

*Supplementary Material*

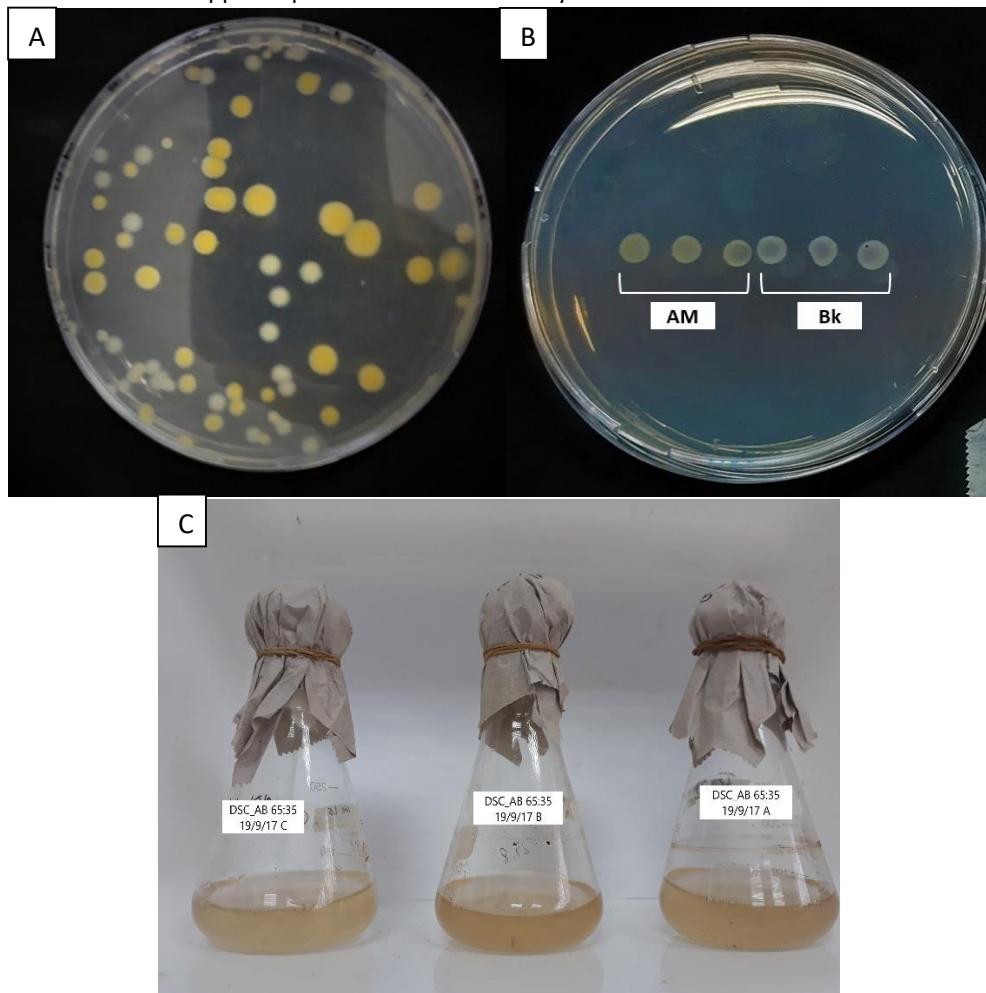
**Metaproteomic and gene expression analysis of interspecies interactions in a PAH-degrading synthetic microbial consortium constructed with the key microbes of a natural consortium**

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**Figure S1:** (A) R2A plate showing morphologies of both strains present in DSC\_AB culture, *Sphingobium* sp. AM (yellow) and *Burkholderia* sp. Bk (white) (B) Growth interference assay between AM and Bk in R2A. (C) DSC\_AB culture in MML supplemented with 200 ppm of phenanthrene after 7 days of incubation



**Table S1:** Target genes and primers sets (Khara et al., 2014) used to amplify genes of the upper and lower PAH degradation pathway of AM strain.

Gene	Primer set	Sequence (5'-3')	Annealing temperature	Reference
Sphingobium 16S rRNA	16S rRNA_RT_F	GTAAGCGGGCGATTTAAGTC	62	Khara et al, 2014
	16S rRNA_RT_R	ACAATCGTCCAGTGAGCC		Khara et al, 2014
Catechol 2,3-dioxygenase (xylE)	xylE_RT_F	CGTGGATATAGCCAGTTCG	60	Khara et al, 2014
	xylE_RT_R	TACCGTCTCGGGATAGC		Khara et al, 2014
(2Fe-2S)-binding protein (ahdB)	ahdB_RT_F	GATGTCCGCAATTATGTCG	55	Khara et al, 2014
	ahdB_RT_R	CTTTACGTTGACGCCGAAG		Khara et al, 2014
2,3-dihydroxybiphenyl 1,2-dioxygenase (ahdC)	ahdC_RT_F	ATGACCTGGCCTACATGG	55	Khara et al, 2014
	ahdC_RT_R	GATCTCGGTCGGATTGCC		Khara et al, 2014
Benzoate 1,2-dioxygenase subunit alpha (ahdA1d)	ahdA1d_RT_F	AGCATTGCGTGGAGCG	58	Khara et al, 2014
	ahdA1d_RT_R	ATAGCCCAGAACCTTGACC		Khara et al, 2014
Benzoate 1,2-dioxygenase subunit aa (ahdA1c)	ahdA1c_RT_F	CTGAGACAGTTACGCACCG	58	Khara et al, 2014
	ahdA1c_RT_R	AAGCTTCCAATCCCGC		Khara et al, 2014
(2Fe-2S)-binding protein (ahdA3)	ahdA3_RT_F	GCAGTATATAATGTTGATGGCG	55	Khara et al, 2014
	ahdA3_RT_R	ATGCCACTTGGCAGG		Khara et al, 2014
Naphthalene dioxygenase (ahdA1f)	ahdA1f_RT_F	ACAAGGGCTTCATTCG	55	Khara et al, 2014
	ahdA1f_RT_R	CTCGTCGGGACTTCC		Khara et al, 2014

**Table S2:** Target genes and primer set designed to amplify genes of the upper and lower PAH degradation pathway of Bk strain.

Gene	Primer set	Sequence (5'-3')	Annealing temprature	Reference
Burkholderia 16S rRNA	Burk_F	CTGCATTTGTGACTGGCGG	60	Pactao Bacosa, 2012
	Burk_R	ACGTTACCAAGTCAATGAAGAC		
Catechol 2,3-dioxygenase	cat_F	CCGCGATGAAGCATACGTG	60	This study
	cat_R	TCAAGGTCGGCCTCTGTTTC		
Naphthalene/biphenyl dioxygenase	Naph_F	TCCGGATAAGGTGTATTCGCT	60	This study
	naph_R	CGGCGTTACGTACGGTAA		
Biphenyl 2,3-dioxygenase	Biph_F	CAGGCCCTGAAGAATTGAC	60	This study
	Biph_R	ATGATATGACCGAGGCCCTG		
Salicylate	Sal_F	TTCTCACCGATGACTGCAC	60	This study
	Sal_R	CCTCCTCGACCTCTTGATG		
Large subunit toluate/benzoate dioxygenase	Tol_F	CGGAAAATACCGTGATCCA	60	This study
	Tol_F	TCCTCCTGCTAGAACCGGA		

**Table S3:** Under-expressed (foldchange > 1) or over-expressed (fold change < -1) proteins for both AM strain (left) and BK strain (right) in coculture, in relation to the axenic culture.

<i>Sphingobium</i> sp. AM			<i>Burkholderia</i> sp. BK		
Protein Name	Accession number-NCBI	fold-change	Protein Name	Accession number-NCBI	Fold change
<b>Xenobiotic Biodegradation and Metabolism</b>					
alkene reductase	KXU30967.1	1,37	hydratase	OWJ56571.1	1,95
benzene 1,2-dioxygenase	KXU30457.1	1,18	alpha/beta hydrolase	OWJ62986.1	1,64
4-oxalocrotonate decarboxylase	KXU30445.1	1,88	NADP(H)-dependent aldo-keto reductase	OWJ60237.1	2,08
2-hydroxy-6-oxo-6-phenylhexa-2,4-dienoate hydrolase	KXU30062.1	2,18	catechol 2,3-dioxygenase	OWJ56357.1	2,15

4-hydroxy-2-oxovalerate aldolase	KXU30446.1	1,12	(2Fe-2S)-binding protein	OWJ56336.1	2,12
anthranilate 1,2-dioxygenase	KXU30454.1	1,98	acetaldehyde dehydrogenase (acetylating)	OWJ61070.1	1,72
haloperoxidase	KXU30540.1	1,62	aromatic-ring-hydroxylating dioxygenase		
aldolase	KXU30430.1	1,55	subunit beta	OWJ56327.1	3,73
oxidoreductase / 4-cresol dehydrogenase	KXU30469.1	1,89	3-oxoadipyl-CoA thiolase	OWJ61198.1	1,20
<b>Carbohydrate Metabolism</b>			aldolase	OWJ56339.1	3,76
malate dehydrogenase	KXU30729.1	1,36	2-keto-4-pentenoate hydratase	OWJ61072.1	5,33
aldehyde dehydrogenase	KXU30518.1	1,26	4-oxalocrotonate decarboxylase	OWJ61071.1	5,14
succinate--CoA ligase subunit alpha	KXU30728.1	2,11	aldehyde dehydrogenase	OWJ56323.1	4,01
phosphoglycerate kinase	KXU32515.1	2,18	hypothetical protein BWU74_31040	OWJ56335.1	4,31
<b>Energy Metabolism</b>			biphenyl-2,3-diol 1,2-dioxygenase	OWJ56344.1	2,89
FOF1 ATP synthase subunit gamma	KXU29630.1	1,47	aldehyde dehydrogenase family protein	OWJ57697.1	-1,58
ETC complex I subunit	KXU29743.1	1,63	<b>Carbohydrate Metabolism</b>		
<b>Translation</b>			dihydroxyacetone kinase subunit DhaK	OWJ57141.1	2,06
30S ribosomal protein S18	KXU32077.1	2,13	phosphoglycerate mutase	OWJ63020.1	5,34
trigger factor	KXU30769.1	1,59	class II aldolase/adducin family protein	OWJ61961.1	1,67
ribonuclease	KXU32371.1	-1,44	fructose-bisphosphatase class I	OWJ59189.1	2,59
isoprenylcysteine carboxyl methyltransferase	KXU32302.1	-1,30	bifunctional D-altronate/D-mannonate		
<b>Cellular Stress</b>			dehydratase	OWJ61184.1	3,82
glutathione S-transferase	KXU30453.1	1,70	isocitrate dehydrogenase (NADP(+))	OWJ58913.1	2,48
thiol:disulfide oxidoreductase	KXU32110.1	1,12	acetate--CoA ligase	OWJ59802.1	-1,03
glutathione peroxidase	KXU32454.1	1,25	pyruvate oxidase	OWJ59284.1	-1,75
Phyllosphere-induced regulator PhyR	KXU32484.1	1,38	<b>Energy Metabolism</b>		
phage shock protein PspA	KXU32744.1	2,19	cytochrome c oxidase subunit II	OWJ63336.1	1,30
co-chaperone YbbN	KXU29975.1	1,46	FOF1 ATP synthase subunit B	OWJ57555.1	2,13
copper resistance protein CopC	KXU31207.1	1,79	<b>Carbohydrates Biosynthesis</b>		
copper resistance protein CopC	KXU33463.1	2,20	alpha-1,4-glucan--maltose-1-phosphate		
exodeoxyribonuclease III	KXU31586.1	1,43	maltosyltransferase	OWJ56795.1	-4,35
ATP-dependent chaperone ClpB	KXU32858.1	-1,19	<b>Translation</b>		
<b>Aminoacid Metabolism</b>			preprotein translocase subunit YajC	OWJ63507.1	1,68
acetylglutamate kinase	KXU30278.1	1,14	[cytidine(C)-cytidine(C)-adenosine (A)]-adding		
acetolactate synthase small subunit	KXU32951.1	1,25	enzyme	OWJ56399.1	2,98
pyrroline-5-carboxylate reductase	KXU31786.1	1,80	30S ribosomal protein S1	OWJ59246.1	1,24
trifunctional transcriptional regulator/proline			50S ribosomal protein L16	OWJ56906.1	2,72
dehydrogenase/L-glutamate gamma-			30S ribosomal protein S9	OWJ58484.1	2,50
semialdehyde dehydrogenase	KXU29470.1	-4,40	50S ribosomal protein L15	OWJ56917.1	1,94
<b>Transporters</b>			50S ribosomal protein L11	OWJ55972.1	1,12
efflux transporter periplasmic adaptor subunit	KXU29878.1	2,40	<b>Transporters</b>		
efflux transporter periplasmic adaptor subunit	KXU30088.1	1,42	50S ribosomal protein L1	OWJ55973.1	1,92
<b>Metabolism of Cofactors and Vitamins</b>			elongation factor G	OWJ56004.1	1,72
pyridoxine 5'-phosphate synthase	KXU30295.1	1,18	50S ribosomal protein L30	OWJ57040.1	3,32
NAD(P)H:quinone oxidoreductase	KXU32046.1	1,39	aspartyl/glutamyl-tRNA amidotransferase		
<b>Iron Metabolism</b>			subunit A	OWJ57736.1	1,19
bacterioferritin	KXU30691.1	2,30	50S ribosomal protein L33	OWJ59111.1	3,90
<b>Cell Cycle</b>			trigger factor	OWJ60122.1	2,28
histidine phosphotransferase	KXU30760.1	1,94	H-NS histone	OWJ57795.1	6,00
<b>Unclassified</b>			ribonuclease E/G	OWJ62377.1	-1,01
oxidoreductase	KXU32266.1	2,08	DNA-directed RNA polymerase subunit beta	OWJ55976.1	-2,81
hypothetical protein AXW74_01365	KXU33610.1	1,60	alanine--tRNA ligase	OWJ62463.1	-1,44
hypothetical protein AXW74_00325	KXU33768.1	1,15	<b>Cellular Stress</b>		
hypothetical protein AXW74_22915	KXU29437.1	2,53	transposase	OWJ56945.1	1,60
hypothetical protein AXW74_00530	KXU33808.1	-1,16	DNA starvation/stationary phase protection		
			protein	OWJ63190.1	1,06
			peroxiredoxin	OWJ61569.1	2,15
			Fertility inhibition FinO-like protein	OWJ60700.1	1,21
			polyisoprenoid-binding protein	OWJ61284.1	6,51
			organic hydroperoxide resistance protein	OWJ56783.1	1,92
			nucleotide exchange factor GrpE	OWJ63506.1	3,36
			exodeoxyribonuclease III	OWJ58150.1	2,55
			FMN-dependent NADH-azoreductase	OWJ57026.1	4,80
			Nucleotid Metabolism		
			NADP-dependent 3-hydroxy acid	OWJ63145.1	1,89
			dehydrogenase		
			Zn-dependent hydrolase	OWJ60603.1	-2,85

phosphoribosylformylglycinamidine cycloligase	OWJ63156.1	-1,06
<b>Environmental Information Processing</b>		
two-component system response regulator	OWJ57723.1	3,85
transcriptional regulator	OWJ62876.1	1,95
<b>Aminoacid Metabolism</b>		
RidA family protein	OWJ63256.1	1,36
succinylglutamate desuccinylase	OWJ61179.1	1,12
3-methylcrotonyl-CoA carboxylase	OWJ61903.1	1,98
3-keto-5-aminohexanoate cleavage protein	OWJ60369.1	3,56
NAD-dependent succinate-semialdehyde dehydrogenase	OWJ59895.1	2,81
3-(cis-5,6-dihydroxycyclohexa-1,3-dien-1-yl)propanoate dehydrogenase	OWJ56326.1	3,42
hypothetical protein BWU74_31035 (choline dehydrogenase)	OWJ56334.1	6,21
dihydrolipoyl dehydrogenase	OWJ60520.1	-1,28
acyl-CoA dehydrogenase	OWJ61976.1	-2,38
NAD-glutamate dehydrogenase	OWJ62164.1	-4,13
tryptophan synthase subunit beta	OWJ56811.1	1,28
histidinol dehydrogenase	OWJ56974.1	2,21
4-hydroxy-tetrahydroadipicolic acid reductase	OWJ58511.1	2,30
anthranilate synthase component I	OWJ57031.1	1,66
aspartate 4-decarboxylase	OWJ63125.1	1,21
methionine adenosyltransferase	OWJ57760.1	1,61
low-specificity L-threonine aldolase	OWJ58269.1	1,98
cystathione beta-lyase	OWJ60154.1	2,46
succinyl-diaminopimelate desuccinylase	OWJ60752.1	3,18
dihydroadipicolic acid synthase family protein	OWJ61754.1	3,97
dihydroadipicolic acid synthase family protein	OWJ56330.1	3,50
acetolactate synthase small subunit	OWJ62559.1	-1,19
argininosuccinate synthase	OWJ58355.1	-1,16
<b>Transporters</b>		
ABC transporter substrate-binding protein	OWJ61663.1	2,12
metal ABC transporter substrate-binding protein	OWJ63323.1	1,86
sugar ABC transporter	OWJ62205.1	1,75
amino acid ABC transporter	OWJ57892.1	2,56
metal ABC transporter substrate-binding protein	OWJ59258.1	2,45
carbohydrate ABC transporter substrate-binding protein	OWJ62924.1	1,14
sulfate transporter subunit	OWJ60229.1	2,19
ABC transporter substrate-binding protein	OWJ60343.1	1,79
thiosulfate transporter subunit	OWJ63418.1	1,11
branched-chain amino acid ABC transporter substrate-binding protein	OWJ61825.1	2,03
D-xylose ABC transporter substrate-binding protein	OWJ58976.1	2,77
amino acid ABC transporter substrate-binding protein	OWJ58492.1	-1,96
sugar ABC transporter substrate-binding protein	OWJ62209.1	-2,95
<b>Metabolism of Cofactors and Vitamins</b>		
molybdopterin adenylyltransferase	OWJ62346.1	3,83
methylenetetrahydromethanopterin dehydrogenase	OWJ61862.1	1,30
dihydropteroate synthase	OWJ62434.1	2,81
methenyltetrahydromethanopterin cyclohydrolase	OWJ61971.1	1,17
<b>LPS Biosynthesis and Outer Membrane Stability</b>		
3-deoxy-8-phosphooctulonate synthase	OWJ60498.1	1,54
UDP-3-O-(3-hydroxymyristoyl)glucosamine N-acetyltransferase	OWJ60389.1	1,59
phosphomannomutase/phosphoglucomutase	OWJ63004.1	1,30

UDP-N-acetylglucosamine diphosphorylase/glucosamine-1-phosphate N-acetyltransferase	OWJ56404.1	1,06
NlpB/DapX lipoprotein domain protein /bamC outer membrane protein assembly factor	OWJ60590.1	2,26
BamB	OWJ60478.1	3,02
peptidoglycan-associated lipoprotein	OWJ63151.1	3,26
Peptidoglycan Biosynthesis		
UDP-N-acetylglucosamine 1-carboxyvinyltransferase	OWJ57047.1	-2,40
<b>Fatty acids metabolism</b>		
oxidoreductase	OWJ60701.1	1,83
acetyl-CoA carboxylase carboxyl transferase subunit alpha	OWJ60449.1	1,62
beta-ketoacyl-[acyl-carrier-protein] synthase II	OWJ62391.1	2,11
oxidoreductase	OWJ60321.1	2,33
NAD(P)-dependent oxidoreductase	OWJ56852.1	2,35
acyl-CoA dehydrogenase	OWJ63410.1	-2,85
<b>Glyoxylate cycle</b>		
bifunctional glyoxylate/hydroxypyruvate reductase B	OWJ56588.1	2,18
hydroxypyruvate isomerase	OWJ61196.1	2,29
<b>Quorum sensing</b>		
response regulator	OWJ57306.1	1,74
<b>Virulence Factors</b>		
polyketide cyclase	OWJ62224.1	3,66
Hcp1 family type VI secretion system effector	OWJ57950.1	3,36
polysaccharide deacetylase	OWJ56854.1	2,08
<b>Iron Metabolism</b>		
porphobilinogen synthase	OWJ56928.1	2,34
TonB-dependent siderophore receptor	OWJ59615.1	1,33
glutamate-1-semialdehyde-2,1-aminomutase	OWJ59169.1	2,87
ATP-cob(I)alamin adenosyltransferase	OWJ56324.1	4,87
<b>Motility</b>		
flagellar motor protein MotB	OWJ57956.1	4,39
flagellin	OWJ57514.1	-2,31
<b>Cellular Cycle</b>		
septum site-determining protein MinD	OWJ62907.1	-1,59
<b>Unclassified</b>		
LLM class flavin-dependent oxidoreductase	OWJ56342.1	3,30
2-aminoethylphosphonate aminotransferase	OWJ58158.1	2,61
cupin	OWJ57093.1	2,93
disulfide bond formation protein DsbA	OWJ63400.1	2,84
carbonic anhydrase	OWJ61947.1	2,97

**Table S4:** AM and BK proteins expressed uniquely in the axenic cultures or in DSC-AB.

<b>BK Proteins observed uniquely in DSC_AB culture</b>		
<b>Motility</b>		
methyl-accepting chemotaxis protein [Burkholderia sp. Bk]	OWJ56685.1	
<b>BK Proteins observed uniquely in axenic culture</b>		
<b>Xenobiotic metabolism</b>		
catechol 1,2-dioxygenase [Burkholderia sp. Bk]	OWJ59967.1	
(2Fe-2S)-binding protein [Burkholderia sp. Bk]	OWJ56343.1	
1,6-dihydroxycyclohexa-2,4-diene-1-carboxylate dehydrogenase [Burkholderia sp. Bk]	OWJ59963.1	
<b>Carbohydrate Metabolism</b>		
2-methylcitrate dehydratase [Burkholderia sp. Bk]	OWJ56714.1	
<b>Translation</b>		
23S rRNA pseudouridylate synthase B [Burkholderia sp. Bk]	OWJ59750.1	

50S ribosomal protein L31 [Burkholderia sp. Bk]	OWJ60184.1
peptidylprolyl isomerase [Burkholderia sp. Bk]	OWJ60461.1
<b>LPS Biosynthesis and Outer Membrane Stability</b>	
glycosyltransferase [Burkholderia sp. Bk]	OWJ60203.1
3-deoxy-manno-octulosonate cytidylyltransferase [Burkholderia sp. Bk]	OWJ58348.1
<b>Aminoacid Metabolism</b>	
glutamate synthase [Burkholderia sp. Bk]	OWJ56947.1
cytosine deaminase [Burkholderia sp. Bk]	OWJ60529.1
hypothetical protein BWU74_02270 [Burkholderia sp. Bk]	OWJ63233.1
<b>Cellular stress</b>	
copper resistance protein CopC [Burkholderia sp. Bk]	OWJ57235.1
<b>Unclassified</b>	
oxidoreductase [Burkholderia sp. Bk]	OWJ62976.1
<b>AM Proteins observed uniquely in DSC_AB culture</b>	
<b>Aminoacid Metabolism</b>	
cysteine synthase [Sphingobium sp. AM]	KXU31626.1
<b>AM Proteins observed uniquely in axenic culture</b>	
<b>Transcription</b>	
DEAD/DEAH box helicase [Sphingobium sp. AM]	KXU30904.1
<b>Cellular stress</b>	
peroxidase [Sphingobium sp. AM]	KXU31240.1
<b>Fatty acid metabolism</b>	
acetyl-CoA carboxylase carboxyltransferase subunit alpha [Sphingobium sp. AM]	KXU30696.1
<b>Transporters</b>	
ABC transporter ATP-binding protein [Sphingobium sp. AM]	KXU30162.1