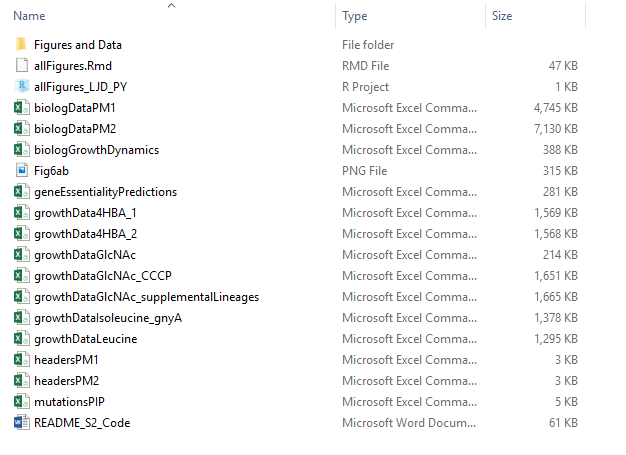
**S2 Code. R code and data to regenerate figures**

**Software information:**

R Version 3.4.0

**Installation:**

* Download RStudio (open source license) from <https://www.rstudio.com/products/rstudio/download/>
* Create a new project in the S1 Data folder by opening RStudio, selecting File->New Project… -> New Directory -> Empty Project. Enter a directory name (can be anything you want), and choose S1 Data as the subdirectory. Make sure that the project is in the S1 Data folder and not in a subfolder.
* Your S1 Data folder should now look like this: (where in this case allFigures\_LJD\_PY is the project file)



**Required R packages:**

Once RStudio is running, the following packages need to be installed in order for the supplied script to be able to run:

* readr
* dplyr
* ggplot2
* ggthemes
* tidyr
* viridis
* gridExtra
* grid
* knitr
* reshape2
* gplots
* RColorBrewer
* ggsignif
* cowplot
* ggpubr
* gtable
* png

All available using install.packages(‘packageName’), or the first chunk of code in allFigures.Rmd can be uncommented the first time the file is run.

**Scripts:**

* allFigures.Rmd

**Data files:**

* biologDataPM1.csv – All growth data on Phenotypic Microarray Plate PM1
* biologDataPM2.csv – All growth data on Phenotypic Microarray Plate PM2a
* headersPM1.csv – Carbon source labels from plate PM1
* headersPM2.csv – Carbon source labels from plate PM2a
* biologGrowthDynamics.csv – All calculated growth dynamics
* growthDataGlcNAc.csv – Growth data from Figure 4
* mutationsPIP.csv – Mutated genes in the piperacillin-evolved lineage
* geneEssentialityPredictions.csv – Predicted essential genes by carbon source
* growthDataLeucine.csv – Growth data of ancestor, PIP, and *gnyA* on 40mM L-Leucine (all 4 biological replicates)
* growthData4HBA\_1.csv – Half of the growth data on 4HBA
* growthData4HBA\_2.csv – The other half of the growth data on 4HBA
* growthDataIsoleucine\_gnyA.csv – Growth data from S6 Fig.
* growthDataGlcNAc\_supplementalLineages.csv – Growth data from S3 Fig.
* growthDataGlcNAc\_CCCP.csv – Growth data from S4 Fig.
* Fig6ab.png – Figure 6A-B which is incorporated into the code for Fig 6.
* Figures and Data folder – Empty folder where figures/data generated by the script will be saved

**Instructions:**

* Prior to this point, RStudio must be installed with the appropriate packages and a new project must be created in the S1 Data folder.
* Open the project in the S1 Data folder.
* Open the script allFigures.Rmd in the project (File -> Open File…).
* Run the script by clicking Knit->Knit to HTML in the upper left-hand corner of the window.
* This will generate the following:
  + An HTML file containing all of the figures and figure captions
  + Figures 2-6 from the main manuscript as .pdf files
  + Figures S1-S6 as .pdf files
  + Data S1-S4 as .csv files

Figures can easily be changed to different file formats (e.g. jpg, png, etc.) or saved to different locations by changing adjusting the filenames and paths within ggsave functions. A call to ggsave can be found at the bottom of the code for each figure.