoheterotroph	Wirsen et al. 1986
Shewanella benthica DB6101	10	50	0.35	0.1–70	Ryukyu Trench sediment (5110)		Chemoorganoheterotroph	Kato et al. 1995
Shewanella benthica DB5501	15	60	0.35	0.1–70	Suruga Bay, sediment (2485)		Chemoorganoheterotroph	Kato et al. 1995
Shewanella benthica DB6705	15	60	0.4	0.1–70	Japan Trench, sediment (6356)		Chemoorganoheterotroph	Kato et al. 1995
Shewanella benthica DB6906	15	60	0.35	0.1–70	Japan Trench, sediment (6269)		Chemoorganoheterotroph	Kato et al. 1995
Shewanella benthica DB172R	10	60	0.45	0.1–70	Izu-Bonin Trench, sediment (6499)		Chemoorganoheterotroph	Kato et al. 1996
Shewanella benthica DB172F	10	70	0.41	50–100	Izu-Bonin Trench, sediment (6499)		Chemoorganoheterotroph	Kato et al. 1996
Shewanella benthica DB21MT-2	10	70	0.17	60–100	Mariana Trench sediment (10,898)		Chemoorganoheterotroph	Kato et al. 1998
Shewanella benthica KT99	2	98		40–140	Kermadec Trench, amphipod homogenate (9856)	ABIC00000000	Chemoorganoheterotroph	Lauro et al. 2007
Order *Chromatiales*								
Family *Thioalkalispiraceae*								
Thioprofundum lithotrophica 106	50	15	0.3	0.1–50	Mid-Atlantic Ridge, black smoker chimney (3626)		Chemolithoautotrophic	Takai et al. 2009

Table 1 continued

Isolate	T_{opt} (°C)	P_{opt} (MPa)	Maximal growth rate (h^{-1})	Pressure range (MPa)	Isolation source (depth [m])	GenBank genome sequence accession number	Metabolism	Reference(s)
Order *Oceanospirillales*								
Family *Oceanospirillaceae*								
Profundimonas piezophila YC-1	8	50	0.017	20–70	Puerto Rico Trench, water column (6000)		Chemoorganoheterotroph	Cao et al. 2014
Order *"Vibrionales"*								
Family *Vibrionaceae*								
Photobacterium profundum DSJ4	10	10	0.45	0.1–70	Ryukyu Trench, sediment (5110)		Chemoorganoheterotroph	Nogi et al. 1998
Photobacterium profundum SS9	15	28	0.5	0.1–70	Sulu Trough, amphipod homogenate (2551)	NC_006370, NC_006371	Chemoorganoheterotroph	De Long et al. 1997
Phylum: *Thermotogae*								
Class *Thermotogae*								
Order *Thermotogales*								
Family *Thermotogaceae*								
Thermosipho japonicus IHB 1^T	72	20	1.33	0.1–60	Okinawa, Iheya (972)		Chemoorganoheterotroph	Takai and Horikoshi 2000
Marinitoga piezophila $KA3^T$	65	40	1.9	0.1–60	East Pacific Rise, hydrothermal vent (2630)	CP003257	Chemoorganoheterotroph	Alain et al. 2002a
Domain: *Archaea*								
Phylum: *Euryarchaeota*								
Class *Methanococci*								
Order *Methanococcales*								
Family *Methanocaldococcaceae*								
Methanocaldococcus jannaschii JAL-1^T	86	75	2.36	0.1–75	East Pacific Rise, hydrothermal vent (2,610)	NC_000909	Methanogenesis	Jones et al. 1983
Family *Methanococcaceae*								
Methanococcus thermolithotrophicus	65	50	0.58	0.1–100	Italy, geothermally heated sediments (0.5)	NZ_AQXV01000000	Methanogenesis	Bernhardt et al. 2007
Class *Methanopyri*								
Order *Methanopyrales*								
Family *Methanopyraceae*								
Methanopyrus kandleri 116	105	20	0.73	0.1–50	Aleutian Trench, water column (2500)	NC_003551	Methanogenesis	Takai et al. 2008

Table 1 continued

Isolate	T_{opt} (°C)	P_{opt} (MPa)	Maximal growth rate (h^{-1})	Pressure range (MPa)	Isolation source (depth [m])	GenBank genome sequence accession number	Metabolism	Reference(s)
Class *Thermococci*								
Order *Thermococcales*								
Family *Thermococcaceae*								
Palaeococcus ferrophilus DMJ^T	83	30	0.5	0.1–60	The Myojin Knoll in the Ogasawara-Bonin Arc, Japan (1338)	Ongoing	Chemoorganoheterotroph	Takai et al. 2000
Palaeococcus pacificus DY20341^T	80	30	0.91	0.1–80	East Pacific Ocean hydrothermal field (2737)	CP006019	Chemoorganoheterotroph	Zeng et al. 2013
Pyrococcus abyssi GE5	96	20	0.98	0.1–50	Fiji Basin, hydrothermal vent (2000)	NC_000868	Chemoorganoheterotroph	Erauso et al. 1993
Pyrococcus yayanosii CH1^T	98	52	1.2	20–120	Mid-Atlantic Ridge, hydrothermal vent (4100)	NC_015680	Chemoorganoheterotroph	Zeng et al. 2009
Thermococcus aggregans TY^T	75	20		0.1–30	Guaymas bassin (2000)		Chemoorganoheterotroph	Canganella et al. 2000
Thermococcus guaymasensis TYS^T	85	20–35		0.1–50	Guaymas bassin (2000)		Chemoorganoheterotroph	Canganella et al. 1998
Thermococcus peptonophilus DSM 10343^T	90	45		0.1–60	Back-arc Bonin, Izu (1380)		Chemoorganoheterotroph	Canganella et al. 1997
Thermococcus eurythermalis A501^T	85	0.1–30	1.25	0.1–70	an oil-immersed hydrothermal chimney, Guaymas Basin (2000)	CP008887, CP008888	Chemoorganoheterotroph	Zhao et al. 2015
Thermococcus barophilus MP^T	85	40	1.5	0.1–80	Mid-Atlantic Ridge, hydrothermal vent chimney (3550)	CP002372, CP002373	Chemoorganoheterotroph	Marteinsson et al. 1999

n.d. not determined

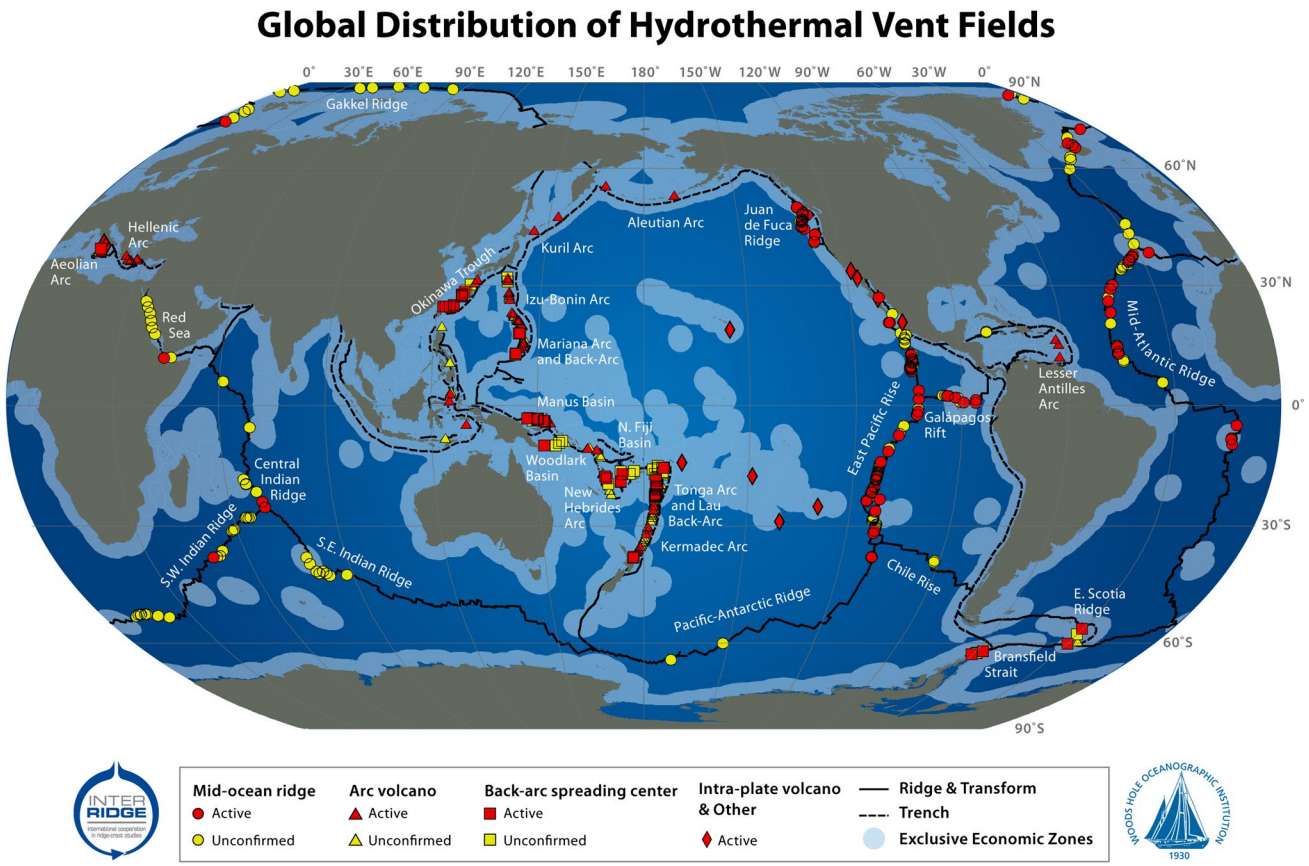

Fig. 1 Global map of hydrothermal vents identified so far according to data compiled in Interridge vents database (http://vents-data.interridge.org/maps) [Beaulieu SE (2013) InterRidge Global Database of Active Submarine Hydrothermal Vent Fields: prepared for InterRidge, Version 3.1. World Wide Web electronic publication. Version 3.2. http://vents-data.interridge.org. Accessed 2015-02-11]

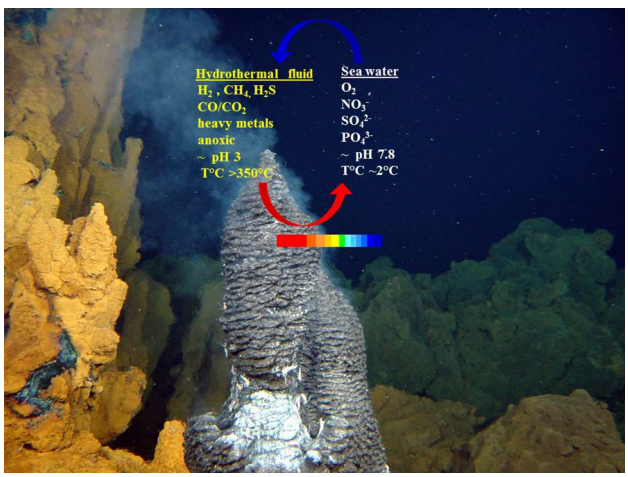

Fig. 2 Illustration showing mixing between the hot hydrothermal fluid enriched in dissolved reduced gas as H_2, CH_4 H_2S, CO, CO_2 and metals etc. (electron donors) and cold seawater that contains O_2, SO_4^{-2} and NO_3^- (electron acceptors) that constitutes the basis of "bacterial chemosynthesis" occurring in deep-sea vents. Photograph of a chimney from Rainbow location (Mid-Atlantic Ridge) reproduced with the permission from Ifremer©

hot water (270–460 °C) forms a cloud of precipitated material upon mixing with oxygenated cold seawater (2–3 °C); these hydrothermal vents are called black smokers.

Interactions between deep basaltic or ultramafic rocks and sea water brought to high temperature and high pressure, which therefore has a high solvent power, will produce hydrothermal fluids. These fluids may reach a temperature as high as 460 °C, with an acidic pH. They are anoxic and contain high concentrations of dissolved gases (H_2S, CH_4, CO, CO_2, and H_2) and minerals (Mn^{2+}, Fe^{2+}, Si^+, Zn^{2+}, etc.) (Fig. 2) (Jannasch and Mottl 1985; Johnson et al. 1986; Von Damm 2000; Charlou et al. 2002, 2010). In contact with sea water (cold, oxic with alkaline pH), minerals precipitate and form black smokers.

Metabolic and phylogenetic diversity of deep-sea hydrothermal vents

Many *Bacteria* and *Archaea* can use sulfur, sulfates, thiosulfate, and Iron (III) oxide as electron acceptors in anaerobic respiration and drive metabolisms like sulfur respiration, sulfato respiration and thiosulfate respiration, iron

respiration and Anaerobic Oxidation of Methane (AOM). Fermentative metabolism is also a principal feature of many archaeal and bacterial species isolated from deep-sea hydrothermal vents (Takai and Nakamura 2011).

Except for some *Thaumarchaeota* that perform nitrification, oxygen and nitrate are used as electrons acceptors by almost all bacteria that specific drive metabolisms such as aerobic respiration, hydrogen oxidation, nitrification, methanotrophy and methylotrophy, sulfur compound oxidation, iron oxidation, manganese oxidation, denitrification and Annamox. Some metabolisms like methanogenesis and ammonia oxidation are specifically to *Archaea*. The culture-independent approach was used to describe the microbial phylogenetic diversity in active deep-sea hydrothermal vents from chimney fluid sediments and macrofauna samples. *Archaea* are associated with hydrothermal edifices, fluids and sediments and encompass archaeal groups like *Thermococcales*, *Archaeoglobales*, *Desulfurococcales*, *Ignicoccales*, *Methanococcales* and *Methanopyrales* (Huber et al. 1989; Takai and Horikoshi 1999; Takai et al. 2001; Teske et al. 2002; Schrenk et al. 2004; Nunoura et al. 2010; Roussel et al. 2011; Takai and Nakamura 2011). Other groups like *Halobacteriales*, *Thaumarchaeota*, DHVE2 and MCG divisions were also detected in these ecosystems (Takai et al. 2001; Roussel et al. 2011; Orcutt et al. 2011; Flores et al. 2012).

The *Bacteria* domain is dominated by *Epsilonproteobacteria* detected in hydrothermal fluids, in sea water, hydrothermal sediments, microbial mats and in association with macrofauna (Reysenbach et al. 2000, 2002; Alain et al. 2002b; Teske et al. 2002; Huber et al. 2003; Page et al. 2004; Suzuki et al. 2005; Campbell et al. 2006; Gerasimchuk et al. 2010; Crépeau et al. 2011; Sylvan et al. 2012); other bacterial groups were also detected, such as *Alpha-*, *Beta-*, *Delta* and *Gammaproteobacteria*, *Aquificales*, *Desulfobacteriales*, *Thermotogales*, *Deinococcus-Thermus*, *Deferribacteres*, *Firmicutes*, CFB (*Cytophaga-Flavobacteria-Bacteroidetes*), *Acidobacteria*, *Verrumicrobia* and *Planctomycetes* (Alain et al. 2002a; Reysenbach et al. 2002; Teske et al. 2002; Huber et al. 2003; Page et al. 2004; Crépeau et al. 2011; Orcutt et al. 2011; Sylvan et al. 2012). More than one hundred species of *Bacteria* (17 phyla, 72 genera, 113 species) and *Archaea* (3 phyla, 17 genera and 62 species) were isolated and cultured from deep-sea hydrothermal vents, mainly from the Pacific ocean (69–77 % of species) and, in lesser numbers, from the Atlantic (19–28 % of species) and Indian (2–4 % of species) Oceans.

Knowledge on piezophiles vs other extremophiles from deep-sea hydrothermal vents

The field of piezomicrobiology has been held back by requirements for expensive high-pressure laboratory equipment for sample containment and culture. The first HHP-adapted prokaryotes were bacteria isolated from deep-sea sediments in 1949 by ZoBell and Johnson (Zobell and Johnson 1949). In 1979, the group of Professor Yayanos reported the isolation of the first piezophilic bacterium from the cold deep ocean and 2 years later (Yayanos et al. 1981) isolated the first obligate piezophile microorganism, a psychrophilic bacterium isolated from a decaying amphipod fished from the bottom of the Mariana Trench.

However, although deep-sea hydrothermal vent fields were explored at depths ranging from 800 to 5000 m, rather few attempts to enrich isolates under in situ pressures have been carried out. Almost all hyper/thermophilic vent prokaryotes have been isolated under atmospheric pressure, and few of them have been exposed to HHP. To our knowledge, only a few microorganisms have been described that are both piezotolerant and piezophilic, these include representatives from across both *Archaea* and *Bacteria* domains: *Pyrococcus abyssi* (Erauso et al. 1993) *Thermococcus barophilus* (Marteinsson et al. 1999), *Thermococcus aggregans* (Canganella et al. 2000), *Thermococcus guaymasensis* (Canganella et al. 1998), *Thermococcus peptonophilus* (Canganella et al. 1997), *Thermococcus eurythermalis* (Zhao et al. 2015), *Palaeococcus ferrophilus* (Takai et al. 2000), *Palaeococcus pacificus* (Zeng et al. 2012), *Methanopyrus kandleri* (Takai et al. 2008), *Marinitoga piezophila* (Alain et al. 2002a), *Thermosipho japonicus* (Takai and Horikoshi, 2000), *Thioprofundum lithotrophica*, *Piezobacter thermophilus* (Takai et al. 2009) and *Desulfovibrio hydrothermalis* (Alazard 2003). The first obligate piezophilic anaerobic hyperthermophilic archaeon discovered was *Pyrococcus yayanosii*, isolated from ultramafic a deep-sea hydrothermal vent field named “Ashadze” located on the Mid-Atlantic Ridge at 4100 m depth (Zeng et al. 2009; Birrien et al. 2011). *P. yayanosii*, *T. barophilus* and *M. piezophila* were isolated after enrichment cultures performed under both high temperatures and HHP; they showed the highest growth rates when grown under hydrostatic pressures and their genomes were entirely sequenced and annotated (Jun et al. 2011; Vannier et al. 2011; Lucas et al. 2012). However, when *T. barophilus* and *M. piezophila* grew under atmospheric pressure, their growth rates were lower.

Microbes in the deep-sea hydrothermal environment face contrasted and fluctuating environmental conditions, to which they need to adapt or die. Hydrothermal vents are characterized by large fluctuations in salinity and temperature, from 0.1 to twice the salinity of seawater (Jannasch and Mottl 1985), and from fluid temperatures as high as 460 °C at the heart of the vent, to 2 °C, the average temperature of the surrounding deep ocean waters (Oger and Jebbar 2010). Prokaryotes residing in the vent ecosystem will thus experience situations of heat, cold, acidic,

or salinity stresses. In addition in the deepest parts of the oceans, hydrothermal vent ecosystems are also submitted to extremely HHP, which is known to impact the structure of several cellular components and functions, such as membrane fluidity, protein activity and structure (Oger and Jebbar 2010). Physically the impact of pressure bears resemblance to both a lowering of temperature, since it will reinforce the structure of certain molecules, such as membrane lipids, and an increase in temperature, since it will as well destabilize other structures, such as proteins. These environmental stressors will affect the cell structure and metabolism.

The hyper/thermophiles piezophiles prokaryotes may have evolved a combination of several adaptive mechanisms (compatible solutes accumulation, efficient expression and activity of prefoldins, membrane fluidity maintaining, robust biocatalysts, etc.) to face harsh and fluctuating conditions prevailing in deep-sea hydrothermal vents, thus their cellular processes such as motility, cell division, nutrient uptake, membrane protein function, protein synthesis and replication are not or less impaired by HHP such as it was observed in piezosensitive species *E. coli* or *S. cerevisiae*.

In the following two sections we will, respectively, address a quick overview of the main physiological adaptive strategies of vent prokaryotes and molecular adaptations in the structure of proteins.

Physiology and adaptation

Due to the environmental fluctuations, microorganisms from hydrothermal vents are expected to show strong osmotic adaptation. In a simplistic way, the effect of salinity and heat stresses may be reduced to one factor, e.g. the reduced activity of water within or in the vicinity of the cells, with a number of consequences, e.g. for the inward or outward fluxes of cellular salts and the destabilization of the function and structure of cellular components. Under reduced water activity, proteins fold incorrectly, which reduces or stops protein activity. We know two strategies of osmoadaptation to high temperature or salinity in hyper-thermophilic prokaryotes isolated from shallow or deep-sea hydrothermal vents (da Costa et al. 1998). Extremely halophilic *Archaea* and a few halophilic *Bacteria* accumulate K^+, Na^+ and Cl^- in response to changes in extracellular salinity (Vreeland 1987; Csonka and Hanson 1991; Galinski and Truper 1994), while a common strategy among microorganisms to cope with osmotic stress is the accumulation of low-molecular-mass organic compounds, also known as compatible solutes because they do not interfere with cellular metabolism (Brown 1976; Galinski 1995; Ventosa et al. 1998). Compatible solutes of hyperthermophiles are similar to those used by mesophiles, i.e. sugars, amino acids, polyols. In addition, hyperthermophiles also accumulate solutes little or never encountered in mesophiles such as mannosylglycerate (MG) (Martins et al. 1997), di-myo-1,1′-inositol phosphate (DIP) (Martins et al. 1996), diglycerol phosphate and derivatives of these compounds (da Costa et al. 1998; Santos and da Costa 2001). These two solutes are essentially restricted to thermophiles (Santos and da Costa 2002). MG is accumulated mostly in response to salt stress in *Archaea* such as *Pyrococcus furiosus*, *Pyrococcus horikoshii*, *Thermococcus celer*, *Thermococcus stetteri* or *Thermococcus litoralis* and *Bacteria* such as *Thermus thermophilus* (Martins and Santos 1995; Nunes et al. 1995; Lamosa et al. 1998; Empadinhas et al. 2001). In the same *Archaea*, DIP is accumulated in response to supra-optimal temperature of growth. In the family *Thermococcales*, there are three noticeable exceptions to this shared osmolyte accumulation pattern. First, *Thermococcus kodakarensis*, which does not seem to accumulate MG under heat or salt stress; second, *Pyrolobus fumarii*, which only accumulates DIP as a response to both stresses (Goncalves et al. 2008); and finally *P. ferrophilus*, which lacks the DIP synthesis genes and accumulates only MG as a response to both stresses. MG is mostly accumulated in response to high salinity, in amounts above 0.6 μmol/mg protein, in the genera *Palaeococcus* (Neves et al. 2005), *Pyrococcus* (Martins and Santos 1995; Empadinhas et al. 2001) or *Thermococcus* (Lamosa et al. 1998). DIP is usually accumulated in response to high temperatures in amounts above 1 μmol/mg of protein in *T. celer* (Lamosa et al. 1998) and *P. furiosus* (Martins and Santos 1995). Osmolytes, such as MG or DIP, create a protective shell surrounding the proteins, which helps maintain proper folding and protein functions (Lamosa et al. 2003). Natural osmolytes increase protein thermal stability in vitro (Santos and da Costa 2002). In hyperthermophiles, MG has been shown to preserve protein folding through an increase of protein rigidity (Borges et al. 2002).

In piezophiles, studies devoted to characterizing the role of compatible solutes in response to HHP have been very limited. Interestingly, *P. profundum* accumulates β-hydroxybutyrate (both monomers and oligomers) in response to hydrostatic pressure (Martin et al. 2002), *M. piezophila* accumulates only amino acids (α-glutamate, proline and alanine) under atmospheric pressure, but the role of these compatible solutes in adaptation to HHP has not been investigated (Lamosa et al. 2013).

As the first and ultimate barrier between the intracellular space and the outside world, biological membranes play a fundamental role in the adaptation of microbes to their environment. The function of the membrane is threefold: (1) to act as a physical barrier to regulate inward and outward trafficking, (2) to play a central role in energy storage and processing via ion gradients, and (3) to provide a

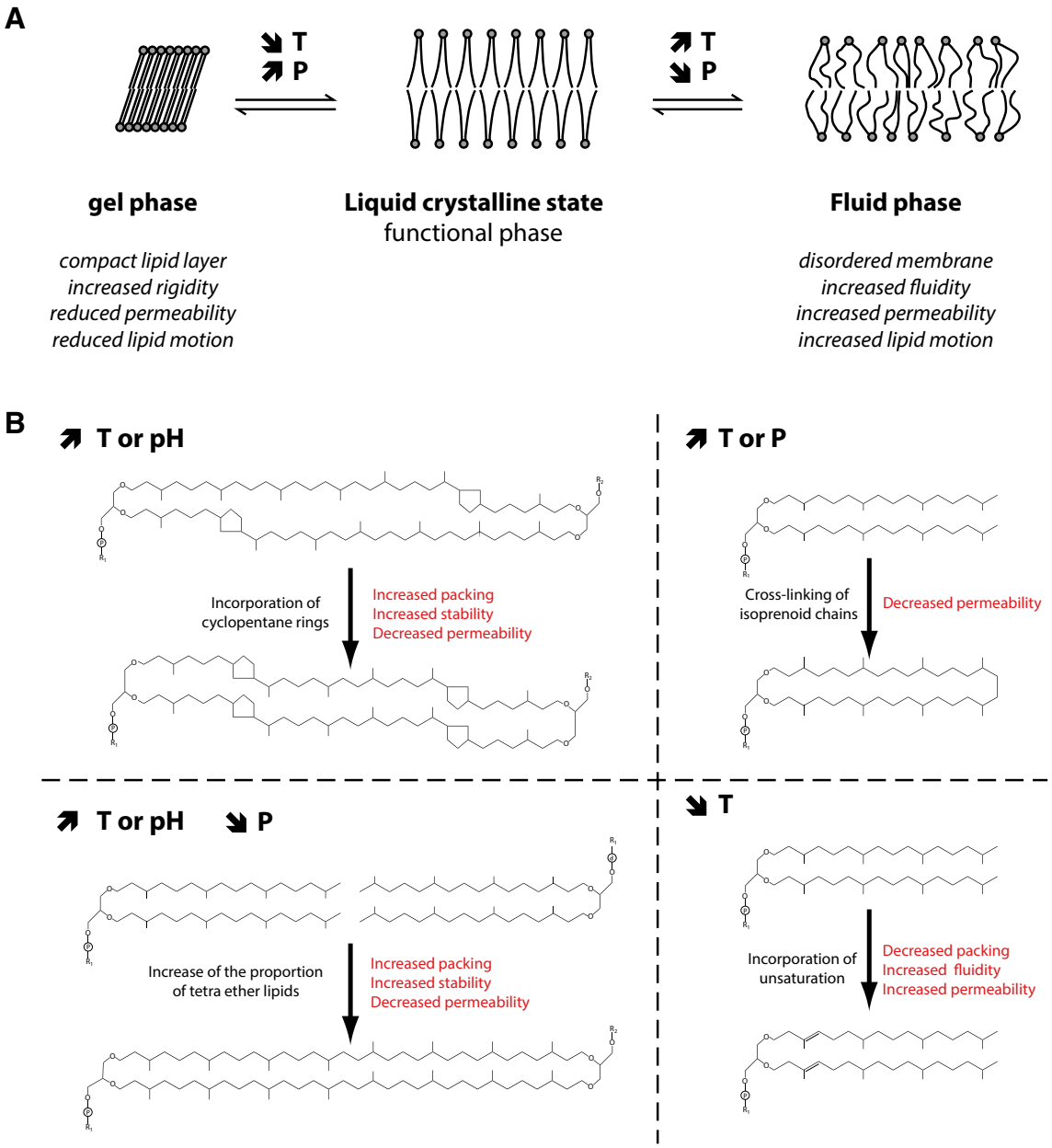

Fig. 3 Homeoviscous adaptation in *Archaea*. **a** In its functional state the membrane is in the liquid crystalline state. Upon an increase in temperature or a decrease in hydrostatic pressure, the lipid motion increases and the membrane enters the fluid phase. Conversely, when temperature drops or hydrostatic pressure increase, the lipid molecules pack more tightly and enter a gel phase. Membranes in both gel phase and fluid phase, have impaired membrane function. **b** Known membrane lipid composition adaptive mechanisms in Archaea

matrix for environmental sensing, multicomponent metabolic and signaling pathways and motility. Thus, maintaining optimal membrane biological function is crucial for any organism. Temperature-, pH-, salinity- or hydrostatic pressure-induced perturbations in membrane organization pose a serious challenge for the cell. Archaeal and bacterial membranes are very dissimilar in structure, although they perform identical functions. However, as will be demonstrated below, the mechanisms employed to adapt to extreme conditions and fluctuating environments are essentially similar. Bacterial polar lipids, apart from a few rare exceptions, are based on straight chain hydrocarbons linked by ester bonds on the *sn*-1 and *sn*-2 positions of glycerol. Archaeal polar lipids are composed of isoprenoid hydrocarbon chains bound by ether bonds to the *sn*-2 and *sn*-3 positions of glycerol (Fig. 3). Polar headgroups consist of phosphodiester-linked polar groups or sugar moieties on the *sn*-1 (*Archaea*) or *sn*-3 (*Bacteria*) positions of the glycerol

backbone (*sn*-glycerol-1-phosphate, or G-1-P, structure and *sn*-glycerol-3-phosphate, or G-3-P, structure).

Based on the observation that the membrane lipids of *E. coli* cells grown under the contrasting temperatures of 43 and 15 °C were different (Marr and Ingraham 1962; Sinensky 1971) while the respective membranes displayed similar physical properties at their respective growth temperatures, Sinensky modeled the basis of homeoviscous adaptation (Sinensky 1974; Oger and Cario 2013). This theory states that organisms adapt their membrane lipid composition to favor the maintenance of the appropriate membrane fluidity for it to function optimally. This concept is now understood in a broader sense to include adaptation to proton/water permeability and the dynamic nature of the plasmic membranes (McElhaney 1984a, b; van de Vossenberg et al. 1999; Oger and Cario 2013). Homeoviscous adaptation must also be understood as a means to rapidly adapt the composition, and thus the functionality, of the membrane to brutal environmental fluctuations, or aggressions, including those of heat and cold, salinity, osmotic stress, pressure and pH.

Under normal physiological conditions, membranes are relatively fluid, disordered liquid crystalline phases. When the temperature drops, or hydrostatic pressure increases, the membrane lipids may undergo the fluid to gel phase transition. When the temperature increases above the physiological conditions, or the pressure decreases from these, the rate of motion of lipids in the membrane will be increased, this may impact membrane stability and intrinsic permeability. As can be expected, perturbation in lipid phase state has profound consequences on membrane structure and function (Lee 2003, 2004). Transition to the gel phase may induce the clustering of membrane proteins, which appear de facto, excluded from the zones in the gel phase, reducing the diffusion and the activity of proteins in the membrane and slowing the flux of transported solutes, but increasing the permeability to cations and water.

Adaptation of bacterial membrane properties follows four major routes. (1) The variation of acyl chain length: an increase of the chain length by two carbons increases the phase transition temperature of the lipid by 10–20 °C, and decreases membrane permeability to proton and water (Winter 2002). (2) The accumulation of unsaturated fatty acids: the incorporation of a single unsaturation can shift the fluid/gel phase transition by 10–20 °C (Russell and Nichols 1999; Winter 2002). (3) The accumulation of specific polar headgroups such as phosphatidylcholine (PC) or phosphatidylglycerol (PG) in place of phosphatidylethanolamine (PE). The presence of PC as a polar headgroup results in a drastic shift of the fluid/gel transition temperature (Yano et al. 1998; Winter 2002; Mangelsdorf et al. 2005; Winter and Jeworrek 2009). This is due in part to the reduced hydration and stearic bulk of the ethanolamine compared to choline, and the capacity of PE, and incapacity of PC, groups to form hydrogen bonds. (4) The accumulation of branched fatty acid.

Archaeal lipid membranes generally have much lower phase transition temperature than fatty acyl ester lipids (Yamauchi et al. 1993). Part of the adaptation of the archaeal membrane to extreme environments may originate from the original structure of its lipids. While membranes made of fatty acyl ester lipids are in the gel phase or in the liquid crystalline phase depending mostly on their fatty acid composition, archaeol- and caldarchaeol-based polar lipid membranes of *Archaea* are assumed to be in the liquid crystalline phase at a wide temperature range of 0–100 °C (Stewart et al. 1990; Dannenmuller et al. 2000). In addition, most if not all *Archaea* from the hydrothermal environments synthesize membrane-spanning bipolar tetraether lipids that form monolayers. The monolayer organization provides extreme rigidity to these membranes. Lateral mobility studies demonstrate that the lateral diffusion rate at 80 °C is comparable between *Sulfolobus acidocaldarius* or *T. acidphilum* and *E. coli* at 37 °C. However, in contrast to bacterial lipids, which exhibit similar lateral diffusion rates for temperatures close to the phase transition temperature, in *T. acidphilum* these values are observed approximately 65 °C above the nominal lipid phase transition temperature (Jarrell et al. 1998).

The adaptation of archaeal membrane properties is very similar in its physics to that of bacteria, although it takes slightly different routes to converge to the same effects. There exist several different routes, as follows. (1) The incorporation of cyclopentane rings along the isoprenoid chain as a function of fluctuating temperature (De Rosa et al. 1980a, b; Ernst et al. 1998; Uda et al. 2001, 2004) or pH (Shimada et al. 2008) increases the packing efficiency of the membrane lipids (Gliozzi et al. 1983), which increases membrane stability as a function of increasing temperature or salinity and decreasing pressure or pH, and consequently lowers the permeability (Chong et al. 2012). (2) The regulation of the tetraether-to-diether lipid ratio (Sprott et al. 1991; Lai et al. 2008; Matsuno et al. 2009): increase in tetraether lipids will stabilize the membranes by forming monolayer-type membranes or domains in the membrane, helping to regulate the flux of solutes and protons across the membrane. (3) The crosslinking of the two acyl-chains of the lipids to yield macrocyclic archaeol or caldarchaeol derivatives by a covalent bond between the isoprenoid chains reduces the motion of the molecule creating a more closely-packed structure and increasing the membrane stability, creating an efficient barrier against water, proton and solute leakage (Dannenmuller et al. 2000; Mathai et al. 2001). (4) The increase in unsaturation along the isoprenoid chains of the lipids as a function of temperature (Nichols et al. 2004) or salinity (Dawson et al.

2012) has to date only been described in the psychrophilic methanogen *Methanococcoides burtonii* (Franzmann et al. 1992; Nichols et al. 2004), but unsaturated lipids have been characterized in several species of hyperthermophiles (Hafenbradl et al. 1993; Gonthier et al. 2001), which might indicate the occurrence of a similar adaptive strategy in hydrothermal vent organisms.

Adaptation of bacterial and archaeal membranes to the harsh environment of the hydrothermal system is clearly visible in the lipids most commonly present. However, responding to the variations of environmental stressors might only involve a fraction of the adaptive traits mentioned above. Indeed, in order to be efficient, the membrane composition adaptation response needs to be very quick. The routes described require different timeframes. Thus, certain adaptive mechanisms will prevail over others. For example, the increasing unsaturation of membrane lipids will decrease the gel/fluid transition temperature to the same extent as the shortening of one of the acyl chains, or the substitution of a phosphatidylcholine by a phosphatidylethanolamine polar head, but will be quicker because it is performed inside the cytoplasmic membrane on existing lipids by a membrane protein (Kasai et al. 1976; Cybulski et al. 2002; Aguilar and de Mendoza 2006; Beranova et al. 2008), while the other actions would need de novo lipid synthesis.

Molecular adaptation of deep-sea enzymes

While extremophilic adaptation has been extensively studied for temperature and salt adapted enzymes, very little is known about pressure adaptation. Pressure-induced modifications of the protein volume may arise from a global elastic compression within closely related conformational sub-states or from promotion of conformational sub-states presenting lower specific volumes, up to complete unfolding of the protein (Fourme et al. 2006; Akasaka 2006; Akasaka et al. 2013). These modifications originate from changes in the various interactions between amino acids (hydrogen bond, ionic, van der Waals and hydrophobic interactions), from changes in the internal voids and cavities found in proteins, and from changes in the hydration properties (Li et al. 1998; Marchi and Akasaka 2001; Boonyaratanakornkit et al. 2002; Refaee et al. 2003; Girard et al. 2005; Nisius and Grzesiek 2012). As protein folding, substrate recognition, protein–protein interactions and protein hydration rely upon a combination of these various interactions, pressure may affect all protein mechanisms through its influence on them (Masson et al. 2004; Occhipinti et al. 2006; Ohmae et al. 2008; Rosenbaum et al. 2012).

Studies that link pressure-induced structural modifications to alterations or stimulations of biological functions are rare. However, it was shown that, for example, pressure induces minor changes in the orientation of a chromophore in the protein citrine, leading to a pressure-induced continuous shift of the fluorescence peak (Barstow et al. 2008). The effect of pressure on the internal solvent-excluded void volume has been proposed as a major contribution to protein activity (Fourme et al. 2006; Girard et al. 2010), but mainly in protein pressure-induced unfolding (Collins et al. 2005; Rouget et al. 2011; Roche et al. 2012). Indeed, the role of a hydrophobic cavity, positioned close to the active site pocket, has been proposed to play a major role in urate oxidase (Girard et al. 2010). Pressure induced an expansion of the active site pocket correlated with a volume reduction of the hydrophobic cavity, leading to an inactivation of the protein. It was proposed (Girard et al. 2010) and confirmed (Marassio et al. 2011) that this particular cavity acts as ballast, providing the required plasticity of the active site pocket during the urate oxidase catalytic process. A recent study (Roche et al. 2012) provided evidence that cavities that are present in the folded state and absent in the unfolded state make a large contribution to the volume difference between folded and unfolded states that govern pressure-induced unfolding of proteins. Such internal cavities might also play a role in the pressure dissociation of protein oligomers and aggregates (Foguel et al. 2003; Girard et al. 2010).

In general, monomeric proteins are quite resistant to high pressure and do not undergo denaturation under a pressure of 300 MPa (Robb and Clark 1999; Sun et al. 1999), but in some cases unfolding has been observed at moderate pressure (Gorovits et al. 1995).

Oligomeric proteins can be dissociated by lower pressure than monomeric proteins; for example, in the GroEl chaperonin complex (14 subunits) from *E. coli*, dissociation occurred at 130 MPa (Gorovits et al. 1995), while the RuBisCO from *Rhodospirillum rubrum* already started to dissociate at 40 MPa (Erijman et al. 1993). Interestingly, these pressure values lie within the physiological range of many deep-sea organisms. However, the picture is more complex than this would make it appear because some oligomeric assemblies have been described to withstand pressure up to 1 GPa for the dimeric protein, bovine erythrocyte Cu, Zn superoxide dismutase (Ascone et al. 2010), up to 400 MPa for the cowpea mosaic virus capsid (Fourme et al. 2002) or up to 300 MPa and 90 °C for the 12-subunit TET aminopeptidase from the archaeon *P. horikoshii* (Rosenbaum et al. 2012).

The existence of a specific molecular adaptation to prolonged exposure to pressure such as the one encountered in the deep sea or in the sediments is suggested by the recent discovery of obligate piezophilic microbes such as *P. yayanosii* (Zeng et al. 2009; Birrien et al. 2011) or *Shewanella benthica* (Lauro et al. 2013). Moreover, as already

mentioned above, piezophilic strains display a typical cellular stress response when they are grown at atmospheric pressure. This suggests that a significant part of the proteome is adapted at the molecular level and, conversely, that the stabilization and enzymatic processes of many proteins must be adapted to colonize the deeper part of the biosphere. As said before, each protein will experience pressure differently depending on its specific 3D structure and the pressure sensitivity of an organism may be controlled by a limited number of proteins.

Most deep-sea organisms are exposed to hydrostatic pressures of 20–110 MPa. Due to the prominent effects of pressure on intermolecular surfaces, it is commonly believed that large molecular systems are significantly affected in this pressure range. It is apparently the case, for instance, for the translational activity that is entirely lost when the pressure was up to 90 MPa (Lu et al. 1997). Pressure-induced dissociation of ribosomes has been considered a major cause of the inhibition of bacterial growth in the deep sea. Cell free assays showed that the post-translocational complex represents the most pressure-sensitive intermediate of the elongation cycle and is possibly the limiting factor for the pressure-induced block of protein biosynthesis (Gross et al. 1993). High hydrostatic pressure (HHP) has also been suggested to influence the structure and function of membrane proteins. The effect of hydrostatic pressure on mitochondrial H^+-ATPase revealed that the complex was inactivated in the pressure range of 60–180 MPa (Dreyfus et al. 1988). The effect of high hydrostatic pressure was also studied on the bacterial mechanosensitive channel. In this case, pressure significantly affected channel kinetics between 0 and 90 MPa. The reduced activity was attributed to a shortening of the channel openings due to lateral compression of the bilayer under high hydrostatic pressure (Macdonald and Martinac 2005). Dimer dissociation of *Vibrio cholerae* ToxR membrane protein was observed at 20–50 MPa in vivo in *E. coli* reporter strains (Macdonald and Martinac 2005). These observations indicated that piezophilic adaptation principally targeted large molecular machines and membrane systems. Accordingly, the volume change upon Actin polymerization was found to be smaller in the case of a deep-sea fish than commonly measured in surface organisms (Morita 2003). The transcriptional activities and the subunit dissociations of the RNA polymerases from the piezophile *Shewanella violacea* and the one of *E. coli* were compared and revealed a better resistance of the deep-sea machinery after pressure treatment (Kawano et al. 2004). All these observations suggest that important structural modifications occur in large supramolecular assemblies that make it possible to adapt to a high-pressure environment. However, the structure determination of such large protein complexes is a technical challenge in structural biology that hampers the comparative structural and biophysical studies of large molecular systems arising from different pressure environments.

Compared to complex cellular machines, the metabolic enzymes represent tractable systems to reveal the structural determinants of piezophilicity. For many enzymes, the biochemistry involves simple substrates and cofactors, and kinetics parameters can be extracted straightforwardly to assess whether or not the reaction is favoured by high pressure. It also exists a consistent knowledge for their structure–function relationships and, in some cases, such as the Lactate-Malate deshydrogenase family, the molecular determinants for thermal or halophilic adaptations have already been explored (Madern et al. 2000; Luke et al. 2007; Sawle and Ghosh 2011). Considering that the stability and the enzymatic properties of most studied mesophilic proteins do not show important alterations in the pressure range that is encountered in the deep sea; the existence of a piezophilic adaptation at the enzyme level remains controversial. However, comparative biochemical studies on enzymes arising from surfaces and deep-sea organisms revealed differences indicating that catalytic reactions are more efficient under high pressure for deep-sea enzymes. The study of tetrameric lactate dehydrogenases (LDH) from hagfish living at different depth showed that pressure inactivation was less pronounced for the abyssal species (Nishiguchi et al. 2008). Several studies were performed on thermophilic enzymes. They revealed an increase in half-life at high temperature with increasing pressure, thus suggesting that high pressure should be taken into account in temperature adaptation (Hei and Clark 1994; Sun et al. 1999; Mombelli et al. 2002). However, these studies focused on residual activities and do not allow us to conclude whether pressure inactivation and thermal stabilization are due to differences in oligomers (Hei and Clark 1994) dissociation, unfolding or subtle structural changes, for example at the active site level. A study on a large aminopeptidase tetrahedral complex (TET) from the deep-sea archaeon *P. horikoshii*, in which small angle X-ray scattering (SAXS) and activity measurements were performed under high pressure and high temperature, showed that the 12-subunits molecular edifice maintains its quaternary structure up to 300 MPa (Rosenbaum et al. 2012). This study suggested that large molecular complexes could maintain high-pressure stability far beyond the pressure that the organisms could encounter in the deepest part of the oceans. Interestingly, the catalytic behaviour of the system was enhanced by pressure. The determination of the volume changes associated with catalysis indicated a change in the reaction rate-limiting step at 180 MPa. This suggested that pressure adaptation could occur at the active site level in this peptidase family. However, comparative studies on different TET peptidases arising from surface and deep-sea organisms are essential to draw a conclusion on this matter. Such

comparative studies have been performed on dihydrofolate reductases (DHFR) from *Shewanella* species living in deep-sea and atmospheric-pressure environments (Ohmae et al. 2012). The stabilities of these enzymes were found to be similar and the pressure effects did not indicate that the catalytic processes of deep-sea DHFR enzymes are better adapted to high-pressure environments. However, 3-isopropylmalate dehydrogenase from the obligate piezophile *Shewanella benthica* DB21MT-2 (SbIPMDH) remains active in extreme conditions whereas that isolated from the land bacterium *S. oneidensis* MR-1 (SoIPMDH) becomes inactivated. Comparison of the structures of the two enzymes shows that SbIPMDH has a larger internal cavity volume than SoIPMDH. This loosely packed structure of SbIPMDH may limit pressure-induced modification of the native structure, keeping it active at higher pressures compared to SoIPMDH (Nagae et al. 2012). This study highlighted the potential role of internal cavity in high-pressure adaptation.

Conclusions

Based on the studies described above, a number of mechanisms explaining pressure resistance and possible adaptation to HHP can be proposed. However, comparative experimental studies combining enzymology, biophysics, structure and microbiology approaches performed under high pressure are still essential to make a decisive statement on the existence of a specific kind of pressure adaptation in deep-sea organisms. As already underlined, proteins are dynamic entities. High-pressure adaptive traits might therefore only be revealed by molecular dynamics studies such as high-pressure crystallography or neutron spectroscopy. Finally, transcriptomic and proteomic studies are vital for revealing the identity of the proteins that are mostly impacted by high or low pressure, either to compensate for losses of cellular functions or as bona fide key players in pressure adaptation.

At this time it is not yet clear if HHP adaptations require just a change of one or a few genes in a few pathways, an overall alteration of many genes in a genome, or mainly regulatory modulations. Several reasons may explain our present inability to identify specific signatures associated with HHP. First, thus far, only piezosensitive (*E. coli*, *S. cerevisiae*) or moderately piezophile strains (*P. profundum* strain SS9, Popt = 28 MPa) have been studied extensively. As a consequence, HHP adaptation, if it exists, might not be sufficient to be demonstrated. Secondly, all results converge to demonstrate a large overlap between cold and HHP regulation.

The diversity of piezophiles isolated so far from deep-sea hydrothermal vents is narrow and consists of prokaryotes driving metabolisms such as sulfatoreduction, sulforeduction, methanogenesis and fermentation, which does not represent the large metabolic diversity encountered in these ecosystems. Autochthonous microorganisms of the deep-sea hydrothermal vents are inherently adapted to the extreme conditions of their environment, i.e. to the high pressure, low to high temperature, low to high pH, anaerobiosis to aerobiosis conditions found throughout the deep-sea hydrothermal vents. Much more effort should be put into isolating new and varied piezophiles from deep-sea hydrothermal vents, including (i) development of adapted and specialized high-pressure equipment to maintain samples at in situ pressure and temperature once onboard ship, (ii) improvement of knowledge and imagination about geochemistry of their ambient environment, extracellular milieu and try to re-create viable laboratory conditions, and (iii) patience, which has sometimes been rewarded by very long-term incubation of cultures.

Acknowledgments This work was supported by the Agence Nationale de la Recherche (ANR-10-BLAN-1725 01-Living deep). We are indebted to Helen McCombie [Bureau de Traduction de l'Université (BTU), Université de Bretagne Occidentale-Brest] for helpful language improvement.

References

Aguilar PS, de Mendoza D (2006) Control of fatty acid desaturation: a mechanism conserved from bacteria to humans. Mol Microbiol 62:1507–1514

Akasaka K (2006) Probing conformational fluctuation of proteins by pressure perturbation. Chem Rev 106:1814–1835. doi:10.1021/cr040440z

Akasaka K, Kitahara R, Kamatari YO (2013) Exploring the folding energy landscape with pressure. Arch Biochem Biophys 531:110–115. doi:10.1016/j.abb.2012.11.016

Alain K, Marteinsson VT, Miroshnichenko ML et al (2002a) *Marinitoga piezophila* sp. nov., a rod-shaped, thermo-piezophilic bacterium isolated under high hydrostatic pressure from a deep-sea hydrothermal vent. Int J Syst Evol Microbiol 52:1331–1339

Alain K, Olagnon M, Desbruyères D et al (2002b) Phylogenetic characterization of the bacterial assemblage associated with mucous secretions of the hydrothermal vent polychaete *Paralvinella palmiformis*. FEMS Microbiol Ecol 42:463–476

Alazard D (2003) *Desulfovibrio hydrothermalis* sp. nov., a novel sulfate-reducing bacterium isolated from hydrothermal vents. Int J Syst Evol Microbiol 53:173–178. doi:10.1099/ijs.0.02323-0

Ascone I, Savino C, Kahn R, Fourme R (2010) Flexibility of the Cu, Zn superoxide dismutase structure investigated at 0.57 GPa. Acta Crystallogr D Biol Crystallogr 66:654–663. doi:10.1107/S0907444910012321

Bale SJ, Goodman K, Rochelle PA et al (1997) Desulfovibrio profundus sp. nov., a novel barophilic sulfate-reducing bacterium from deep sediment layers in the Japan Sea. Int J Syst Bacteriol 47:515–521

Barstow B, Ando N, Kim CU, Gruner SM (2008) Alteration of citrine structure by hydrostatic pressure explains the accompanying spectral shift. Proc Natl Acad Sci USA 105:13362–13366. doi:10.1073/pnas.0802252105

Beranova J, Jemiola-Rzeminska M, Elhottova D et al (2008) Metabolic control of the membrane fluidity in *Bacillus subtilis* during cold adaptation. Biochim Biophys Acta-Biomembr 1778:445–453

Birrien JL, Zeng X, Jebbar M et al (2011) *Pyrococcus yayanosii* sp. nov., an obligate piezophilic hyperthermophilic archaeon isolated from a deep-sea hydrothermal vent. Int J Syst Evol Microbiol 61:2827–2831. doi:10.1099/ijs.0.024653-0

Boonyaratanakornkit BB, Park CB, Clark DS (2002) Pressure effects on intra- and intermolecular interactions within proteins. Biochim Biophys Acta 1595:235–249

Borges N, Ramos A, Raven NDH et al (2002) Comparative study of the thermostabilizing properties of mannosylglycerate and other compatible solutes on model enzymes. Extremophiles 6:209–216

Brown AD (1976) Microbial water stress. Bacteriol Rev 40:803–846

Campbell B, Engel A, Porter M, Takai K (2006) The versatile epsilon-proteobacteria: key players in sulphidic habitats. Nat Rev Microbiol 4:458–468

Canganella F, Gonzalez JM, Yanagibayashi M, Kato C, Horikoshi K (1997) Pressure and temperature effects on growth and viability of the hyperthermophilic archaeon *Thermococcus peptonophilus*. Arch Microbiol 168:1–7

Canganella F, Jones WJ, Gambacorta A, Antranikian G (1998) *Thermococcus guaymasensis* sp. nov. and *Thermococcus aggregans* sp. nov., two novel thermophilic archaea isolated from the Guaymas Basin hydrothermal vent site. Int J Syst Bacteriol 48:1181–1185

Canganella F, Gambacorta A, Kato C, Horikoshi K (2000) Effects of hydrostatic pressure and temperature on physiological traits of *Thermococcus guaymasensis* and Thermococcus aggregans growing on starch. Microbiol Res 154:297–306

Cao Y, Chastain RA, Eloe EA, Nogi Y, Kato C, Bartlett DH (2014) Novel psychropiezophilic Oceanospirillales species *Profundimonas piezophila* gen. nov., sp. nov., isolated from the deep-sea environment of the Puerto Rico trench. Appl Environ Microbiol 80(1):54–60. doi:10.1128/AEM.02288-13

Charlou JL, Donval JP, Fouquet Y et al (2002) Geochemistry of high H(2) and CH(4) vent fluids issuing from ultramafic rocks at the Rainbow hydrothermal field (36 degrees 14′N, MAR). Chem Geol 191:345–359. doi:10.1016/s0009-2541(02)00134-1

Charlou JL, Donval JP, Konn C et al (2010) High production of H-2 and CH4 and abiotic hydrocarbons in ultramafic-hosted hydrothermal systems on the Mid-Atlantic Ridge. Geochim Cosmochim Acta 74:A163–A163

Chong PL, Ayesa U, Daswani VP, Hur EC (2012) On physical properties of tetraether lipid membranes: effects of cyclopentane rings. Archaea 2012:138439. doi:10.1155/2012/138439

Collins MD, Hummer G, Quillin ML et al (2005) Cooperative water filling of a nonpolar protein cavity observed by high-pressure crystallography and simulation. Proc Natl Acad Sci USA 102:16668–16671. doi:10.1073/pnas.0508224102

Corliss JB, Ballard RD (1977) Oases of life in the cold abyss. Natl Geogr Mag 152:440–453

Crépeau V, Cambon Bonavita MA, Lesongeur F, Randrianalivelo H et al (2011) Diversity and function in microbial mats from the Lucky Strike hydrothermal vent field. FEBS Lett 76:524–540

Csonka LN, Hanson AD (1991) Prokaryotic osmoregulation—genetics and physiology. Annu Rev Microbiol 45:569–606

Cybulski LE, Albanesi D, Mansilla MC et al (2002) Mechanism of membrane fluidity optimization: isothermal control of the *Bacillus subtilis* acyl-lipid desaturase. Mol Microbiol 45:1379–1388

Da Costa MS, Santos H, Galinski EA (1998) An overview of the role and diversity of compatible solutes in Bacteria and Archaea. Adv Biochem Eng Biotechnol 61:117–153

Dannenmuller O, Arakawa K, Eguchi T et al (2000) Membrane properties of archaeal macrocyclic diether phospholipids. Chemistry (Easton) 6:645–654

Dawson KS, Freeman KH, Macalady JL (2012) Molecular characterization of core lipids from halophilic archaea grown under different salinity conditions. Org Geochem 48:1–8

Deming JW, Somers LK, Straube WL, Swartz DG, MacDonell MT (1988) Isolation of an obligately barophilic bacterium and description of a new genus, *Colwellia* gen. nov. Syst Appl Microbiol 10:152–160

De Long EF, Franks DG, Yayanos AA (1997) Evolutionary relationships of cultivated psychrophilic and barophilic deep-sea bacteria. Appl Env Microbiol 63:2105–2108

De Rosa M, Esposito E, Gambacorta A et al (1980a) Complex lipids of *Caldariella acidophila*, a thermoacidophile archaebacterium. Phytochemistry 19:821–826

De Rosa M, Esposito E, Gambacorta A et al (1980b) Effects of temperature on ether lipid composition of *Caldariella acidophila*. Phytochemistry 19:827–831

Dreyfus G, Guimaraes-Motta H, Silva J (1988) Effect of hydrostatic pressure on the mitochondrial ATP synthase. Biochemistry 27:6704–6710

Eder W, Ludwig W, Huber R (1999) Novel 16S rRNA gene sequences retrieved from highly saline brine sediments of kebrit deep, red sea. Arch Microbiol 172:213–218

Edmond JM, Vondamm KL, McDuff RE, Measures CI (1982) Chemistry of hot springs on the east pacific rise and their effluent dispersal. Nature 297:187–191. doi:10.1038/297187a0

Eloe EA, Malfatti F, Gutierrez J et al (2011) Isolation and characterization of a psychropiezophilic alphaproteobacterium. Appl Env Microbiol 77:8145–8153. doi:10.1128/AEM.05204-11

Empadinhas N, Marugg JD, Borges N et al (2001) Pathway for the synthesis of mannosylglycerate in the hyperthermophilic archaeon *Pyrococcus horikoshii*—biochemical and genetic characterization of key enzymes. J Biol Chem 276:43580–43588

Erauso G, Reysenbach AL, Godfroy A et al (1993) *Pyrococcus abyssi* sp-nov, a new hyperthermophilic archaeon isolated from a deep-sea hydrothermal vent. Arch Microbiol 160:338–349

Erijman L, Lorimer G, Weber G (1993) Reversible dissociation and conformational stability of dimeric ribulose bisphosphate carboxylase. Biochemistry 32:5187–5195

Ernst M, Freisleben HJ, Antonopoulos E et al (1998) Calorimetry of archaeal tetraether lipid—indication of a novel metastable thermotropic phase in the main phospholipid from *Thermoplasma acidophilum* cultured at 59 degrees C. Chem Phys Lipids 94:1–12

Euzéby J (2013) List of prokaryotic names with standing in nomenclature. http://www.bacterio.net/number.html. Accessed 26 Jul 2013

Flores GE, Wagner ID, Liu Y, Reysenbach A-L (2012) Distribution, abundance, and diversity patterns of the thermoacidophilic "deep-sea hydrothermal vent euryarchaeota 2". Front Microbiol 3:47. doi:10.3389/fmicb.2012.00047

Foguel D, Suarez MC, Ferrão-Gonzales AD et al (2003) Dissociation of amyloid fibrils of alpha-synuclein and transthyretin by pressure reveals their reversible nature and the formation of water-excluded cavities. Proc Natl Acad Sci USA 100:9831–9836. doi:10.1073/pnas.1734009100

Fourme R, Ascone I, Kahn R et al (2002) Opening the high-pressure domain beyond 2 kbar to protein and virus crystallography. Structure 10:1409–1414

Fourme R, Girard E, Kahn R et al (2006) High-pressure macromolecular crystallography (HPMX): status and prospects. Biochim Biophys Acta 1764:384–390. doi:10.1016/j.bbapap.2006.01.008

Franzmann PD, Springer N, Ludwig W et al (1992) A methanogenic archaeon from Ace Lake, Antarctica: *Methanococcoides burtonii* sp. nov. Syst Appl Microbiol 15:573–581

Galinski EA (1995) Osmoadaptation in bacteria. Adv Microb Physiol 37:272–328

Galinski EA, Truper HG (1994) Microbial behavior in salt-stressed ecosystems. FEMS Microbiol Rev 15:95–108

Gerasimchuk AL, Shatalov AA, Novikov AL et al (2010) The search for sulfate-reducing bacteria in mat samples from the lost city hydrothermal field by molecular cloning. Microbiology 79:96–105. doi:10.1134/S0026261710010133

Girard E, Kahn R, Mezouar M et al (2005) The first crystal structure of a macromolecular assembly under high pressure: CpMV at 330 MPa. Biophys J 88:3562–3571. doi:10.1529/biophysj.104.058636

Girard E, Marchal S, Perez J et al (2010) Structure-function perturbation and dissociation of tetrameric urate oxidase by high hydrostatic pressure. Biophys J 98:2365–2373. doi:10.1016/j.bpj.2010.01.058

Gliozzi A, Rolandi R, De Rosa M, Gambacorta A (1983) Monolayer black membranes from bipolar lipids of archaebacteria and their temperature-induced structural changes. J Membr Biol 75:45–56

Goncalves LG, Lamosa P, Huber R, Santos H (2008) Di-myo-inositol phosphate and novel UDP-sugars accumulate in the extreme hyperthermophile *Pyrolobus fumarii*. Extremophiles 12:383–389. doi:10.1007/s00792-008-0143-0

Gonthier I, Rager MN, Metzger P et al (2001) A di-O-dihydrogeranylgeranyl glycerol from *Thermococcus* S557, a novel ether lipid, and likely intermediate in the biosynthesis of diethers in Archaea. Tetrahedron Lett 42:2795–2797

Gorovits B, Raman C, Horowitz P (1995) High hydrostatic pressure induces the dissociation of cpn60 tetradecamers and reveals a plasticity of the monomers. J Biol Chem 270:2061–2066

Gross M, Auerbach G, Jaenicke R (1993) The catalytic activities of monomeric enzymes show complex pressure dependence. FEBS Lett 321:256–260

Hafenbradl D, Keller M, Thiericke R, Stetter KO (1993) A novel unsaturated archael ether core lipid from the hyperthermophile *Methanopyrus kandleri*. Syst Appl Microbiol 16:165–169

Hei DJ, Clark DS (1994) Pressure stabilization of proteins from extreme thermophiles. Appl Env Microbiol 60:932–939

Huber R, Kurr M, Jannasch K, Stetter K (1989) A novel group of abyssal methanogenic archaebacteria (*Methanopyrus*) growing at 110°C. Nature 342:833–834

Huber J, Butterfield D, Baross J (2003) Bacterial diversity in a subseafloor habitat following a deep-sea volcanic eruption. FEMS Microbiol Ecol 43:393–409

Jannasch HW, Mottl MJ (1985) Geomicrobiology of deep-sea hydrothermal vents. Science 229:717–725. doi:10.1126/science.229.4715.717

Jarrell HC, Zukotynski KA, Sprott GD (1998) Lateral diffusion of the total polar lipids from *Thermoplasma acidophilum* in multilamellar liposomes. Biochim Biophys Acta-Biomembr 1369:259–266

Johnson K, Beehler C, Sakamoto-Arnold C, Childress J (1986) In situ measurements of chemical distributions in a deep-sea hydrothermal vent field. Science 231:1139–1141

Jun X, Lupeng L, Minjuan X et al (2011) Complete genome sequence of the obligate piezophilic hyperthermophilic archaeon *Pyrococcus yayanosii* CH1. J Bacteriol 193:4297–4298. doi:10.1128/JB.05345-11

Kallmeyer J, Pockalny R, Adhikari RR et al (2012) Global distribution of microbial abundance and biomass in subseafloor sediment. Proc Natl Acad Sci USA 109:16213–16216. doi:10.1073/pnas.1203849109

Kasai R, Kitajima Y, Martin CE et al (1976) Molecular control of membrane properties during temperature acclimatation—membrane fluidity regulation of fatty acid desaturase action. Biochemistry 15:5228–5233

Kawano H, Nakasone K, Matsumoto M et al (2004) Differential pressure resistance in the activity of RNA polymerase isolated from *Shewanella violacea* and *Escherichia coli*. Extremophiles 8:367–375. doi:10.1007/s00792-004-0397-0

Khelaifia S, Fardeau M-L, Pradel N et al (2011) *Desulfovibrio piezophilus* sp. nov., a piezophilic, sulfate-reducing bacterium isolated from wood falls in the Mediterranean Sea. Int J Syst Evol Microbiol 61:2706–2711. doi:10.1099/ijs.0.028670-0

Lai D, Springstead JR, Monbouquette HG (2008) Effect of growth temperature on ether lipid biochemistry in Archaeoglobus fulgidus. Extremophiles 12:271–278

Lamosa P, Martins LO, Da Costa MS, Santos H (1998) Effects of temperature, salinity, and medium composition on compatible solute accumulation by *Thermococcus* spp. Appl Env Microbiol 64:3591–3598

Lamosa P, Turner DL, Ventura R et al (2003) Protein stabilization by compatible solutes. Effect of diglycerol phosphate on the dynamics of *Desulfovibrio gigas* rubredoxin studied by NMR. Eur J Biochem 270:4606–4614

Lamosa P, Rodrigues MV, Gonçalves LG et al (2013) Organic solutes in the deepest phylogenetic branches of the Bacteria: identification of α(1-6)glucosyl-α(1-2)glucosylglycerate in *Persephonella marina*. Extremophiles 17:137–146. doi:10.1007/s00792-012-0500-x

Lauro F, Chastain R, Ferriera S et al (2013) Draft Genome Sequence of the Deep-Sea Bacterium *Shewanella benthica* Strain KT99. Genome Announc 1:e00210–e00213. doi:10.1128/genomeA.00210-13.Copyright

Lee AG (2003) Lipid–protein interactions in biological membranes: a structural perspective. Biochim Biophys Acta 1612:1–40

Lee AG (2004) How lipids affect the activities of integral membrane proteins. Biochim Biophys Acta-Biomembr 1666:62–87

Li H, Yamada H, Akasaka K (1998) Effect of pressure on individual hydrogen bonds in proteins. Basic Pancreatic trypsin inhibitor. Biochemistry 37:1167–1173

Lu B, Li Q, Liu W, Ruan K (1997) Effects of hydrostatic pressure on the activity of rat ribosome and cell-free translation system. Biochem Mol Biol Int 43:499–506

Lucas S, Han J, Lapidus A et al (2012) Complete genome sequence of the thermophilic, piezophilic, heterotrophic bacterium *Marinitoga piezophila* KA3. J Bacteriol 194:5974–5975. doi:10.1128/JB.01430-12

Luke KA, Higgins CL, Wittung-Stafshede P (2007) Thermodynamic stability and folding of proteins from hyperthermophilic organisms. FEBS J 274:4023–4033. doi:10.1111/j.1742-4658.2007.05955.x

Macdonald AG, Martinac B (2005) Effect of high hydrostatic pressure on the bacterial mechanosensitive channel MscS. Eur Biophys J 34:434–441. doi:10.1007/s00249-005-0478-8

Madern D, Ebel C, Zaccai G (2000) Halophilic adaptation of enzymes. Extremophiles 4:91–98

Mangelsdorf K, Zink KG, Birrien JL, Toffin L (2005) A quantitative assessment of pressure dependent adaptive changes in the membrane lipids of piezosensitive deep sub-seafloor bacterium. Org Geochem 36:1459–1479

Marassio G, Prangé T, David HN et al (2011) Pressure-response analysis of anesthetic gases xenon and nitrous oxide on urate oxidase: a crystallographic study. FASEB J 25:2266–2275. doi:10.1096/fj.11-183046

Marchi M, Akasaka K (2001) Simulation of hydrated BPTI at high pressure: changes in hydrogen bonding and its relation with NMR experiments. J Phys Chem B 105:711–714

Marr AG, Ingraham JL (1962) Effect of temperature on the composition of fatty acids in *Escherichia coli*. J Bacteriol 84:1260–1267

Marteinsson VT, Birrien JL, Reysenbach AL et al (1999) *Thermococcus barophilus* sp. nov., a new barophilic and hyperthermophilic archaeon isolated under high hydrostatic pressure from a deep-sea hydrothermal vent. Int J Syst Bacteriol 49(Pt 2):351–359

Martin DD, Bartlett DH, Roberts MF (2002) Solute accumulation in the deep-sea bacterium *Photobacterium profundum*. Extremophiles 6:507–514. doi:10.1007/s00792-002-0288-1

Martins LO, Santos H (1995) Accumulation of mannosylglycerate and di-myo-inosytol-phosphate by *Pyrococcus furiosus* in response to salinity and temperature. Appl Environ Microbiol 61:3299–3303

Martins LO, Carreto LS, DaCosta MS, Santos H (1996) New compatible solutes related to di-myo-inositol-phosphate in members of the order Thermotogales. J Bacteriol 178:5644–5651

Martins LO, Huber R, Huber H et al (1997) Organic solutes in hyperthermophilic Archaea. Appl Environ Microbiol 63:896–902

Masson P, Bec N, Froment M-T et al (2004) Rate-determining step of butyrylcholinesterase-catalyzed hydrolysis of benzoylcholine and benzoylthiocholine. Volumetric study of wild-type and D70G mutant behavior. Eur J Biochem 271:1980–1990. doi:10.1111/j.1432-1033.2004.04110.x

Mathai JC, Sprott GD, Zeidel ML (2001) Molecular mechanisms of water and solute transport across archaebacterial lipid membranes. J Biol Chem 276:27266–27271

Matsuno Y, Sugai A, Higashibata H et al (2009) Effect of growth temperature and growth phase on the lipid composition of the archaeal membrane from *Thermococcus kodakaraensis*. Biosci Biotechnol Biochem 73:104–108. doi:10.1271/bbb.80520

McElhaney RN (1984a) The structure and function of the Acholeplasma laidlawii plasma membrane. Biochim Biophys Acta 779:1–42

McElhaney RN (1984b) The relationship between membrane fluidity and phase state and the ability of bacteria and mycoplasma to grow and survive at various temperatures. In: Manson LA (ed) Kates M. Membr. fluidity. Plenum Press, New York, pp 249–276

Mombelli E, Shehi E, Fusi P, Tortora P (2002) Exploring hyperthermophilic proteins under pressure: theoretical aspects and experimental findings. Biochim Biophys Acta 25:392–396

Morita T (2003) Structure-based analysis of high pressure adaptation of alpha-actin. J Biol Chem 278:28060–28066. doi:10.1074/jbc.M302328200

Nagae T, Kato C, Watanabe N (2012) Structural analysis of 3-isopropylmalate dehydrogenase from the obligate piezophile *Shewanella benthica* DB21MT-2 and the nonpiezophile Shewanella oneidensis MR-1. Acta Crystallogr, Sect F: Struct Biol Cryst Commun 68:265–268. doi:10.1107/S1744309112001443

Neves C, da Costa MS, Santos H (2005) Compatible solutes of the hyperthermophile *Palaeococcus ferrophilus*: osmoadaptation and thermoadaptation in the order thermococcales. Appl Env Microbiol 71:8091–8098. doi:10.1128/AEM.71.12.8091-8098.2005

Nichols DS, Miller MR, Davies NW et al (2004) Cold adaptation in the antarctic archaeon *Methanococcoides burtonii* involves membrane lipid unsaturation. J Bacteriol 186:8508–8515

Nishiguchi Y, Miwa T, Abe F (2008) Pressure-adaptive differences in lactate dehydrogenases of three hagfishes: *Eptatretus burgeri*, *Paramyxine atami* and *Eptatretus okinoseanus*. Extremophiles 12:477–480. doi:10.1007/s00792-008-0140-3

Nisius L, Grzesiek S (2012) Key stabilizing elements of protein structure identified through pressure and temperature perturbation of its hydrogen bond network. Nat Chem 4:711–717. doi:10.1038/nchem.1396

Nogi Y, Masui N, Kato C (1998) *Photobacterium profundum* sp. nov., a new, moderately barophilic bacterial species isolated from a deep-sea sediment. Extremophiles 2(1):1–7

Nogi Y, Kato C, Horikoshi K (2002) *Psychromonas kaikoae* sp. nov., a novel from the deepest piezophilic bacterium cold-seep sediments in the Japan Trench. Int J Syst Evol Microbiol 52(5):1527–1532

Nogi Y, Hosoya S, Kato C, Horikoshi K (2004) *Colwellia piezophila* sp. nov., a novel piezophilic species from deep-sea sediments of the Japan Trench. Int J Syst Evol Microbiol 54(5):1627–1631

Nunes OC, Manaia CM, Dacosta MS, Santos H (1995) Compatible solutes in the thermophilic bacteria *Rhodothermus marinus* and *Thermus thermophilus*. Appl Environ Microbiol 61:2351–2357

Nunoura T, Oida H, Nakaseama M et al (2010) Archaeal diversity and distribution along thermal and geochemical gradients in hydrothermal sediments at the Yonaguni Knoll IV hydrothermal field in the Southern Okinawa trough. Appl Env Microbiol 76:1198–1211. doi:10.1128/AEM.00924-09

Occhipinti E, Bec N, Gambirasio B et al (2006) Pressure and temperature as tools for investigating the role of individual non-covalent interactions in enzymatic reactions *Sulfolobus solfataricus* carboxypeptidase as a model enzyme. Biochim Biophys Acta 1764:563–572. doi:10.1016/j.bbapap.2005.12.007

Oger PM, Cario A (2013) Adaptation of the membrane in Archaea. Biophys Chem. doi:10.1016/j.bpc.2013.06.020

Oger PM, Jebbar M (2010) The many ways of coping with pressure. Res Microbiol 161:799–809. doi:10.1016/j.resmic.2010.09.017

Ohmae E, Tatsuta M, Abe F et al (2008) Effects of pressure on enzyme function of *Escherichia coli* dihydrofolate reductase. Biochim Biophys Acta 1784:1115–1121. doi:10.1016/j.bbapap.2008.04.005

Ohmae E, Murakami C, Tate S et al (2012) Pressure dependence of activity and stability of dihydrofolate reductases of the deep-sea bacterium *Moritella profunda* and *Escherichia coli*. Biochim Biophys Acta 1824:511–519. doi:10.1016/j.bbapap.2012.01.001

Orcutt BN, Bach W, Becker K et al (2011) Colonization of subsurface microbial observatories deployed in young ocean crust. ISME J 5:692–703. doi:10.1038/ismej.2010.157

Page A, Juniper S, Olagnon M et al (2004) Microbial diversity associated with a *Paralvinella sulfincola* tube and the adjacent substratum on an active deep-sea vent chimney. Geobiology 2:225–238

Paull CK, Hecker B, Commeau R et al (1984) Biological communities at the Florida escarpment resemble hydrothermal vent taxa. Science 226:965–967

Refaee M, Tezuka T, Akasaka K, Williamson MP (2003) Pressure-dependent Changes in the solution structure of hen egg-white lysozyme. J Mol Biol 327:857–865. doi:10.1016/S0022-2836(03)00209-2

Reysenbach AL, Longnecker K, Kirshtein J (2000) Novel bacterial and archaeal lineages from an in situ growth chamber deployed at a Mid-Atlantic Ridge hydrothermal vent. Appl Env Microbiol 66:3798–3806

Reysenbach AL, Gotz D, Banta A et al (2002) Expanding the distribution of the *Aquificales* to the deep-sea vents on Mid-Atlantic ridge and central Indian Ocean ridge. Cah Biol Mar 43:425–428

Robb FT, Clark DS (1999) Adaptation of proteins from hyperthermophiles to high pressure and high temperature. J Mol Microbiol Biotechnol 1:101–105

Roche J, Caro JA, Norberto DR et al (2012) Cavities determine the pressure unfolding of proteins. Proc Natl Acad Sci USA 109:6945–6950. doi:10.1073/pnas.1200915109

Rosenbaum E, Gabel F, Durá MA et al (2012) Effects of hydrostatic pressure on the quaternary structure and enzymatic activity of a

large peptidase complex from *Pyrococcus horikoshii*. Arch Biochem Biophys 517:104–110. doi:10.1016/j.abb.2011.07.017
Rouget J-B, Aksel T, Roche J et al (2011) Size and Sequence and the Volume Change of Protein Folding. J Am Chem Soc 133:6020–6027. doi:10.1021/ja200228w.Size
Roussel EG, Konn C, Charlou JL et al (2011) Comparison of microbial communities associated with three Atlantic ultramafic hydrothermal systems. FEMS Microbiol Ecol 77:647–665. doi:10.1111/j.1574-6941.2011.01161.x
Russell NJ, Nichols DS (1999) Polyunsaturated fatty acids in marine bacteria–a dogma rewritten. Microbiology 145(Pt 4):767–779
Santos H, da Costa MS (2001) Organic solutes from thermophiles and hyperthermophiles. Methods Enzym 334:302–315
Santos H, da Costa MS (2002) Compatible solutes of organisms that live in hot saline environments. Env Microbiol 4:501–509
Sawle L, Ghosh K (2011) How do thermophilic proteins and proteomes withstand high temperature? Biophys J 101:217–227
Schrenk M, Kelley D, Bolton S, Baross J (2004) Low archaeal diversity linked to subseafloor geochemical processes at the Lost City Hydrothermal Field, Mid-Atlantic Ridge. Env Microbiol 6:1086–1095
Shimada H, Nemoto N, Shida Y et al (2008) Effects of pH and temperature on the composition of polar lipids in *Thermoplasma acidophilum* HO-62. J Bacteriol 190:5404–5411
Sinensky M (1971) Temperature control of phospholipid biosynthesis in Escherichia coli. J Bacteriol 106:449–455
Sinensky M (1974) Homeoviscous adaptation—Homerostatic process that regulates viscosity of membrane lipids in *Escherichia coli*. Proc Natl Acad Sci USA 71:522–525
Sprott GD, Meloche M, Richards JC (1991) Proportions of diether, macrocyclic diether, and tetraether lipids in *Methanococcus jannaschii* grown at different temperatures. J Bacteriol 173:3907–3910
Stewart LC, Kates M, Ekiel IH, Smith ICP (1990) Molecular order and dynnamics of diphytanylglycerol phospholipids - A 2H NMR and 31P NMR study. Chem Phys Lipids 54:115–129
Sun MM, Tolliday N, Vetriani C et al (1999) Pressure-induced thermostabilization of glutamate dehydrogenase from the hyperthermophile *Pyrococcus furiosus*. Protein Sci 8:1056–1063. doi:10.1110/ps.8.5.1056
Suzuki Y, Sasaki T, Suzuki M et al (2005) Novel chemoautotrophic endosymbiosis between a member of the *Epsilonproteobacteria* and the hydrothermal-vent gastropod *Alvinicoccha aff. hessleri* (Gastropoda: Provannidae) from the Indian Ocean. Appl Env Microbiol 71:5440–5450. doi:10.1128/AEM.71.9.5440-5450.2005
Sylvan JB, Pyenson BC, Rouxel O et al (2012) Time-series analysis of two hydrothermal plumes at 9°50′N East Pacific Rise reveals distinct, heterogeneous bacterial populations. Geobiology 10:178–192. doi:10.1111/j.1472-4669.2011.00315.x
Takai K, Horikoshi K (1999) Genetic diversity of archaea in deep-sea hydrothermal vent environments. Genetics 152:1285–1297
Takai K, Horikoshi K (2000) *Thermosipho japonicus* sp. nov., an extremely thermophilic bacterium isolated from a deep-sea hydrothermal vent in Japan. Extremophiles 4:9–17
Takai K, Nakamura K (2011) Archaeal diversity and community development in deep-sea hydrothermal vents. Curr Opin Microbiol 14:282–291. doi:10.1016/j.mib.2011.04.013
Takai K, Sugai A, Itoh T, Horikoshi K (2000) *Palaeococcus ferrophilus* gen. nov., sp. nov., a barophilic, hyperthermophilic archaeon from a deep-sea hydrothermal vent chimney. Int J Syst Evol Microbiol 50(Pt 2):489–500
Takai K, Komatsu T, Inagaki F, Horikoshi K (2001) Distribution of archaea in a black smoker chimney structure. Appl Env Microbiol 67:3618–3629. doi:10.1128/AEM.67.8.3618-3629.2001
Takai K, Nakamura K, Toki T et al (2008) Cell proliferation at 122 degrees C and isotopically heavy CH4 production by a hyperthermophilic methanogen under high-pressure cultivation. Proc Natl Acad Sci USA 105:10949–10954. doi:10.1073/pnas.0712334105
Takai K, Miyazaki M, Hirayama H et al (2009) Isolation and physiological characterization of two novel, piezophilic, thermophilic chemolithoautotrophs from a deep-sea hydrothermal vent chimney. Environ Microbiol 11:1983–1997. doi:10.1111/j.1462-2920.2009.01921.x
Teske A, Hinrichs K, Edgcomb V et al (2002) Microbial diversity of hydrothermal sediments in the Guaymas basin: evidence for anaerobic methanotrophic communities †. Appl Environ Microbiol 68:1994–2007. doi: 10.1128/AEM.68.4.1994
Uda I, Sugai A, Itoh YH, Itoh T (2001) Variation in molecular species of polar lipids from Thermoplasma acidophilum depends on growth temperature. Lipids 36:103–105
Uda Y, Sugai A, Itoh YH, Itoh T (2004) Variation in molecular species of core lipids from the order Thermoplasmales strains depends on the growth temperature. J Oleo Sci 53:399–404
Van de Vossenberg J, Driessen AJM, da Costa MS, Konings WN (1999) Homeostasis of the membrane proton permeability in Bacillus subtilis grown at different temperatures. Biochim Biophys Acta-Biomembr 1419:97–104
Van Dover CL, German CR, Speer KG et al (2002) Evolution and biogeography of deep-sea vent and seep invertebrates. Science 295:1253–1257. doi:10.1126/science.1067361
Vannier P, Marteinsson VT, Fridjonsson OH et al (2011) Complete genome sequence of the hyperthermophilic, piezophilic, heterotrophic, and carboxydotrophic archaeon *Thermococcus barophilus* MP. J Bacteriol 193:1481–1482. doi:10.1128/JB.01490-10
Ventosa A, Nieto JJ, Oren A (1998) Biology of moderately halophilic aerobic bacteria. Microbiol Mol Biol Rev 62:504–544
Von Damm KL (2000) Chemistry of hydrothermal vent fluids from 9 degrees-10 degrees N, East Pacific Rise: "Time zero", the immediate posteruptive period. J Geophys Res Earth 105:11203–11222. doi:10.1029/1999jb900414
Vreeland RH (1987) Mechanisms of halotolerance in microorganisms. Crit Rev Microbiol 14:311–356. doi:10.3109/10408418709104443
Whitman WB, Coleman DC, Wiebe WJ (1998) Prokaryotes: the unseen majority. Proc Natl Acad Sci USA 95:6578–6583
Winter R (2002) Effect of lipid chain length, temperature, pressure and composition on the lateral organisation and phase behavior of lipid bilayer/gramicidin mixtures. Biophys J 82:153A–153A
Winter R, Jeworrek C (2009) Effect of pressure on membranes. Soft Matter 5:3157–3173
Xu Y, Nogi Y, Kato C, Liang Z, Rüger HJ, De Kegel D, Glansdorff N (2003) *Psychromonas profunda* sp. nov., a psychropiezophilic bacterium from deep Atlantic sediments. Int J Syst Evol Microbiol 53(2):527–532
Yamauchi K (1993) Doi K, Yoshida Y, Kinoshita M Archaebacterial lipids: highly proton-impermeable membranes from 1,2-diphytanyl-sn-glycero-3- phosphocholine. BiochimBiophysActa 1146:178–182
Yano Y, Nakayama A, Ishihara K, Saito H (1998) Adaptive changes in membrane lipids of barophilic bacteria in response to changes in growth pressure. Appl Env Microbiol 64:479–485
Yayanos AA, Dietz AS, Vanboxtel R (1979) Isolation of a deep-sea barophilic bacterium and some of its growth characteristics. Science 205:808–810
Yayanos AA, Dietz AS, Van Boxtel R (1981) Obligately barophilic bacterium from the Mariana trench. Proc Natl Acad Sci USA 78:5212–5215

Zeng X, Birrien JL, Fouquet Y et al (2009) *Pyrococcus* CH1, an obligate piezophilic hyperthermophile: extending the upper pressure-temperature limits for life. ISME J 3:873–876. doi:10.1038/ismej.2009.21

Zeng X, Zhang X, Jiang L et al (2012) *Palaeococcus pacificus* sp. nov., a novel archaeon from a deep-sea hydrothermal sediment. Int J Syst Evol Microbiol 63:2155–2159. doi:10.1099/ijs.0.044487-0

Zeng X, Zhang X, Jiang L, Alain A, Jebbar M, Shao Z (2013) *Palaeococcus pacificus* sp. nov., a novel archaeon from a deep-sea hydrothermal sediment. IJSEM 63:2155–2159

Zeng X, Zhang Z, Li X et al (2015) *Anoxybacter fermentans* gen. nov., sp. nov., a piezophilic, thermophilic, anaerobic, fermentative bacterium isolated from a deep-sea hydrothermal vent. Int J Syst Evol Microbiol 65:710–715

Zhao W, Zeng X, Xiao X (2015) *Thermococcus eurythermalis* sp. nov., a conditional piezophilic, hyperthermophilic archaeon with a wide temperature range for growth, isolated from an oil-immersed chimney in the Guaymas Basin. Int J Syst Evol Microbiol 65:30–35

Zobell CE, Johnson FH (1949) The Influence of Hydrostatic Pressure on the Growth and Viability of Terrestrial and Marine Bacteria. J Bacteriol 57:179–189