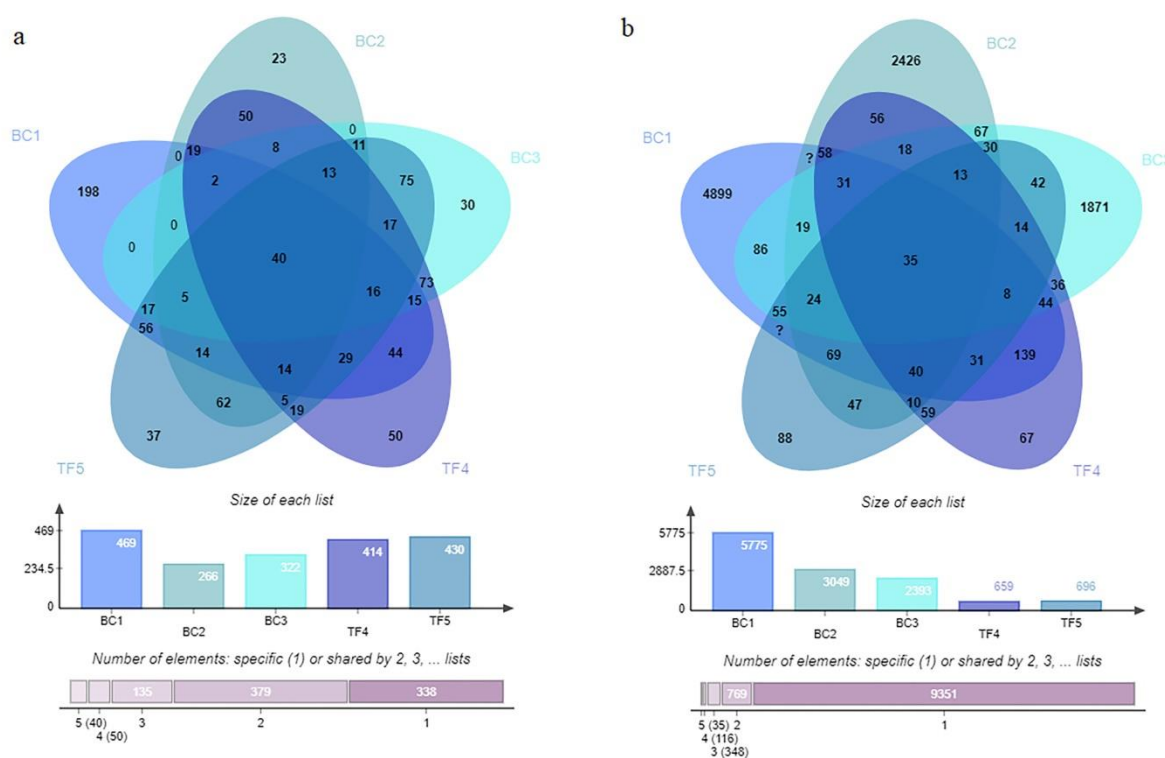
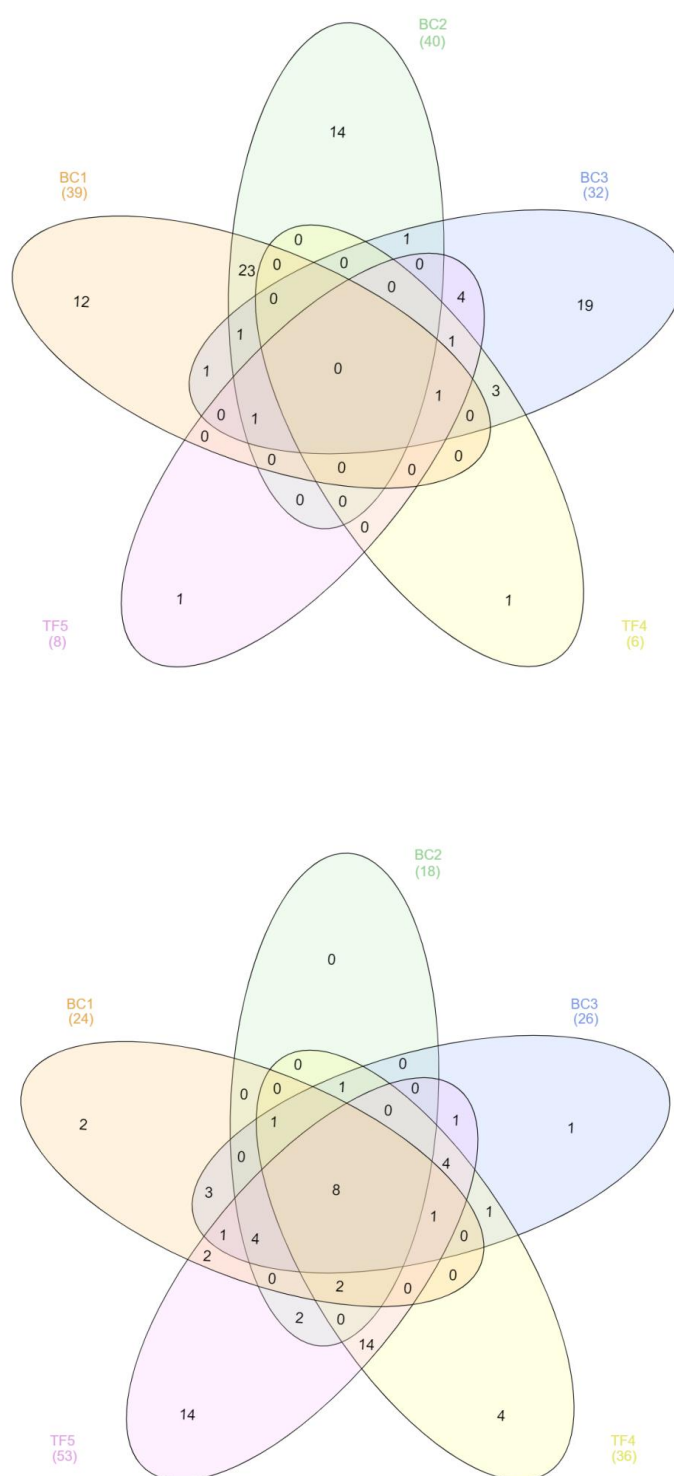


# Supplementary Materials:

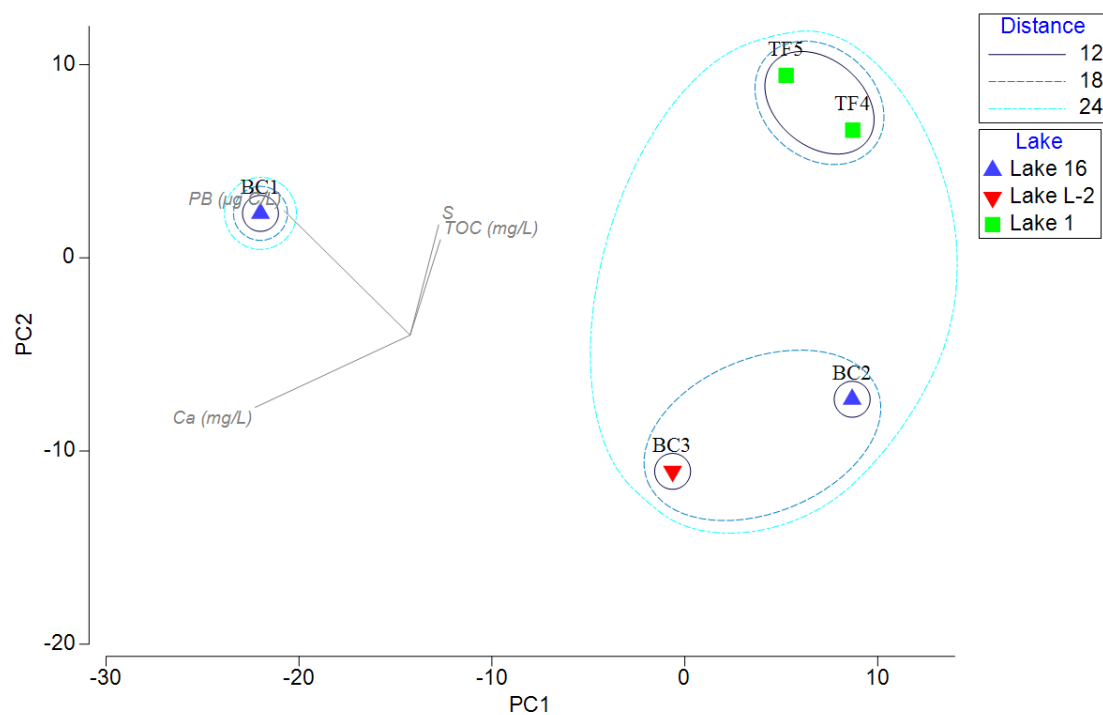
**Figure S1:** Venn diagram for (a) Bacteria and (b) Archaea; **Figure S2:** Venn diagram showing the sharing level of bacterial (a) and archaeal (b) genera between BC and TF brine samples; **Figure S3:** Principal Component Analysis computed on the previous dataset including physicochemical data related to BC and TF brine samples; **Figure S4:** Circos diagrams showing the connection among brine samples and metabolic pathways. The ribbon size represents cell value correspondent to brine/metabolic pathway segment pair, while the stacked bars show ribbon contribution for each segment; **Table S1:** Microbiological and physico-chemical data previously reported for brines; **Table S2:** The total number of sequence reads, good quality reads, observed number of OTUs, Shannon diversity, Evenness and Chao 1 indices per sample of the bacterial and archaeal 16S rRNA gene data sets. Data are from Papale et al. [4] and Lo Giudice et al. [12]; **Table S3:** Relative abundance (% on the total predicted pathways) of predicted metabolic pathways identified in prokaryotic populations produced by the PICRUST analysis. The icons are referred to percentages within the same sub-group (in bold).



**Figure S1. (a-b).** Venn diagram for (a) Bacteria and (b) Archaea.

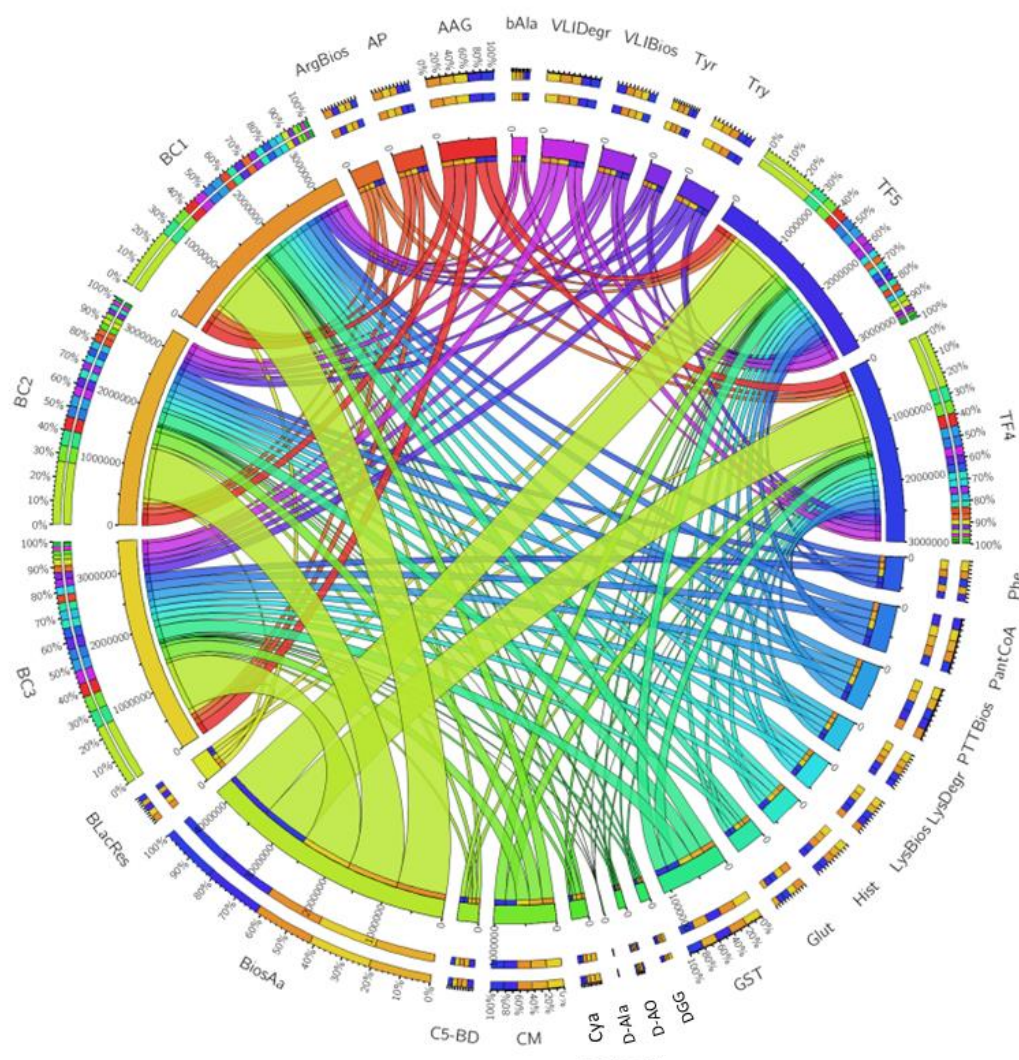


**Figure S2. (a-b):** Venn diagram showing the sharing level of bacterial (a) and archaeal (b) genera between BC and TF brine samples.



**Figure S3.** Principal Component Analysis computed on the previous dataset including physicochemical data related to BC and TF brine samples.

a)

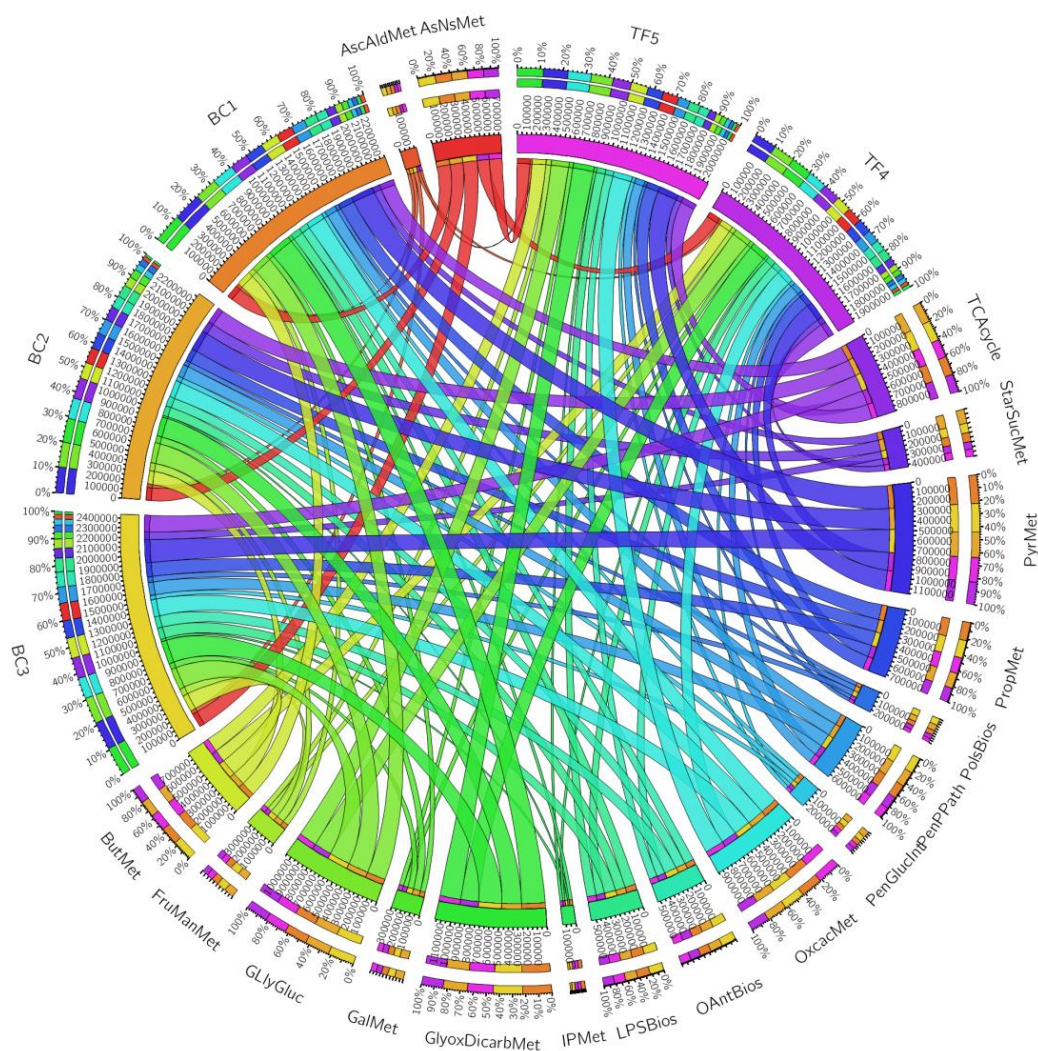


### Amino Acid Metabolism

AAG	Alanine_aspartate_glutamate metabolism	LysBios	Lysine_biosynthesis
AP	Arginine_Proline_metabolism	LysDegr	Lysine_degradation
BiosAa	Biosynthesis_aminoacids	Try	Tryptophan_metabolism
Cya	Cyanoamino_acid_metabolism	VLIBios	Valine_leucine_isoleucine_biosynthesis
CM	Cysteine_methionine_metabolism	VLI Degr	Valine_leucine_isoleucine_degradation
D-Ala	D-Alanine_metabolism	DGG	D-Glutamine_D-glutamate_metabolism
D-AO	D-Arginine_D-ornithine_metabolism	GST	Glycine_serine_threonine_metabolism
ArgBios	Arginine_biosynthesis	Hist	Histidine_metabolism
bAla	beta-Alanine_metabolism	Tyr	Tyrosine_metabolism
BLacRes	beta-Lactam_resistance	C5-BD	C5-Branched_dibasic_metabolism
Phe	Phenylalanine_metabolism	Glut	Glutathione_metabolism
PTTBios	Phenylalanine_tyrosine_tryptophan_biosynthesis	PantCoA	Pantothenate_CoA_biosynthesis



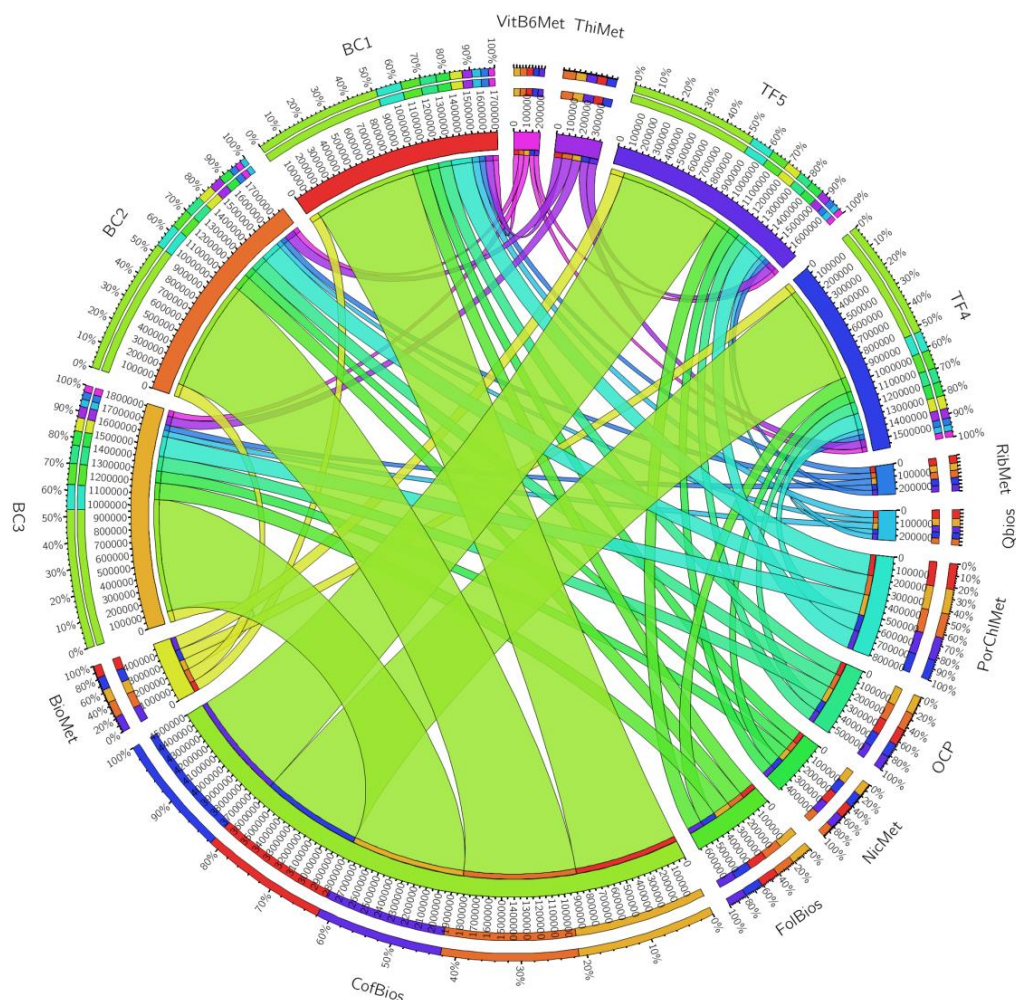
b)



### Carbohydrate Metabolism

OxcacMet:	2-Oxocarboxylic acid metabolism	PenGlucInt:	Pentose and glucuronate interconversions
AsNsMet:	Amino sugar and nucleotide sugar metabolism	PenPPath:	Pentose phosphate pathway
ButMet:	Butanoate metabolism	PolSBios:	Polyketide sugar unit biosynthesis
TCA cycle:	Citrate cycle (TCA cycle)	PyrMet:	Pyruvate metabolism
FruManMet:	Fructose and mannose metabolism	StarSucMet:	Starch and sucrose metabolism
GalMet:	Galactose metabolism	AscAldMet:	Ascorbate and aldarate metabolism
GLyGluc:	Glycolysis / Gluconeogenesis	PropMet:	Propanoate metabolism
GlyoxDicarbMet:	Glyoxylate and dicarboxylate metabolism	IPMet:	Inositol phosphate metabolism
LPSBios:	Lipopolysaccharide biosynthesis	OAntBios:	O-Antigen nucleotide sugar biosynthesis

c)

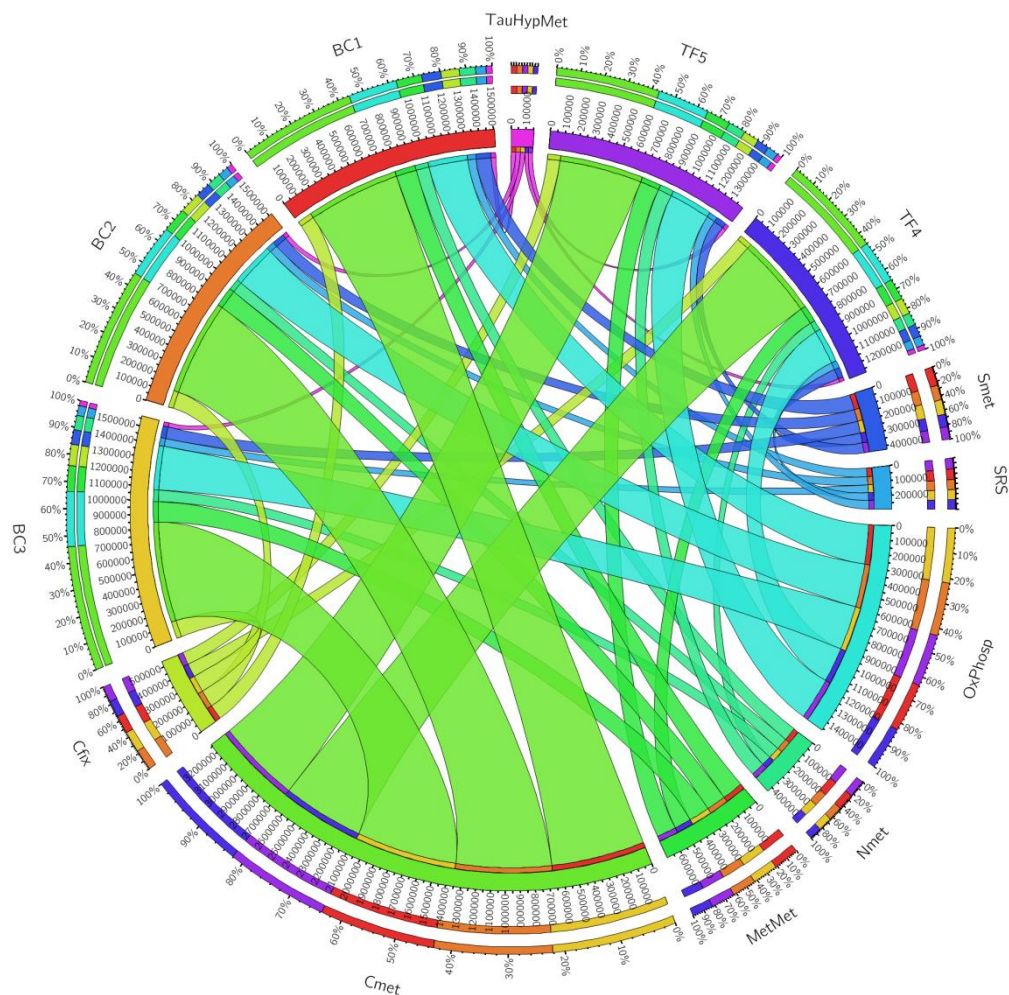


### Metabolism of cofactors and vitamins

FolBios:	Folate biosynthesis	ThiMet:	Thiamine metabolism
NicMet:	Nicotinate and nicotinamide metabolism	Qbios:	Ubiquinone and other terpenoid-quinone biosynthesis
OCP:	One carbon pool by folate	VitB6Met:	Vitamin B6 metabolism
PorChlMet:	Porphyrin and chlorophyll metabolism	BioMet:	Biotin metabolism
RibMet:	Riboflavin metabolism	CofBios:	Biosynthesis of cofactors



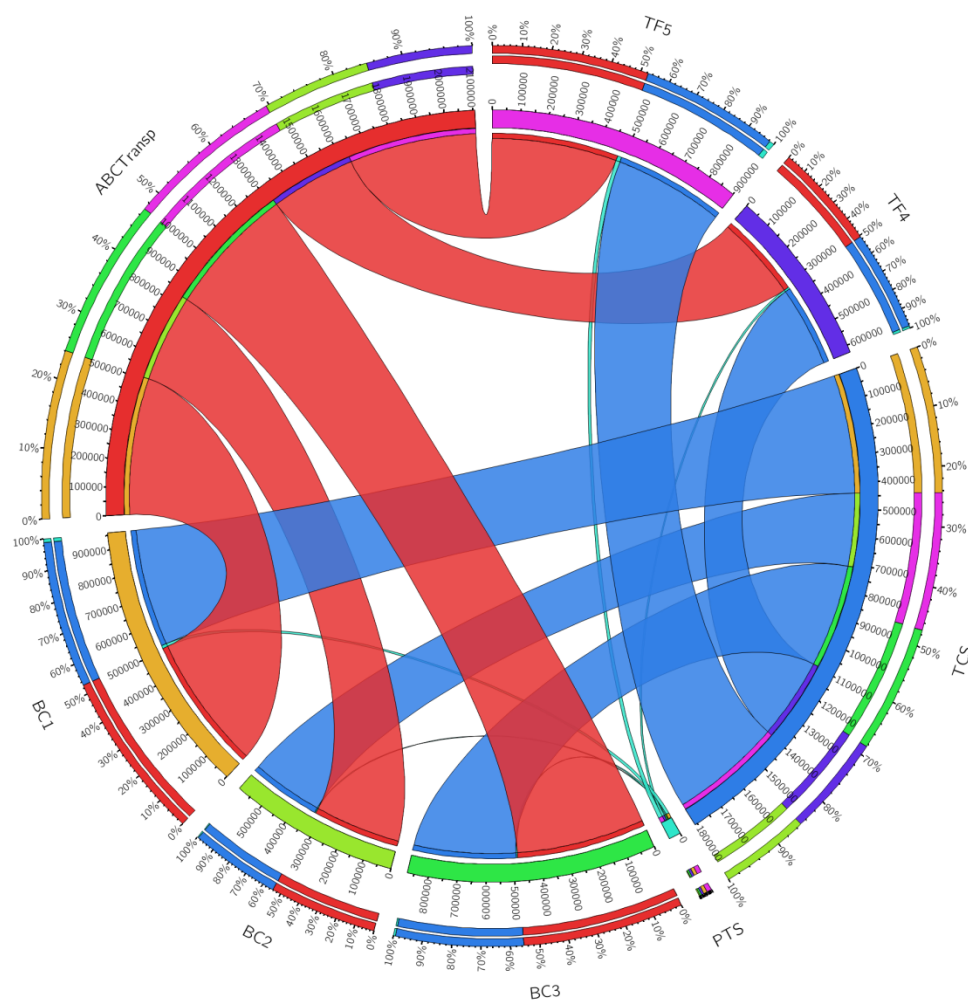
d)



### Energy Metabolism

Cfix:	Carbon fixation in photosynthetic organisms	Smet:	Sulfur metabolism
Cmet:	Carbon metabolism	OxPhosp:	Oxidative phosphorylation
MetMet:	Methane metabolism	SRS:	Sulfur relay system
Nmet:	Nitrogen metabolism	TauHypMet:	Taurine and hypotaurine metabolism

e)



### Transporters

ABCTransp	ABC transporters
PTS	Phosphotransferase system (PTS)
TCS	Two-component system



ArComp:	Degradation of aromatic compounds	Xyl:	Xylene degradation
BA:	Benzoate degradation	Tol:	Toluene degradation
ABA:	Aminobenzoate degradation	Ger:	Geraniol degradation
ClAlk:	Chloroalkane and chloroalkene degradation	Naph:	Naphthalene degradation
RNA:	RNA degradation	Diox:	Dioxin degradation
ClCyclBenz:	Chlorocyclohexane and chlorobenzene degradation	Capr:	Caprolactam degradation
Sty:	Styrene degradation	NT:	Nitrotoluene degradation
FBA:	Fluorobenzoate degradation	PAH:	Polycyclic aromatic hydrocarbon degradation
LimPin:	Limonene and pinene degradation	Gly:	Other glycan degradation

**Figure S4. (a-f):** Circos diagrams showing the connection among brine samples and metabolic pathways. The ribbon size represents cell value correspondent to brine/metabolic pathway segment pair, while the stacked bars show ribbon contribution for each segment.

**Table S1.** Microbiological and physico-chemical data previously reported for brines.

		TF4	TF5	BC1	BC2	BC3
<i>Trace elements</i> (mg L <sup>-1</sup> )	Al	17	>5	nd	nd	nd
	Ba	1.4	0.8	nd	nd	nd
	Ca	0.8	0.8	362.9	2.7	148.1
	Cd	0.3	0.1	0.0	0.1	0.1
	Cl	nd	nd	3410.9	14.1	476.4
	Co	0.7	0.7	1.1	0.0	0.0
	Cr	0.4	0.2	nd	nd	nd
	Cu	7.3	>5	nd	nd	nd
	Fe	29.7	5.7	nd	nd	nd
	I	1.2	2	nd	nd	nd
	K	0.5	0.9	70.7	0.3	18.7
	Li	1.17	1.77	nd	nd	nd
	Mg	2.0	2.8	167.4	0.8	21.9
	Mn	1.58	2.05	nd	nd	nd
	Na	nd	nd	2263.7	9.3	546.4
	Ni	1.7	1.8	5.3	0.2	0.1
	Sr	5.7	7.8	nd	nd	nd
	Ti	263.0	118.0	nd	nd	nd
	U	7.0	6.0	nd	nd	nd
	V	1.4	2.9	nd	nd	nd
	Zn	17.7	> 5	nd	nd	nd
<i>Anions</i>	NO <sub>3</sub> (mg L <sup>-1</sup> )	>0.004	>0.004	nd	nd	nd
	PO <sub>4</sub> (g L <sup>-1</sup> )	>0.002	>0.002	nd	nd	nd
	SO <sub>4</sub> (mg L <sup>-1</sup> )	10.6	10.9	1695.8	5.6	1043.8
<i>Prokaryotic community</i>						
<i>AC_Diversity Indices</i>	Shannon	6045.0	6863.0	5.9	5.7	5.3
	Eveness	0.4	0.2	0.7	0.7	0.7
	Chao I	3738.0	3892.0	9559.2	10419.8	7762.2
<i>BC_Diversity Indices</i>	Shannon	4804.0	4971.0	2.7	2.2	2.3
	Eveness	0.3	0.1	0.5	0.4	0.4
	Chao I	1377.0	2148.0	1079.6	590.3	941.1
<i>AC (%)</i>	AAG	1.6	1.8	2.0	1.1	2.2
	CREN	7.8	7.8	15.3	12.4	14.0
	EURY	55.8	57.1	81.4	85.7	82.4
	KORAR	0.1	0.0	0.3	0.8	1.1
	MHV Group I	0.2	0.1	1.0	0.0	0.3
<i>BC (%)</i>	UNC	34.6	33.1	0.0	0.0	0.0
	ACID	<0.1	<0.1	<0.1	<0.1	<0.1
	ACT	17.2	13.7	2.7	5.8	4.0
	ALF	1.9	4.3	3.5	0.1	<0.1

	AQU	0.0	<0.1	0.2	0.1	0.0
	BAC	79.7	69.0	28.6	9.4	52.7
	BET	2.9	4.6	14.3	4.5	17.7
	CHLAM	<0.1	<0.1	<0.1	<0.1	<0.1
	Chlorobi	<0.1	<0.1	<0.1	<0.1	<0.1
	Chloroflexi	<0.1	0.1	0.4	0.0	0.0
	CYA	0.4	0.6	0.2	0.0	<0.1
	DEF	<0.1	<0.1	<0.1	<0.1	<0.1
	DEL	1.6	22.0	0.0	0.0	0.0
	EPS	1.4	5.4	0.8	0.0	0.0
	FIR	1.5	0.6	0.1	<0.1	0.0
	GAM	33.4	29.6	3.9	0.2	0.4
	PAR	0.0	0.0	1.6	<0.1	0.3
	PLAN	1.6	0.1	0.0	0.0	0.0
	PROT UNC	0.0	0.0	5.0	0.1	0.3
	SPIR	2.1	0.8	0.0	0.0	0.0
	OTHERS	7.3	8.6	14.9	9.6	8.2
<i>Microbial Activities</i>	AP (nmol L <sup>-1</sup> h <sup>-1</sup> )	1.1	1.0	3.9	0.6	1.1
	β-GLU (nmol L <sup>-1</sup> h <sup>-1</sup> )	2.5	0.1	0.1	1.2	1.6
	LAP (nmol L <sup>-1</sup> h <sup>-1</sup> )	2.0	2.0	2.0	1.3	0.5
<i>Physiological Profiles (PP)</i>						
<i>Complex carbon sources</i>	Tween 40	6.8	2.0	11.4	3.9	3.1
	Tween 80	7.4	3.7	10.6	3.9	7.9
	α_cyclodextrin	11.8	1.0	16.6	1.7	1.2
<i>Carbohydrates</i>	Glycogen	0.0	1.2	1.5	4.2	2.7
	D_cellobiose	3.4	3.2	15.0	3.2	1.6
	α_D_lactose	2.7	2.3	0.0	0.0	0.0
	β_methyl_D_glu coside	0.6	4.0	0.0	0.0	0.0
	D_xylose	10.7	8.2	0.2	5.7	3.2
	i_erythritol	4.4	5.2	0.2	3.7	2.2
	D_mannitol	0.2	2.4	0.2	4.6	20.3
	N_acetyl_D_glu cosamine	0.1	3.2	3.0	3.1	3.2
	Glucose_1_phos phste	2.0	1.2	0.5	4.2	2.1
<i>Phosphate-carbon</i>	D,l_α_glycerol phosphate	2.5	4.3	0.0	0.0	0.0
<i>Carboxylic acid</i>	Pyruvic acid methyl ester	4.1	2.0	7.6	3.9	13.6
	D_glucosamic acid	1.4	1.1	0.4	6.0	4.5
	D_galactonic acid	1.0	4.8	0.5	5.2	2.0
	2_hydroxy	3.3	1.5	0.0	3.5	3.7

Amino acids	benzoic acid					
	4_hydroxy benzoic acid	6.8	2.5	12.0	4.6	4.7
	$\gamma$ _hydroxy butyric acid	2.4	2.5	0.3	4.1	1.2
	Itaconic acid	5.3	8.2	0.3	5.2	3.1
	$\alpha$ _ketobutyric acid	2.5	3.4	0.3	3.0	3.1
	D_malic Acid	2.3	6.9	0.0	0.0	0.0
	L_arginine	1.9	1.6	0.0	0.0	0.0
	L_asparagine	0.1	1.3	10.0	3.3	1.4
	l_phenylalanine	5.6	1.1	1.8	5.4	2.4
	L_serine	0.4	1.2	0.3	5.4	1.8
	L_threonine	0.8	1.2	0.0	2.7	2.5
	Glycyl_L_glutamic acid	1.2	1.4	7.1	3.2	2.3
	Phenylethyl_amine	0.0	1.0	0.1	5.5	4.4
	Putrescine	7.8	4.5	0.0	0.6	1.7
Microbial abundance	BVC TSA1 (CFU mL <sup>-1</sup> )	3.1	7.2	15.5	0.0	0.0
	BVC TSA50 (CFU mL <sup>-1</sup> )	4.7	5.5	4.3	0.5	0.9
	BVC TSA100 (CFU mL <sup>-1</sup> )	3.3	5.5	5.3	0.1	2.3
	BVC DSMZ97 (CFU mL <sup>-1</sup> )	0.0	0.0	0.0	0.0	0.1
	BVC R2A (CFU mL <sup>-1</sup> )	0.1	0.1	0.0	0.0	0.0
	CTC (%)	18.0	30.2	8.5	1.0	7.1
	DEAD cells (%)	33.0	59.2	55.5	94.7	90.5
	LIVE cells (%)	67.0	40.8	44.5	5.3	9.5
	PA (cells L <sup>-1</sup> x 10 <sup>9</sup> )	5.0	8.1	6.1	0.4	0.4
	PB ( $\mu$ g C L <sup>-1</sup> )	67.0	242.0	567.5	32.5	31.8
Morphometric features	Cocci (%)	44	28	31	23	0
	Coccobacilli (%)	26	33	0	50	26
	Curved rods (%)	0	5	4	0	0
	Filamentous bacteria (%)	0	0	27	0	0
	Rods (%)	30	33	35	27	74
	Vibrios (%)	0	0	2	0	0
	VOL ( $\mu$ m <sup>3</sup> )	0.04	0.1	0.2	0.2	0.1
Environmental parameters	pH	7.2	6.7	6.5	7.9	7.0
	S (psu)	90	75	6.3	0.04	1.6
	TIC (mg L <sup>-1</sup> )	628.3	674.8	nd	nd	nd
	TOC (mg L <sup>-1</sup> )	82.0	102.5	11.5	6.6	6.4



**Table S2.** The total number of sequence reads, good quality reads, observed number of OTUs, Shannon diversity, Evenness and Chao 1 indices *per* sample of the bacterial and archaeal 16S rRNA gene data sets. Data are from Papale et al. [4] and Lo Giudice et al. [12].

		Good quality reads				Diversity indices			
	Sample	Total reads (n.)	n.	%	Chimeras (%)	OTUs (n.)	Shannon H	Evenness	Chao 1
<i>Bacteria</i>	TF4	31863	18322	57.5024	2.9	414	4.804	0.347	1377.31
	TF5	32823	18394	56.0400	0	430	4.971	0.146	2148.849
<i>Archaea</i>	TF4	36555	13425	36.7255	0	659	6.045	0.397	3738.437
	TF5	29464	11713	39.7536	0.1	696	6.863	0.185	3892.03
<i>Bacteria</i>	BC1	37280	11396	30.5687	0.1	469	2.753	0.469	1079.625
	BC2	33932	17780	52.3989	0	266	2.178	0.390	590.294
	BC3	30005	14961	49.8617	0	322	2.333	0.404	941.138
<i>Archaea</i>	BC1	40415	14161	35.0390	3.1	5775	5.864	0.723	9559.173
	BC2	36550	13152	35.9836	8	3049	5.745	0.711	10419.855
	BC3	29649	10018	33.7887	7.2	2393	5.341	0.680	7762.220



**Table S3.** Relative abundance (% on the total predicted pathways) of predicted metabolic pathways identified in prokaryotic populations produced by the PICRUSt analysis. *The icons are referred to percentages within the same subgroup (in bold).*

Total Pathways	BC1	BC2	BC3	TF4	TF5
Aminoacyl-tRNA biosynthesis	0.89	0.93	0.95	1.04	0.98
Bacterial chemotaxis	0.52	0.20	0.30	0.48	0.77
Bacterial secretion system	0.86	0.59	0.66	0.76	0.92
Base excision repair	0.37	0.44	0.39	0.40	0.35
Biosynthesis of secondary metabolites	9.07	9.64	9.16	9.48	8.99
Biosynthesis of various secondary metabolites	0.08	0.12	0.11	0.10	0.10
Cationic antimicrobial peptide (CAMP) resistance	0.36	0.33	0.33	0.28	0.33
DNA replication	0.67	0.83	0.75	0.71	0.63
Flagellar assembly	0.87	0.27	0.50	0.76	0.94
Homologous recombination	0.88	1.01	0.92	0.92	0.81
Metabolic pathways	19.11	19.17	19.04	18.86	18.92
Microbial metabolism in diverse environments	5.41	5.39	5.57	5.32	5.12
Mismatch repair	0.69	0.82	0.75	0.74	0.67
Non-homologous end-joining	0.02	0.02	0.03	0.03	0.01
Peptidoglycan biosynthesis	0.68	0.74	0.71	0.77	0.70
Phenazine biosynthesis	0.07	0.09	0.08	0.09	0.08
Phosphonate and phosphinate metabolism	0.01	0.01	0.00	0.00	0.01
Plant-pathogen interaction	0.21	0.15	0.18	0.20	0.23
Protein export	0.55	0.60	0.57	0.64	0.63
Quorum sensing	1.60	1.19	1.53	1.32	1.99
Ribosome	1.92	2.30	2.15	2.34	2.07
RNA polymerase	0.14	0.14	0.14	0.16	0.15
Selenocompound metabolism	0.32	0.38	0.36	0.35	0.32
Sesquiterpenoid and triterpenoid biosynthesis	0.00	0.00	0.00	0.02	0.00
Vancomycin resistance	0.23	0.27	0.26	0.25	0.23
<b>Amino acid metabolism</b>					
Alanine, aspartate and glutamate metabolism	0.89	0.91	0.82	0.94	0.92
Arginine and proline metabolism	0.59	0.48	0.49	0.47	0.44
Biosynthesis of amino acids	3.91	4.16	3.81	4.11	4.06
Cyanoamino acid metabolism	0.25	0.40	0.26	0.22	0.17
Cysteine and methionine metabolism	0.92	1.05	0.98	0.94	0.93
D-Alanine metabolism	0.11	0.11	0.12	0.11	0.13
D-Arginine and D-ornithine metabolism	0.01	0.00	0.01	0.01	0.02
Arginine biosynthesis	0.54	0.41	0.41	0.44	0.48
beta-Alanine metabolism	0.24	0.26	0.26	0.24	0.21
beta-Lactam resistance	0.44	0.37	0.38	0.35	0.40
Phenylalanine metabolism	0.51	0.50	0.61	0.57	0.46
Phenylalanine, tyrosine and tryptophan biosynthesis	0.67	0.72	0.70	0.77	0.71
Lysine biosynthesis	0.47	0.50	0.49	0.54	0.50
Lysine degradation	0.47	0.49	0.53	0.53	0.43
Tryptophan metabolism	0.57	0.61	0.67	0.60	0.45
Valine, leucine and isoleucine biosynthesis	0.52	0.52	0.45	0.54	0.64
Valine, leucine and isoleucine degradation	0.75	0.64	0.80	0.68	0.64
D-Glutamine and D-glutamate metabolism	0.16	0.15	0.16	0.17	0.15
Glycine, serine and threonine metabolism	1.08	1.02	1.03	1.09	1.06
Histidine metabolism	0.52	0.55	0.53	0.55	0.47
Tyrosine metabolism	0.36	0.34	0.41	0.34	0.29
C5-Branched dibasic acid metabolism	0.32	0.35	0.27	0.32	0.40
Glutathione metabolism	0.58	0.33	0.49	0.47	0.44
Pantothenate and CoA biosynthesis	0.68	0.73	0.68	0.74	0.79

**Table S3.** Continues.

Energy Metabolism										
Carbon fixation in photosynthetic organisms		0.49		0.58		0.53		0.49		0.44
Carbon metabolism		3.08		3.25		3.14		3.06		2.93
Methane metabolism		0.69		0.67		0.64		0.64		0.68
Nitrogen metabolism		0.44		0.40		0.36		0.38		0.50
Sulfur metabolism		0.50		0.41		0.37		0.38		0.35
Oxidative phosphorylation		1.30		1.46		1.38		1.31		1.47
Sulfur relay system		0.28		0.28		0.25		0.29		0.31
Taurine and hypotaurine metabolism		0.16		0.16		0.13		0.14		0.15
Metabolism of cofactors and vitamins										
Folate biosynthesis		0.60		0.63		0.63		0.68		0.63
Nicotinate and nicotinamide metabolism		0.44		0.40		0.43		0.50		0.46
One carbon pool by folate		0.52		0.59		0.55		0.52		0.48
Porphyrin and chlorophyll metabolism		0.81		0.80		0.75		0.73		0.78
Riboflavin metabolism		0.24		0.23		0.21		0.25		0.22
Thiamine metabolism		0.31		0.41		0.33		0.35		0.36
Ubiquinone and other terpenoid-quinone biosynthesis		0.28		0.19		0.23		0.22		0.22
Vitamin B6 metabolism		0.20		0.21		0.21		0.21		0.19
Biotin metabolism		0.39		0.52		0.40		0.48		0.55
Biosynthesis of cofactors		4.04		4.49		4.16		4.38		4.30
Metabolism of terpenoids and polyketides										
Carotenoid biosynthesis		0.01		0.00		0.01		0.01		0.01
Terpenoid backbone biosynthesis		0.39		0.25		0.39		0.41		0.43
Nonribosomal peptide structures		0.02		0.04		0.02		0.01		0.03
Transporters										
ABC transporters		2.33		1.54		2.10		1.66		2.31
Phosphotransferase system (PTS)		0.05		0.01		0.02		0.04		0.07
Two-component system		2.03		1.28		1.67		1.58		2.02
Antibiotics										
Acarbose and validamycin biosynthesis		0.11		0.16		0.15		0.12		0.09
Carbapenem biosynthesis		0.08		0.07		0.06		0.07		0.08
Monobactam biosynthesis		0.26		0.29		0.27		0.30		0.27
Novobiocin biosynthesis		0.10		0.10		0.11		0.10		0.09
Staurosporine biosynthesis		0.01		0.00		0.00		0.00		0.00
Streptomycin biosynthesis		0.29		0.30		0.35		0.32		0.25