Supplemental material for:

A metabolomics and proteomics study of the adaptation of *Staphylococcus aureus* to glucose starvation

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Supplemental figures:

Figure S1

Selected results of metabolome and proteome analyses for *S. aureus* COL grown in chemical defined medium used for reconstructing growth-phase dependent central metabolism. **(A)** Overlay of GC-MS spectra from all nine sampling points across growth, selected metabolites were indicated by numbers. **(B)** 2-D gel-based expression analysis visualized by Dual Channel Imaging: The overlay of the false color image of growing cells t_0/OD = 0.5 (green) and non-growing cells t_{24}/OD = 2.5 (red) is shown. This overlay results in the following color code: i) proteins, whose level strongly increased during entry into stationary phase (red color), ii) proteins the level of which did not respond to the nutrient status and remained unchanged (yellow color) and iii) proteins which were present at diminished level in stationary phase compared to exponentially growing cells (green color). Selected protein abbreviations were colored depending on their pathway they are belonging to: white for glycolysis and gluconeogenesis, blue for TCC and green for PPP. Protein abbreviations were listed in **tab.S4A**, TMS –trimethylsilyl-derivative

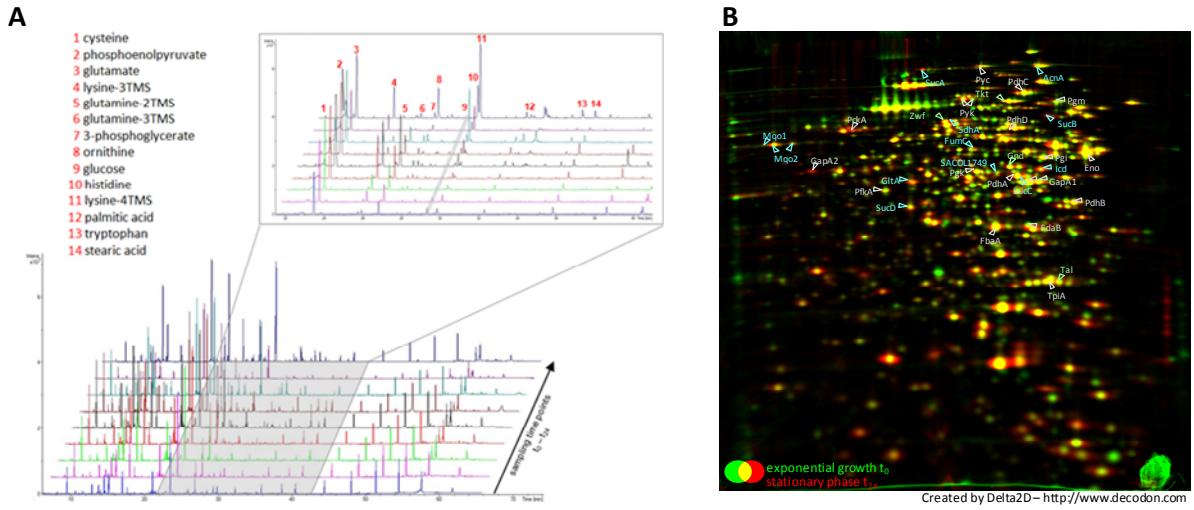


Figure S2

Growth curve of *Staphylococcus aureus* COL in chemical defined medium with glucose as main carbon source and 1 mM amino acids as described in material and methods. The arrow marks the addition of 7.5 mM glucose-solution to stationary phase cells and indicates recovering of growth.

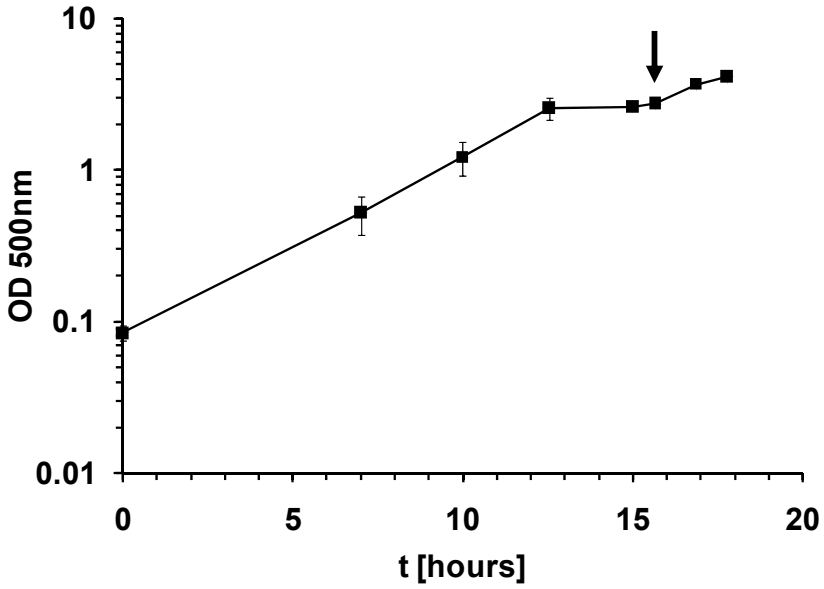


Figure S3

Statistical evaluation of proteome data: (A) Proteins from selected pathways (see material and methods main text for details) with significantly changed expression profiles determined by ANOVA (α=0.05, distribution based on 1000 permutations). For each time point 3 biological replicates were analyzed Red – significantly induced proteins, green – significantly repressed proteins. (Grey – no data available for respective replicate) **(B)** Expression profiles of significantly changed proteins (red – induced, green – repressed). Significance was determined by ANOVA**.**

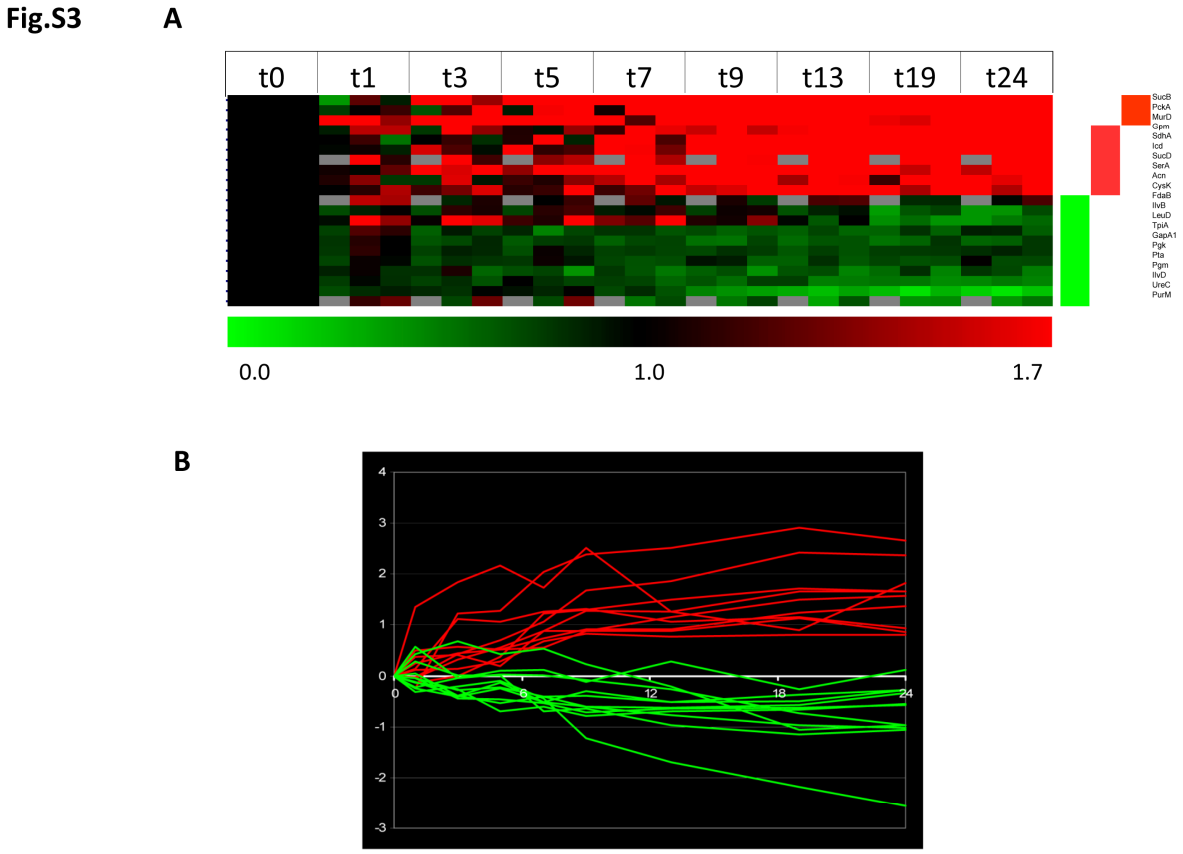
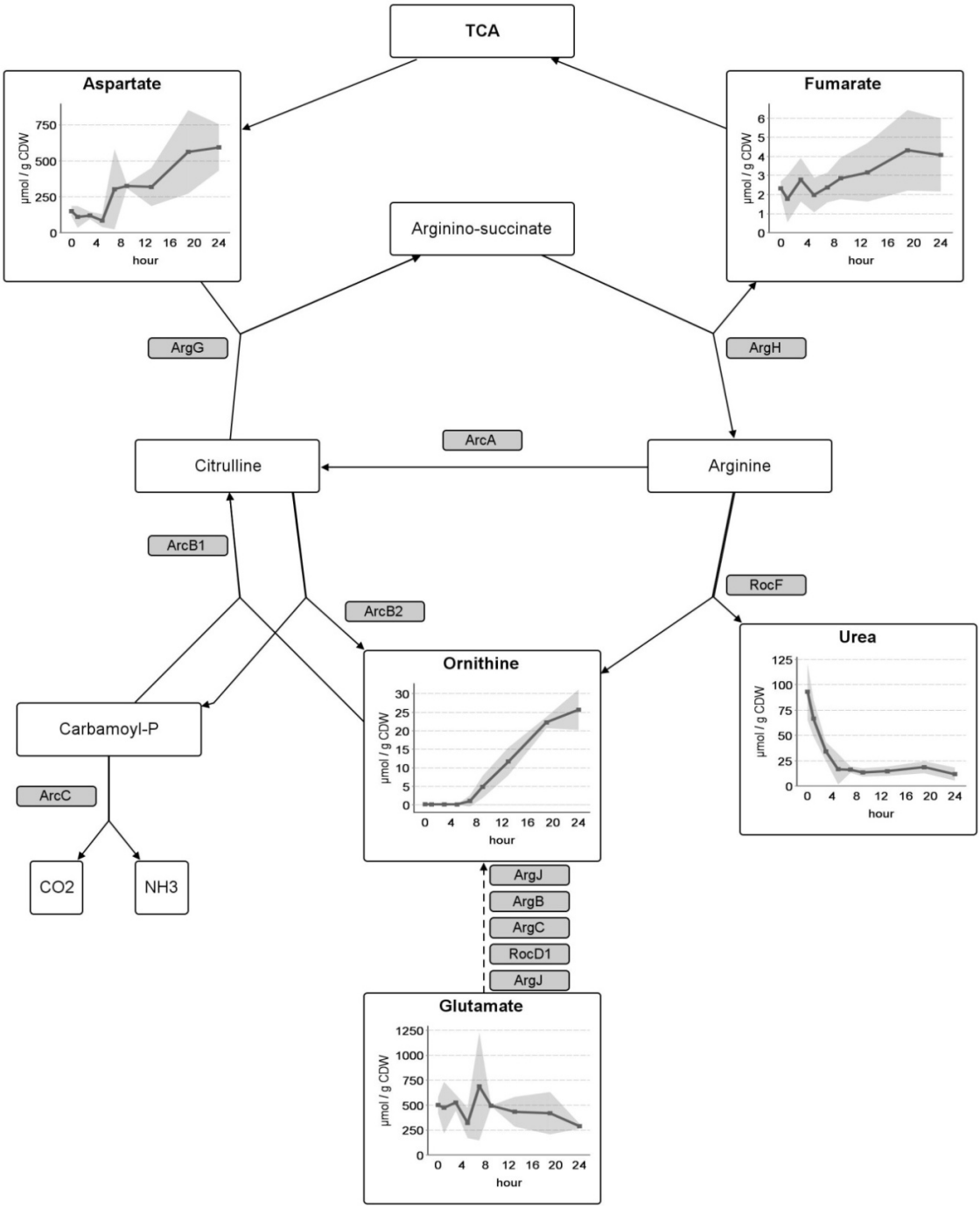


Figure S4

Arginine and urea metabolism in *S. aureus*. Absolute quantified metabolite concentrations are shown for identified metabolites in [µmol/g CDW], see **tab.1**. The grey shadow represents the standard deviation of three biological replicates. CDW = cell dry weight.



Supplemental tables:

All supplemental tables indicated here are completely listed in a separately attached excel spreadsheet.

Table S1

Gene distance matrix results for analyzed metabolites and proteins based on Pearson correlation. High similarity between two variables is shown by orange colored boxes (higher 0.9 correlation). A high inverse similarity between variables is shown by blue colored boxes (lower -0.9 correlation). Distance Metric: Pearson Correlation.

Table S2a

Extracellular metabolite concentrations [mM] for CDM and nine sample time points (see fig.2 for growth curve) determined with 1H-NMR analysis. Data represent mean values of 3 biological replicates. CDM = chemical defined medium, SD = standard deviation.

Table S2b

Relative intracellular metabolite values for nine sample time points (see fig.2 for growth curve), quantified and normalized to internal standard and CDW. Metabolites were determined with LC-MS analysis. Data represent mean values of 3 biological replicates. CDW = cell dry weight, SD = standard deviation.

* Metabolites were only identified by exact masses and not by analysis of standard compounds.

Table S2c

Intracellular metabolite concentrations for nine sample time points (see fig.2 for growth curve), absolute quantified and normalized to CDW [µmol / g CDW]. Metabolites were determined with GC-MS and LC-MS measurements. Data represent mean values of 3 biological replicates. CDW = cell dry weight, SD = standard deviation.

Table S3

Selected fold changes in abundance of identified proteins. Given are the the ratios of the relative spot volumes and the respective standard deviations (SD). Reference value is the relative spot intensity at time point t0 (at optical density = 0.5). Ratios are average values of three independent experiments

(proteins with available data from two experiments are labeled with *), proteins with significant changes (ANOVA) are marked with an “x”.

Table S4

In the manuscript used abbreviations and names of proteins A) and metabolites B).