Supplementary methods.

Metabolite Measure

Choline, TMA, TMAO, betaine and carnitine were assayed in serum samples using liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS). Serum samples were prepared for metabolomics analysis by adding 400 μL of ice-cold HPLC-grade methanol to 100 μL of serum to extract metabolites and precipitate protein. Samples were thereafter vortexed for 10 minutes and centrifuged at 14,000 *g* x 10 minutes at 4°C. The supernatants were transferred to new vials and dried *in vacuo* using a Thermo Savant vacuum concentrator at 35°C. Once dry, the samples were resuspended in 60:40 acetonitrile:water and transferred to LC-MS vials containing 200 μl glass inserts. All samples were kept at 4°C in the autosampler compartment until 2 μL was injected for analysis. LC-MS based metabolomics analysis was performed using an Agilent 1290 Infinity UPLC coupled to an Agilent 6495 Triple Quadrupole (QQQ) mass spectrometer. Chromatographic separation of metabolites was achieved using a Millipore (SeQuant) Zic-HILIC 2.1x100mm 3μm column maintained at 40°C using a flow rate of 0.5mL/min. Compounds were eluted via a 30 minute gradient starting using mobile phases A (water with 0.1% formic acid and 10mM ammonium formate) and B (90:10 acetonitrile:water with 0.1% formic acid and 10mM ammonium acetate) running at 100% B from 0 to 1.00 minutes, 100% to 90% B from 1.00 to 5.00 minutes, 90% to 55% B from 5.00 to 10.00 minutes, 55% B from 10.00 to 15.00 minutes, 55% to 100% B from 15.00 to 19.00 minutes, and 100% B from 19.00 to 30.00 minutes. An Agilent QQQ mass spectrometer was operated in positive ion mode using a heated electrospray ionization source with a gas temperature of 200°C, a gas flow rate of 14 units, a nebulizer gas flow rate of 20 units, a sheath gas temperature of 300°C, a sheath gas flow rate of 11 units, a spray voltage of 1.5kV and capillary voltage 3.0kV. Tandem mass spectra were collected using a dwell time of 200 ms, fragmentation voltage of 380V and a cell accessory voltage of 5V. The MRM transitions monitored

were carnitine 162.1 → 85.2 at a collision energy of 25V, betaine 118 → 58.2 at a collision energy of 30V, choline 104.1 → 60.1 at a collision energy of 27V, TMAO 76 → 42 at a collision energy of 50V, and TMA 60 → 60 at a collision energy of 5V. Acquired data was analyzed using Agilent Masshunter Quantitative Analysis (version B.07.01/7.1.524.0). Compounds were identified and quantified using retention time and MRM transitions from a commercial standard for each target analyte. Concentrations are provided as ng/ml.

Data Analysis

The data, consisting of 38 patient samples measured across 5 choline metabolites and 14 clinical outcomes were processed using R, Version 3.4.1. (www.r-project.org). Continuous variables were expressed as mean ± standard deviation (SD) and the categorical variables as percentage in tables. Pearson correlation was used to check correlation between disease activity scores. T-test was used for the comparison of continuous variables and the comparisons were then adjusted for age, gender and disease activity, by including the latter as covariates in a logistic regression model. Hierarchically clustered heatmaps were generated for correlations between choline metabolites and clinical outcomes. Further, linear regression was performed between each choline metabolite- clinical outcome pair, controlling for patient age, gender and disease activity as covariates in the model. Normally distributed independent variables were standardized so they had mean 0 and standard deviation 1.