

Supplementary Table S5: Substrate utilization of *Bradyrhizobium* strains a described *Bradyrhizobium* type strains of the *B. elkanii* supergroup.

Substrate utilization	<i>Bradyrhizobium</i> strains <sup>a</sup>														<i>Bradyrhizobium</i> type strains <sup>b</sup>										
	<i>B. oropedii</i>				<i>B. acaciae</i>				<i>B. brasilense</i>				<i>B. ivorensis</i>	<i>B. altum</i>											
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	a	b	c	d	e	f	g	h	i	j	k
Reduction of nitrates to nitrogen	+	-	-	-	+	-	+	+	-	-	-	+	-	+	+	+							+	-	+
L-tryptophane (indole production)	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-							-	-	-
D-glucose (glucose fermentation)	+	-	-	-	-	-	-	-	-	-	-	-	-	-	+	+	-							+	-
L-arginine DiHydrolase	+	-	+	-	-	+	-	-	-	+	-	-	+	-	-	-							+	+	+
Urease	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+		+	+				+	-	+	+
Esculin ferric citrate (hydrolysis $\beta$ -glucosidase)	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	w							+	+	w
gelatin (bovin origin)	+	-	-	-	-	-	-	-	-	-	-	-	-	+	-								-	-	-
4-nitrophenyl- $\beta$ D-galactoyranoside ( $\beta$ -galactosidase)	+	+	-	w	+	w	+	+	-	w	-	-	-	-	-	-							-	-	-
Potassium gluconate	-	+	-	+	+	+	+	+	-	-	w	+	+	-	+		+	-				-	-		+
Capric acid	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-										-
Adipic acid	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-										-
Malic acid	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-									-	w
Trisodium citrate (Citrate utilization)	+	-	-	-	-	-	-	-	-	w	-	-	-	-	-	-								w	
Phenylacetic acid	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-									w	
$\alpha$ -cyclodextrin	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-										
Dextrin	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-										
Glycogen	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-		+	-				-	v		
Tween 40	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+				+	+					
Tween 80	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+					-	+				
N-acetyl-D-galactosamine	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-										
N-Acetyl-D-glucosamine	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-				-	-		+
Adonitol	-	-	-	-	-	-	-	-	+	-	-	-	-	+	-		-	w				w	w		
L-arabinose	+	+	-	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	-	+	+	+	+	+	+
D-arabitol	+	+	-	+	-	-	-	-	+	w	+	+	+	+	+		w	w	+	+		w	v		
D-cellobiose	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-				-			
i-erythritol	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-							-	v		
D-fructose	+	+	+	w	w	-	-	-	+	w	w	+	+	+	+			w	+	+	+	+	+	+	w
L-fructose	w	+	+	+	+	+	+	+	+	+	+	+	+	+	+										

D-galactose	w	+	-	w	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	w	+	+	+	+	
Gentiobiose	-	w	-	-	-	-	-	-	-	-	-	-	-	-	+	-	+	-	-	-	-	-	-	-	-	
$\alpha$ -D-glucose	w	w	+	-	w	w	w	+	w	w	+	+	w	+	w					-	+					
m-inositol	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-							-	v			
$\alpha$ -D-lactose	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-								w		w	
Lactulose	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-											
Maltose	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	v	-	-				-	v	-	+	-
D-mannitol	+	+	+	+	-	-	-	-	+	w	-	+	w	+	+	+	w	w	+	+	w	+	+	+	+	+
D-mannose	-	w	+	-	w	-	w	-	-	-	-	-	w	+	w	-	+	+	-	-	+	+		+		
D-melibiose	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-							-				
$\beta$ -methyl-D-glucoside	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-											
D-psicose	w	-	-	-	-	-	-	-	w	-	-	-	-	+	-											
D-raffinose	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-			-	-			-				
L-rhamnose	+	w	-	-	+	w	+	+	w	w	w	+	w	+	w	+	w	w	+	+	w	w				
D-sorbitol	w	+	+	-	-	-	-	-	-	w	w	w	w	+	w			w	w	-	+	w	+			
Sucrose	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-			-				-	-	-		-
D-trehalose	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-			-	-			-	-			
Turanose	-	-	-	-	w	-	-	-	-	-	-	-	-	+	-			-	-			-	w			
Xylitol	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-			-	-			w	+			
mono-methyl-succinate	+	+	-	+	+	+	+	+	+	+	+	+	w	+	+											
acetic acid	w	w	-	-	-	-	-	w	-	-	-	-	-	-	w						+	+				
cis-aconitic acid	w	-	-	-	-	-	-	-	-	-	-	-	-	+	-						+	-				
citric acid	w	w	-	-	w	+	w	+	-	+	w	+	+	+	+						+	+			-	-
formic acid	w	w	+	-	w	+	w	+	-	+	w	+	+	+	+						+	+				
D-galactonic acid lactone	w	w	-	-	w	w	w	w	-	+	w	+	+	+	w						+	+				
D-galacturonic acid	w	w	-	-	w	w	w	-	+	-	-	w	-	+	w						-	+				
D-gluconic acid	+	+	+	w	+	+	+	+	w	+	+	+	+	+	+						-	-				
D-glucosaminic acid	w	w	-	-	-	-	-	-	-	+	+	+	+	+	+						+	+				
D-glucuronic acid	w	-	-	-	-	-	-	-	w	-	-	-	-	+	-						-	-				
$\alpha$ -hydroxy butyric acid	+	+	+	w	+	+	+	+	+	+	+	+	+	-	+						+	+				
$\beta$ -hydroxy butyric acid	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+											
$\gamma$ -hydroxy butyric acid	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+						+	+				
p-hydroxy phenylacetic acid	w	+	+	+	w	+	+	+	+	+	+	+	+	+	+						-	-				
itaconic acid	-	w	-	-	-	-	-	-	-	-	-	w	+	-	-						+	+				
$\alpha$ -keto butyric acid	+	+	-	+	+	+	+	+	w	+	+	+	+	-	+						+	+				
$\alpha$ -keto glutaric acid	+	w	+	-	+	+	+	w	w	+	+	+	+	+	w						+	+				
$\alpha$ -keto valeric acid	-	w	-	-	+	+	-	w	-	w	w	w	+	-	w						+	-				
D,L-lactic acid	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+						-	+				

malonic acid	w	-	-	-	-	-	-	-	-	-	-	w	+	+	-					+	-						
propionic acid	w	+	-	w	w	+	-	+	-	w	w	+	+	+	+					+	+						
quinic acid	w	-	-	-	-	-	-	-	-	-	-	-	-	+	-					+	-						
succinic acid	+	+	+	-	+	+	+	+	-	+	+	+	+	+	+					+	+						
D-saccharic acid	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+					-	+						
sebacic acid	+	+	-	w	+	+	+	+	+	+	+	+	+	+	+					+	+						
bromo succinate acid	+	w	-	-	+	+	+	+	w	w	w	w	+	+	+					+	+						
succinamic acid	+	+	-	w	+	+	+	+	+	+	+	+	+	+	+												
Glucuronamide	+	-	+	-	+	+	+	w	+	w	+	+	w	+	+					-	+						
L-alaninamide	+	+	+	w	+	+	+	+	w	+	+	+	+	w	+					+	+						
D-alanine	+	+	-	w	+	+	w	+	w	+	+	+	+	w	w					+	+						
L-alanine	+	+	-	w	+	+	+	+	w	+	+	+	+	+	w					-	+						
L-alanyl-glycine	+	+	-	w	+	+	w	+	w	-	w	w	+	+	w					+	+						
L-asparagine	+	w	-	-	+	+	+	+	-	+	+	+	+	+	w					+	+				w		-
L-aspartic acid	+	w	-	-	+	w		+	-	+	w	+	+	+	w					+	+						
L-glutamic acid	+	w	-	-	+	+	+	+	-	+	w	+	+	+	w					+	+				+		+
glycyl-L-aspartic acid	w	w	-	-	-	-	-	-	-	-	w	w	-	-	-												
glycyl-L-glutamic acid	w	+	-	-	+	+	+	+	-	w	+	+	w	-	+												
L-histidine	-	-	+	-	-	-	-	-	-	-	-	-	-	+	-												
hydroxy-L-proline	-	-	+	-	-	-	-	-	-	-	-	-	-	+	-												
L-leucine	+	+	+	w	+	+	+	+	w	+	+	+	+	+	+					+	+						
L-ornithine	w	-	-	-	-	-	-	-	-	-	-	-	-	+	-												
L-phenylalanine	+	w	+	-	w	+	w	w	-	-	w	w	+	+	w					+	+						
L-proline	+	w	-	-	w	-	w	-	-	-	w	w	-	+	w					-	-						
L-pyroglutamic acid	+	+	-	w	+	+	+	+	w	+	+	w	+	+	+					+							
D-serine	w	w	-	-	w	-	w	-	w	-	-	-	-	-	-					+	+						
L-serine	w	-	-	-	w	-	w	-	-	-	-	-	-	-	-					+	-						
L-threonine	-	-	-	-	+	-	w	-	-	-	-	w	-	-	-					-	+						
D,L-carnitine	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-												
$\gamma$ -amino butyric acid	-	-	-	-	-	-	w	-	-	-	-	-	w	+	w					+	-						
urocanic acid	+	+	+	w	+	+	+	+	+	w	w	+	+	+	w					+	+						
Inosine	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-												
Uridine	-	-	-	-	-	-	-	-	-	-	w	-	-	-	-												
Thymidine	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-												
Phenyethylamine	w	-	-	-	-	-	-	-	-	-	-	-	-	-	-					+	-						
Putrescine	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-												
2-aminoethanol	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-												
2,3-butanediol	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-												

<b>Glycerol</b>	+	+	-	+	+	+	+	+	w	+	+	+	-	+	+			w	+	+	+	+	w		w
<b>D,L-<math>\alpha</math>-glycerol phosphate</b>	w	-	-	-	-	-	-	-	-	-	-	-	-	+	-										
<b>glucose-1-phosphate</b>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-										
<b>glucose-6-phosphate</b>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-										
<b>Oxidase</b>	+	+	+	+	+	+	+	+	+	+	+	+	+	+											+
<b>Catalase</b>	+	+	+	+	+	+	+	+	+	+	+	+	+	+											-

Results are indicated as ‘+’ positive for substrate utilization, ‘w’ weak for substrate utilization, ‘-’ negative for substrate utilization, ‘v’ for variable results between strains within a species and grey blocks indicates no data available. Thick black lines are used to separate strains of the same species.

<sup>a</sup> Isolates are numbered (1-14): (1) Pear128, (2) Pear129, (3) Pear76<sup>T</sup>, (4) Leo176, (5) 19AH, (6) 19AJ, (7) 10BB<sup>T</sup>, (8) 16BA no1,) (9) Arg62, (10) R5, (11) Cham227, (12) Cham231, (13) Arg68 (14) Pear77<sup>T</sup>.

<sup>b</sup>*Bradyrhizobium* type strains: (a) *B. elkanii* USDA6<sup>T</sup> (This study), (b) *B. pachyrhizi* PAC48<sup>T</sup> (Ramírez-Bahena et al., 2009), (c) *B. embrapense* SEMIA6208<sup>T</sup> (Delamuta et al., 2015), (d) *B. tropiciagri* SEMIA6148<sup>T</sup> (Delamuta et al., 2015), (e) *B. erythrophlei* CCBAU 53325<sup>T</sup> (Yao et al., 2015), (f) *B. ferriligni* CCBAU 51502<sup>T</sup> (Yao et al., 2015), (g) *B. viridifuturi* SEMIA690<sup>T</sup> (Helene et al., 2015), (h) *B. mercantei* SEMIA6399<sup>T</sup> (Helene et al., 2017), (i) *B. brasilense* (da Costa et al., 2017), (j) *B. ivorensis* CI-1B<sup>T</sup> (Fossou et al., 2019), (k) *B. uaiense* UFLA03-164<sup>T</sup> (Michel et al., 2020). Results of API 20NE and Biolog test for each respective *Bradyrhizobium* described species (i.e. based on results available for all strains within a species) was obtained from published species descriptions.