Phenotype Microarray Report

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Organism: *Genus species* (WT & Mutant)
Phenotype MicroArray Plates: PM 1
Starter Agar/Broth: LB + Carbenicillin (WT), LB+ Chloramphenicol & Kanamycin (Mutant)
Temperature: 37˚C
Protocol: Gram-negative PM
Redox Dye Mix: Dye D
Carbon Source: NA
Additional Additives: NA
Replicates: NA
Report Date: 12/20/2023

Thank you for providing your *Genus specie* isolates for Phenotypic MicroArray (PM) characterization. We have a single run for your *Genus species* WT and Mutant organisms. The *G. species* WT and mutant were subcultured and struck for isolation on LB+Carbenicillin agar and LB + Chloramphenicol & Kanamycin agar, respectively, for 24 hours at 37˚C. We followed the Biolog Gram-negative PM protocol, by inoculating each organism in IF-0a inoculating fluid to a density of 42% transmittance using a turbidimeter. Based on previous data, redox dye D was used to assess cellular metabolism. The cell suspensions with redox dye were inoculated into their respective PM 1 plates at 100 µl per well. Each plate was placed on Odin set to 37˚C and the kinetic data was collected every 20 minutes for 48 hours.

Below you will find the dye reduction kinetic curves generated from Odin. The image of the kinetic curves displays the OD590 values collected over time (48 hours) for PM plate 1, as requested. The graph displays the kinetic overlay of absorbance values for the WT in gold and mutant strain in black over time for each strain (time in hours on the x-axis, absorbance at OD590nm on the y-axis). We have also included a map of the PM plate for your reference.

You will also find pairwise comparisons of the mutant strain compared to the WT. The wild-type or reference strain is in pink, and the mutant or test strain is in light blue. The dark blue area indicates the overlay between the two strains. From the kinetic analysis, we calculated the difference in the maximum rate between the WT and mutant *G. species*. The maximum rate is defined as the single line fit to the plot of kinetic data that has the largest slope (in OD units per hour). Maximum rates with positive values indicate a gain in the metabolic phenotype of the mutant compared to the WT. In contrast, negative maximum rates indicate a loss of metabolic phenotype of the mutant compared to the WT.

Generally, there were no large differences in the maximum rate between the WT and mutant strains. You will find below the top 10 gain of utilization phenotypes in the mutant strain and the top 10 loss of utilization phenotypes in the mutant strain.

In addition to this report, you will find an Excel spreadsheet with the raw OD data values generated from Odin. Each sheet contains the data for each PM plate run. There is a sheet that lists the differences in the maximum rate of each substrate of the mutant compared to the WT.

Thank you for choosing Biolog Lab Services!

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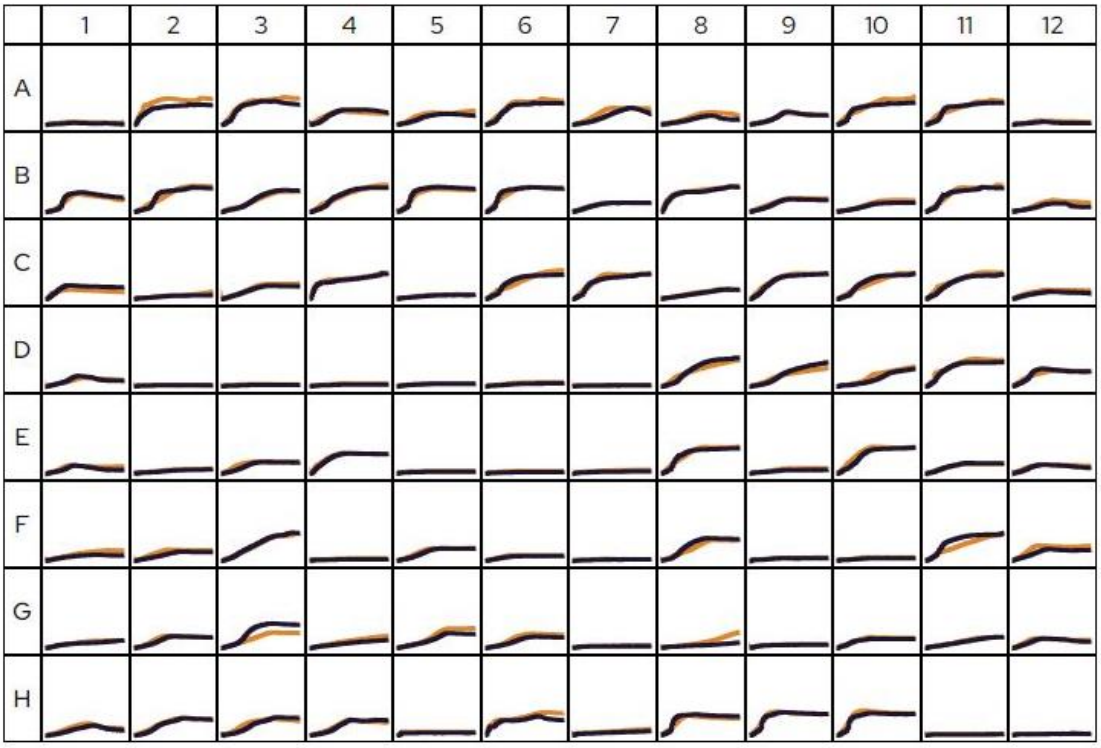
48-hour Dye Reduction Kinetic Curves for *G. specie* (WT and Mutant)

PM1 Carbon Utilization Assays

Catalog #12111

A1 Negative Control	A2 L-Arabinose	A3 N-Acetyl-D-Glucosamine	A4 D-Saccharic Acid	A5 Succinic Acid	A6 D-Galactose	A7 L-Aspartic Acid	A8 L-Proline	A9 D-Alanine	A10 D-Trehalose	A11 D-Mannose	A12 Dulcitol
B1 D-Serine	B2 D-Sorbitol	B3 Glycerol	B4 L-Fucose	B5 D-Glucuronic Acid	B6 D-Gluconic Acid	B7 D,L-α-Glycerol-Phosphate	B8 D-Xylose	B9 L-Lactic Acid	B10 Formic Acid	B11 D-Mannitol	B12 L-Glutamic Acid
C1 D-Glucose-6-Phosphate	C2 D-Galactonic Acid-γ-Lactone	C3 D,L-Malic Acid	C4 D-Ribose	C5 Tween 20	C6 L-Rhamnose	C7 D-Fructose	C8 Acetic Acid	C9 α-D-Glucose	C10 Maltose	C11 D-Melibiose	C12 Thymidine
D1 L-Asparagine	D2 D-Aspartic Acid	D3 D-Glucosaminic Acid	D4 1,2-Propanediol	D5 Tween 40	D6 α-Keto-Glutaric Acid	D7 α-Keto-Butyric Acid	D8 α-Methyl-D-Galactoside	D9 α-D-Lactose	D10 Lactulose	D11 Sucrose	D12 Uridine
E1 L-Glutamine	E2 m-Tartaric Acid	E3 D-Glucose-1-Phosphate	E4 D-Fructose-6-Phosphate	E5 Tween 80	E6 α-Hydroxy Glutaric Acid-γ-Lactone	E7 α-Hydroxy Butyric Acid	E8 ß-Methyl-D-Glucoside	E9 Adonitol	E10 Maltotriose	E11 2-Deoxy Adenosine	E12 Adenosine
F1 Glycyl-L-Aspartic Acid	F2 Citric Acid	F3 myo-Inositol	F4 D-Threonine	F5 Fumaric Acid	F6 Bromo Succinic Acid	F7 Propionic Acid	F8 Mucic Acid	F9 Glycolic Acid	F10 Glyoxylic Acid	F11 D-Cellobiose	F12 Inosine
G1 Glycyl-L-Glutamic Acid	G2 Tricarballylic Acid	G3 L-Serine	G4 L-Threonine	G5 L-Alanine	G6 L-Alanyl-Glycine	G7 Acetoacetic Acid	G8 N-Acetyl-ß-D-Mannosamine	G9 Mono Methyl Succinate	G10 Methyl Pyruvate	G11 D-Malic Acid	G12 L-Malic Acid
H1 Glycyl-L-Proline	H2 p-Hydroxy Phenyl Acetic Acid	H3 m-Hydroxy Phenyl Acetic Acid	H4 Tyramine	H5 D-Psicose	H6 L-Lyxose	H7 Glucuronamide	H8 Pyruvic Acid	H9 L-Galactonic Acid-γ-Lactone	H10 D-Galacturonic Acid	H11 Phenylethyl-amine	H12 2-Aminoethanol



G. specie: Mutant

G. specie: WT

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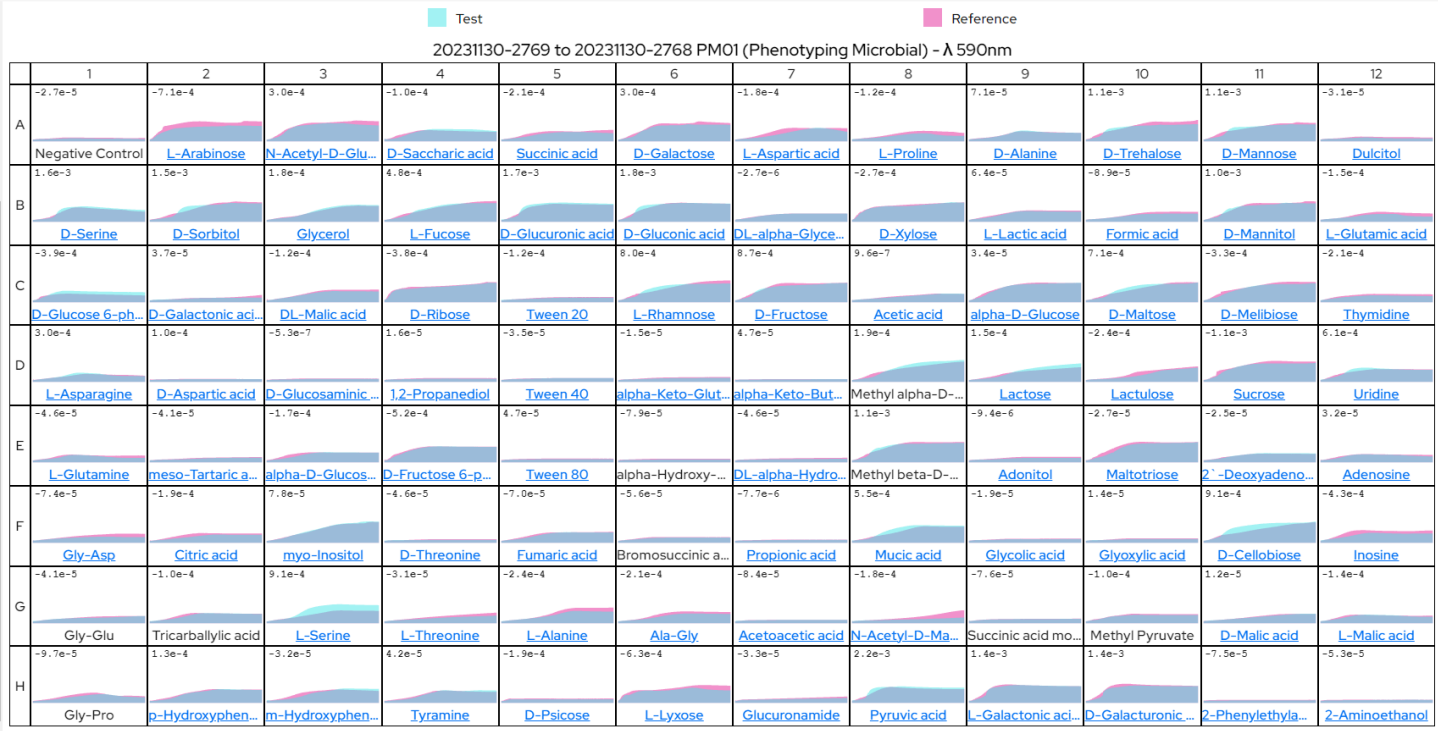
G. species WT vs. Mutant

Test: *G. species* Mutant

Reference: *G. species* WT



Metabolics Gained in Mutant Phenotype

Chemical	Well	Difference in Maximum Rate	Info
Pyruvic acid	H08	2.17E-03	C- Source; carboxylic acid
D-Gluconic acid	B06	1.82E-03	C- Source; carboxylic acid
D-Glucuronic acid	B05	1.74E-03	C- Source; carboxylic acid
D-Serine	B01	1.58E-03	C- Source; amino acid
D-Sorbitol	B02	1.54E-03	C-Source; carbohydrate
L-Galactonic acid lactone	H09	1.38E-03	C- Source; carboxylic acid
D-Galacturonic acid	H10	1.38E-03	C- Source; carboxylic acid
B-Methyl Glucoside	E08	1.14E-03	C-Source; carbohydrate
D-Mannose	A11	1.07E-03	C-Source; carbohydrate
D-Trehalose	A10	1.05E-03	C-Source; carbohydrate

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Metabolics Lost in Mutant Phenotype

Chemical	Well	Difference in Maximum Rate	Info
Sucrose	D11	-1.10E-03	C- Source; carbohydrate
L-Arabinose	A02	-7.11E-04	C- Source; carbohydrate
L-Lyxose	H06	-6.26E-04	C- Source; carbohydrate
Fructose 6 phosphate	E04	-5.22E-04	C- Source; carbohydrate
Inosine	F12	-4.31E-04	C-Source; nucleoside
Glucose 6 phosphate	C01	-3.91E-04	C- Source; carbohydrate
D-Ribose	C04	-3.77E-04	C- Source; carbohydrate
D-Melibiose	C11	-3.29E-04	C- Source; carbohydrate
D-Xylose	B08	-2.71E-04	C- Source; carbohydrate
Lactulose	D10	-2.41E-04	C- Source; carbohydrate