Epsilonproteobacteria dominate bacterial diversity at a natural tar seep

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Supplementary Information

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Table S1: Composition of the underground water sampled at Puy-de-la-Poix.

Element	abundance (mg/l)
C organic	16 mg/l
Na	57 mg/l
Ca	103 mg/l
HCO_3	432 mg/l
NO_3	3 mg/l
SO_4	104 mg/l
Fe	56 µg/l
Al	7 µg/l
Co	5 µg/l
Cu	3 µg/l
H_2S	0.25 mg/l
P	< 1 mg/l
Ni	< 1 µg/l
O_2	4.5 mg/l

Geological and geochemical characteristics of the tar source Puy-de-la-Poix

The Puy-de-la-Poix is a natural, flowing source of heavy oil in Limagne, 1 km west of Clermont-Ferrand (France, 45.7822 N 3.14642 E, 340m, Figure 1). Evidence for petroleum in this region, and at the Puy-de-la-Poix in particular, is dated back to the neolithic period. The Limagne plain takes its name from the latin "*lacus magnus*", e.g. the big lake. The lake was created during the Alps uplift which fractured the granitic bedrock. As a consequence the Limagne region subsided ca. 2000m, which created a vast lacustrine environment which accumulated more than 1500m of organic-rich sediments during the Oligocene period. The sediments of the Limagne are composed of successions of lacustrine limestones, clay-stones, sandstones and black shales. Several attempts at exploiting this resource were made until the late 50's, which proved unsuccessful essentially due to the lack of appropriate reservoir rock formations and trap structures [1]. The Limagne petroleum is heavy, partially oxidized, sulfur-rich [2]. The bitumen is slowly migrating to the surface along numerous faults created by the Alps upraising, and the magmatic chimney of the numerous volcanoes of the region of the Puy, e.g. volcano (Figure 1). The name Puy-de-la-Poix indicates that the tar (Poix) flows from an ancient volcano (Puy). The water composition at Puy-de-la-Poix (Table 1) is typical of the so-called sodium bicarbonate province of the Massif central [3, 4]. Previous isotopic studies of the waters in this geological province clearly indicated that the water originates from the surface and get saturated with carbonates and salts while heated underground before it flows back to the surface [5]. The source at the Puy-

de-la-Poix is however slightly less warm (25°C) and carbonated (549mg/l) than the thermal sources of the area such as Royat, which may indicate that the superficial waters did not migrate to the same depth in the source at Puy-de-la-Poix, or that some mixing with surface meteoritic water occurs during its migration to the surface.

Table S2: Taxononomical allocation of the cloned sequences from the bitumen (B), underground water (U) and surface water (SW) samples. Analyses performed with the classifier fonction of the RDP database (http://rdp.cme.msu.edu/), and confirmed using the arb software (-)

Phylum	Class	Order	Family	Genus	B	UW	SW
Verrucomicrobia	*Verrucomicrobiae*	*Verrucomicrobiales*	*Verrucomicrobiaceae*	*Luteolibacter*			1
				Prosthecobacter			2
Bacteroidetes			*Bacteroidetes incertae sedis*	*Prolixibacter*			1
	Sphingobacteria	*Sphingobacteriales*	*Chitinophagaceae*	unclassified			1
	Flavobacteria	*Flavobacteriales*	*Flavobacteriaceae*	*Flavobacterium*			1
				Aequorivita			1
				Lutibacter			1
Tenericutes	*Mollicutes*	*Acholeplasmatales*	*Acholeplasmataceae*	*Acholeplasma*	1	1	
Firmicutes	*Clostridia*	*Clostridiales*	*Lachnospiraceae*	unclassified	1		
	unclassified					1	
Spirochaetes	*Spirochaetes*	*Spirochaetales*	*Spirochaetaceae*	*Spirochaeta*		1	
Proteobacteria	*Alphaproteobacteria*	*Rhizobiales*	*Rhodobiaceae*	*Parvibaculum*	1		
			Hyphomicrobiaceae	*Filomicrobium*			1
		Caulobacterales	*Hyphomonadaceae*	*Hyphomonas*	1		1
		Rhodobacterales	*Rhodobacteraceae*	*Rhodobacter*			3
				Roseovarius			1
				Seohaeicola		1	
				unclassified	1		
		unclassified			1		1
	Deltaproteobacteria	*Desulfobacterales*	*Desulfobulbaceae*			1	
	Betaproteobacteria	*Rhodocyclales*	*Rhodocyclaceae*				2
		Burkholderiales	*Burkholderiales incertae sedis*				1
			Comamonadaceae	*Variovorax*			1
				Polaromonas			1
				Hydrogenophaga			5
				Simplicispira		1	
				unclassified			2

Gammaproteobacteria	*Pseudomonadales*	*Pseudomonadaceae*	*Pseudomonas*	1		
		Moraxellaceae	*Perlucidibaca*			1
	Alteromonadales	*Alteromonadaceae*	*Marinobacter*	1		1
	Methylococcales	*Methylococcaceae*	*Methylomonas*			1
	Legionellales	*Legionellaceae*	*Legionella*			1
			unclassified			4
	Xanthomonadales	*Xanthomonadaceae*	*Dokdonella*			1
			unclassified			1
		Sinobacteraceae	*Nevskia*		1	1
	Acidithiobacillales	*Acidithiobacillaceae*	*Acidithiobacillus*		1	
	Oceanospirillales	*Oceanospirillaceae*	*Nitrincola*	2		
		Halomonadaceae	*Halomonas*		1	
	Thiotrichales	*Piscirickettsiaceae*	*Thiomicrospira*		3	1
	unclassified					2
Epsilonproteobacteria	*Campylobacterales*	*Campylobacteraceae*	*Sulfurospirillum*			1
			Arcobacter	6	3	
		Helicobacteraceae	*Sulfuricurvum*		1	
			Sulfurovum	6	3	6
			Sulfuromonas	33	31	2
			unclassified	1		
unclassified				1		
unclassified						2

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