THE DEEP-SEA BRINES OF THE MEDITERRANEAN SEA :











Michail Yakimov and MAMBA Consortium





MedSea deep brine pools



Origin of MedSea deep brine pools. 1. Messinian salinity crisis







Seawater

Upper interface Lower interface

Highly concentrated H₂S (2-20 mM)

<u>Hypersaline (150-330‰)</u>

<u>High pressure (>330 atm)</u>

Origin of MedSea deep brine pools. Why they are so different?

	MedSea Water	Evaporitic brine	Tyro Basin	Urania Basin	Bannock Basin	Discovery Basin
Na	0.49	5.10	5.30	3.52	4.20	0.08
K	0.02	0.12	0.02	0.13	0.13	0.09
Mg	0.06	0.59	0.07	0.32	0.64	5.09
Ca	0.01	0.007	0.04	0.04	0.016	0.001
Cl	0.57	5.41	5.35	3.75	5.38	10.15
SO4	0.03	0.21	0.06	0.11	0.14	0.11
Salinity, g/L	38.5	380	411	283	340	480
		-				

Origin of MedSea deep brine pools. 2. Different types of evaporites



Origin of MedSea deep brine pools. 3. Evaporation of seawater

1000m	DOmI Brine produced by 200X evaporation of seawater and precipitation of Anhydrite CaSO4 Halite NaCl Halite NaCl Kieserite MgSO4'H2O Carnallite KMgCl3'H2O Brine produced by equilibrating of seawater with evaporite minerals Bischofite MgCl2'6H2O Halite NaCl				
486 11 55 566 29 μmol 30	5103 115 7 591 5786 205 300	166 71 1 5410 10100 173 3350	99 112 3 5480 10900 122	Na+ K+ Ca++ Mg++ CI- SO4 Li+	

Origin of MedSea deep brine pools. 2. Different types of brines

	Early stage		evaporites	Late stage	evaporites	Extreme evaporite conditions	
	MedSea Water	Evaporitic brine	Tyro Basin	Urania Basin	Bannock Basin	Discovery Basin	
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Brine of Discovery Basin has the highest salinity ever found in marine environment and one of the most salted ecosystem on our planet.

The brine lake, found in December 1993, has the surface area of about 7.5 km² and a volume of nearly 2x10⁸ m³

5	Discovery	Dead Sea	Lake Assal	Don Juan Pond
	Basin	(depth	(depth	"foot deep"
		50m)	20m)	
Na	0.08	1.84	3.76	0.51
Mg	5.09	1.95	1.14	0.11
Cl	10.15	6.82	4.87	12.19
SO4	0.11	0.01	0.02	<0.001
Salinity, g/L	487	315	398	540
Density	1.33	1.21	1.24	1.35

All MedSea deep anoxic basins possess vast unexplored microbial diversity

The Enigma of Prokaryotic Life in Deep Hypersaline Anoxic Basins

Paul W. J. J. van der Wielen,^{1*}† Henk Bolhuis,¹ Sara Borin,² Daniele Daffonchio,² Cesare Corselli,³ Laura Giuliano,⁴ Giuseppe D'Auria,⁴ Gert J. de Lange,⁵ Andreas Huebner,⁵ Sotirios P. Varnavas,⁶ John Thomson,⁷ Christian Tamburini,⁸ Danielle Marty,⁸ Terry J. McGenity,⁹ Kenneth N. Timmis,^{9,10} BioDeep Scientific Party

Van der Wielen et al., *Science*, 2005, 121-3 Daffonchio et al., *Nature*, 2006, 203-7 Borin *et al.*, *PNAS*, 2009, 9151-6

Stratified prokaryote network in the oxic-anoxic transition of a deep-sea halocline

Daniele Daffonchio¹, Sara Borin¹, Tullio Brusa¹, Lorenzo Brusetti¹, Paul W. J. J. van der Wielen², Henk Bolhuis², Michail M. Yakimov³, Giuseppe D'Auria³, Laura Giuliano³, Danielle Marty⁴, Christian Tamburini⁴, Terry J. McGenity⁵, John E. Hallsworth⁵, Andrea M. Sass⁵, Kenneth N. Timmis^{5,6}, Anastasios Tselepides⁷, Gert J. de Lange⁸, Andreas Hübner⁸, John Thomson⁹, Soterios P. Varnavas¹⁰, Francesco Gasparoni¹¹, Hans W. Gerber¹², Elisa Malinverno¹³, Cesare Corselli¹³ & Biodeep Scientific Party^{*}

Sulfur cycling and methanogenesis primarily drive microbial colonization of the highly sulfidic Urania deep hypersaline basin

Sara Borin^a, Lorenzo Brusetti^a, Francesca Mapelli^a, Giuseppe D'Auria^b, Tullio Brusa^a, Massimo Marzorati^a, Aurora Rizzi^a, Michail Yakimov^c, Danielle Marty^d, Gert J. De Lange^e, Paul Van der Wielen[†], Henk Bolhuis^a, Terry J. McGenity^h, Paraskevi N. Polymenakouⁱ, Elisa Malinvernoⁱ, Laura Giuliano^c, Cesare Corselli^j, and Daniele Daffonchio^{a.1}



Functioning of DHAL ecosystems: understanding the biogeochemical pathways that sustain DHALs microbial metabolic network systems

All MedSea deep anoxic basins possess highly stratified microbial communities

Environmental Microbiology (2009) 11(2), 360-381

Microbial eukaryotes in the hypersaline anoxic L'Atalante deep-sea basin

Eva Alexander, 'Alexandra Stock,' Hans-Werner Breiner,' Anke Behnke,' John Bunge,' Michail M., Yakimov' and Thorsten Stoeck'' 'University of Kaiserslautern, School of Biology, Erwin-Schredingere-Strasse 14, De7773, Kaiserslautern, Germany. 'Department of Statistical Science, Corneil University, Ithaca, NY 14853, USA. 'Institute for Coastal Marine Environment, IAMC-CNR, Spinata S. Rahene, Bé, 98122 Messina, Itaj. ing in the evolution of an exceptional and distinctive assemblage of profists. The deep hypersaline anoxic basins in the Mediterranean See provide an ideal platform to test for this hypothesis and are promising targets for the discovery of undescribed profists with unknown physiological capabilities.

Introduction

In the past few decades, hypersaline environments have attracted increasing notice in biological sciences (Oren,

How and when these communities were formed?

ORIGINAL ARTICLE

Primary producing prokaryotic communities of brine, interface and seawater above the halocline of deep anoxic lake L'Atalante, Eastern Mediterranean Sea

Michail M Yakimov¹, Violetta La Cono¹, Renata Denaro¹, Giuseppe D'Auria¹, Franco Decembrini¹, Kenneth N Timmis², Peter N Golyshin^{2,5,5} and Laura Giuliano^{1,4,5}. 'Institute for coastal Marine Environment (MMC), CNR, Mexison, Italy: 'Environmental Microbiology Laboratory, HZI-Helmholtz Centre for Infection Research, Braunschweig, Germany: ³School of Biological Sciences, University of Bangor, Wales, UK and 'Mediterranean Science Commission, CIESM, Monte-Carlo, Monaco

The ISME Journal (2007) 1, 743-755 o 2007 International Society for Microbial Ecology All rights manued 1751-7362/07 \$30.00

Surface sediments of Discovery Basin have been in contact with bischofite brine for only 2,000 years.

Within this short period the Discovery Basin must have been filled at an average of almost 10,000 m³ yr⁻¹ (Nature 1997, 387:31-32).

chaotropicity defines the window

Highest limit for cultivation in pure Mg⁺⁺ is approx. 1M (Waino et al., Int. J. Syst. Evol. Microbiol., 2000) Highest limit of life in solution of bischofite (MgCl₂) is 2.23M (Hallsworth, Yakimov et al. Environm Microbiol, 2007) In case of deep-sea anoxic hypersaline thalasso- and athalassohaline ecosystems we are facing the existence of peculiar microbial world whose genomic information is yet hidden for us up to now and we cannot go deeper into analysis of functional genes simply because there are no cultivated analogues known.

To metagenome or not to metagenome?

Two major metabolic features, fully functional sulfur metabolism and chemolithoautotrophy, detected by amplification and analysis of functional genes, were plotted against metagenome sequencing data to compare the outcomes of these two analytical platforms: SULPHUR METABOLISM

OXIDATION

--sulfide:quinone reductase SQR (oxidation of sulfide to elemental S0)

--fully functional SOX system (sulfite-, thiosulfate-, sulfur- and hydrogen sulfide reduction)

REDUCTION

- --polysulfide (oxido)reductases
- --dissimilatory sulfite reductases
- ASSIMILATION (in less extend)
- --Few sulphate transporters and no ATP-sulfurylases other than of Delta-PB

CO₂ FIXATION 3-HP/4-HB pathway driven by marine Thaumarchaea MGI (acetyl-CoA carboxylase, 4-hydroxybutyryl-CoA dehydratase)

Reductive pentose phosphate CBB cycle → Gamma-PB (RubisCO, phosphoribulokinases)

Reductive citric acid cycle → Epsilon-PB (2-oxoglutarate synthase, ATP-citrate lyase)

Reductive acetyl–CoA pathway → Euryarchaea (MSBL1?, HC1?, Methanohalophilus?) 5,10-methylenetetrahydromethanopterin reductase, CO dehydrogenase/acetyl-CoA synthase complex

DHALs Heterotrophy

Methylotrophic methanogenesis using methylamines and/or methyl thiols driven by ?*Methanohalophilus*-related archaea

Sulphate-, sulfur-reduction and sulphur disproportionation by Delta-, Epsilon-Proteobacteria, *Halanaerobium*-related bacteria and possibly by members of KB1 candidate division.

IAMC, Istituto per l'Ambiente Marino Costiero, CNR, Messina, Italy

IC CSIC, Institute of Catalysis, Madrid, Spain

Industrial partners: Spain France Germany

Marine metagenomics for new biotechnological applications

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