

Supplemental Table S1: Age and sex determinants of patients who received testosterone replacement therapy (TRT) and their matched controls. Cohort 1, Vitalant Pittsburgh. Cohort 2, Vitalant Denver. p-values were obtained by unpaired t-test.

	Cohort 1			Cohort 2		
	TRT	Control	p value	TRT	Control	p value
Age (mean±SD)	44±3.7	44±1.5	0.896	53±9.8	57±13.1	0.718
RBC count (M/μL) in RBC unit (mean±SD)	6.9±0.3	6.2±0.3	0.07	7.6±0.7	7.0±02	0.218
Hemoglobin (g/dL) in RBC unit (mean±SD)	17.7±1.9	15.6±1.7	0.166	16.5±2.1	15.7±2.5	0.666
Males (n)	5	3		2	2	
Females (n)	0	0		1	1	

Supplemental Table S2 sheet 2: Cohort 1

The image displays a large heatmap visualization of data for Cohort 1. The table is organized into columns and rows, with various cells highlighted in different colors (yellow, blue, red, green, purple, orange, brown, pink, grey, light blue, light green). The left side of the table contains a dense list of gene names, and the right side contains numerical data values. The heatmap shows a complex pattern of high and low values across the different genes and samples, with distinct clusters of high values in yellow and green, and low values in blue and purple.

Supplemental Table S2 sheet 3. Cohort 2

The image displays a large, complex data table, likely a genomic or biological dataset, organized into multiple columns and rows. The table is characterized by several distinct horizontal bands of color, including yellow, blue, red, green, and purple, which likely represent different categories or clusters of data. The top section of the table is dominated by these colored bands, while the bottom section consists of a dense grid of data points, possibly representing individual samples or measurements. The table is structured with a header row at the top, followed by numerous rows of data, and a final section at the bottom containing a detailed list of identifiers or labels. The overall layout is highly structured and detailed, typical of a large-scale data analysis output.

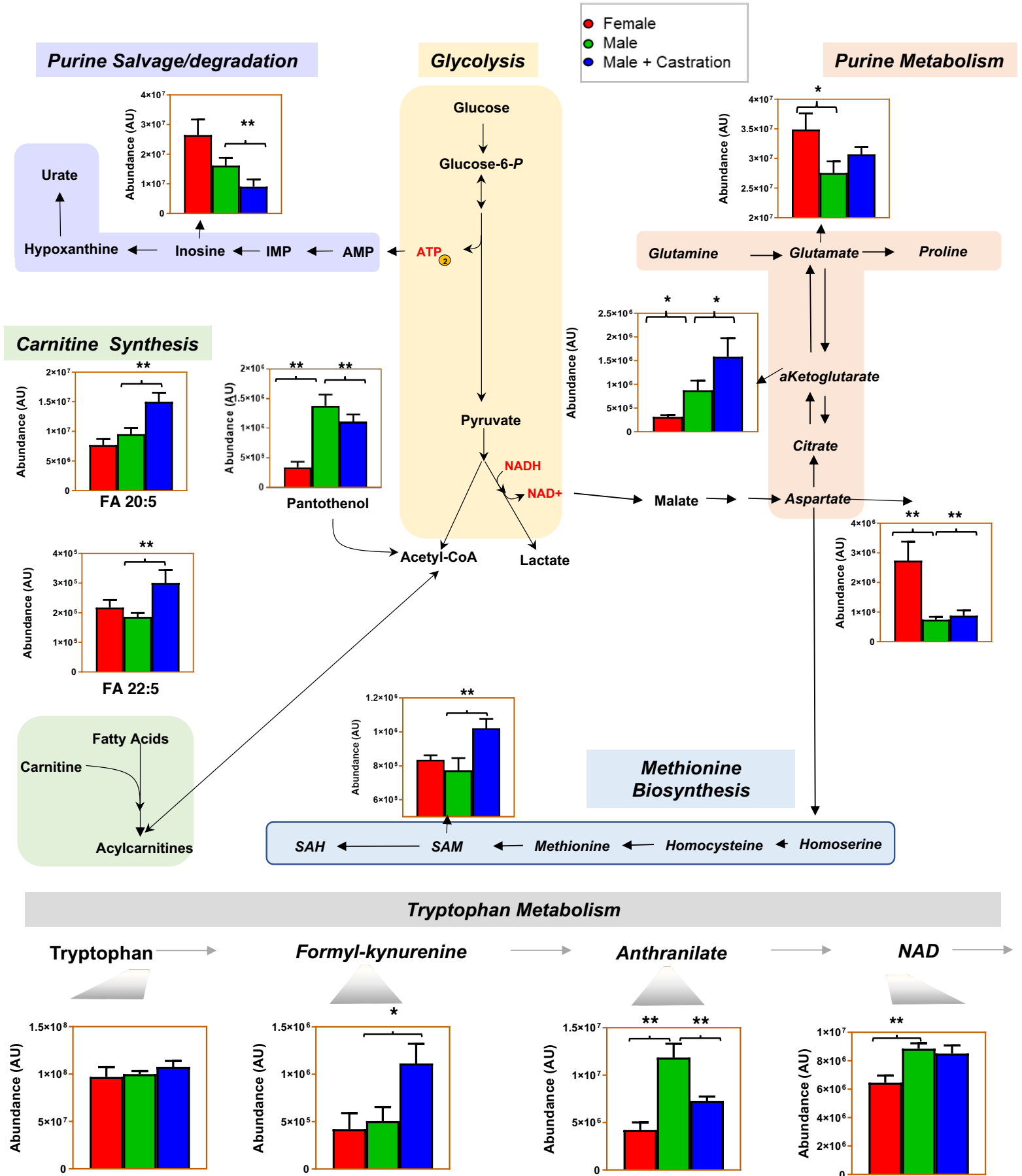
Supplementary Table S3: Evaluation of sex and testosterone-dependent differences in red blood cell metabolites. Numbers represent p values from t-test analyses between testosterone replacement therapy (TRT) and control subjects or between castrated males (Cas) and intact males or males (Cas or intact) and female mice. Highlighted are significant ($p < 0.05$) metabolites common to humans and Cas/intact male mice.

	Human TRT / Controls	Mice Cas male/Male	Mice Cas male / Female	Mice Male / Female
L-alanine	0.004	0.061	0.258	0.019
L-tyrosine	0.080	0.067	0.035	0.037
S-Glutathionyl-L-cysteine	0.047	0.006	0.316	0.010
Dehydroascorbate	0.015	0.095	0.028	0.397
S-Adenosyl-L-methionine	0.019	0.015	0.010	0.247
UDP-N-acetyl-D-glucosamine	0.002	0.047	0.001	0.495
CMP-N-acetylneuraminic acid	0.021	0.082	0.463	0.048
Creatinine	0.034	0.020	0.017	0.128
5-Hydroxyindoleacetate	0.049	0.166	0.044	0.307
Trihydroxycyclohexane-dione	0.009	0.212	0.003	0.004
3-Oxalomalate	0.050	0.083	0.435	0.018
N6-Methyl-L-lysine	0.035	0.047	0.202	0.040
UDP-glucose	0.360	0.414	0.005	0.015
Lactate	0.491	0.270	0.022	0.021
S-Adenosyl-L-homocysteine	0.229	0.346	0.020	0.034
Methenyltetrahydrofolate	0.224	0.336	0.023	0.035
N-Glycoloyl-neuraminic acid	0.124	0.001	0.037	0.015
Anthranilate	0.206	0.003	0.005	0.002
Picolinic acid	0.227	0.221	0.014	0.001
Acetylcholine	0.413	0.160	0.002	0.065
Pyridoxamine	0.462	0.029	0.383	0.040
Oxalosuccinate	0.333	0.037	0.222	0.085
Oxo-methylthioheptanoic acid	0.484	0.125	0.227	0.042
L-Carnitine	0.207	0.162	0.071	0.084
butanoyl-L-carnitine (acyl-C4)	0.048	0.341	0.105	0.121
acyl-C4-DC	0.123	0.003	0.222	0.008
acyl-C5-OH	0.112	0.059	0.442	0.083
acyl-C6	0.128	0.488	0.029	0.265
(acyl-C8)	0.287	0.172	0.016	0.382
(acyl-C8:1)	0.339	0.418	0.179	0.271
(acyl-C12:1)	0.117	0.000	0.041	0.007
(acyl-C16)	0.029	0.286	0.312	0.239
Hexanoic acid (caproate)	0.441	0.208	0.193	0.027
Heptanoic acid	0.079	0.137	0.373	0.163
Octanoic acid (caprylic acid)	0.187	0.162	0.088	0.279
Nonanoic acid (pelargonate)	0.244	0.421	0.178	0.200
Tetradecenoic acid	0.070	0.350	0.205	0.258
Hexadecenoic acid	0.242	0.119	0.116	0.272
Octadecenoic acid	0.212	0.055	0.186	0.105
Linoleate	0.419	0.063	0.106	0.035
Octadecatrienoic acid	0.498	0.069	0.098	0.027
Eicosatetraenoic acid	0.405	0.042	0.345	0.027
Eicosapentaenoic acid	0.231	0.004	0.010	0.125
Docosahexaenoic acid	0.058	0.443	0.127	0.216
Dodecanedioic acid	0.252	0.266	0.033	0.357
Icosatrienoic acid	0.239	0.272	0.137	0.239
Icosapentaenoic acid	0.231	0.004	0.010	0.125
Docosa-pentaenoic acid	0.432	0.094	0.023	0.135

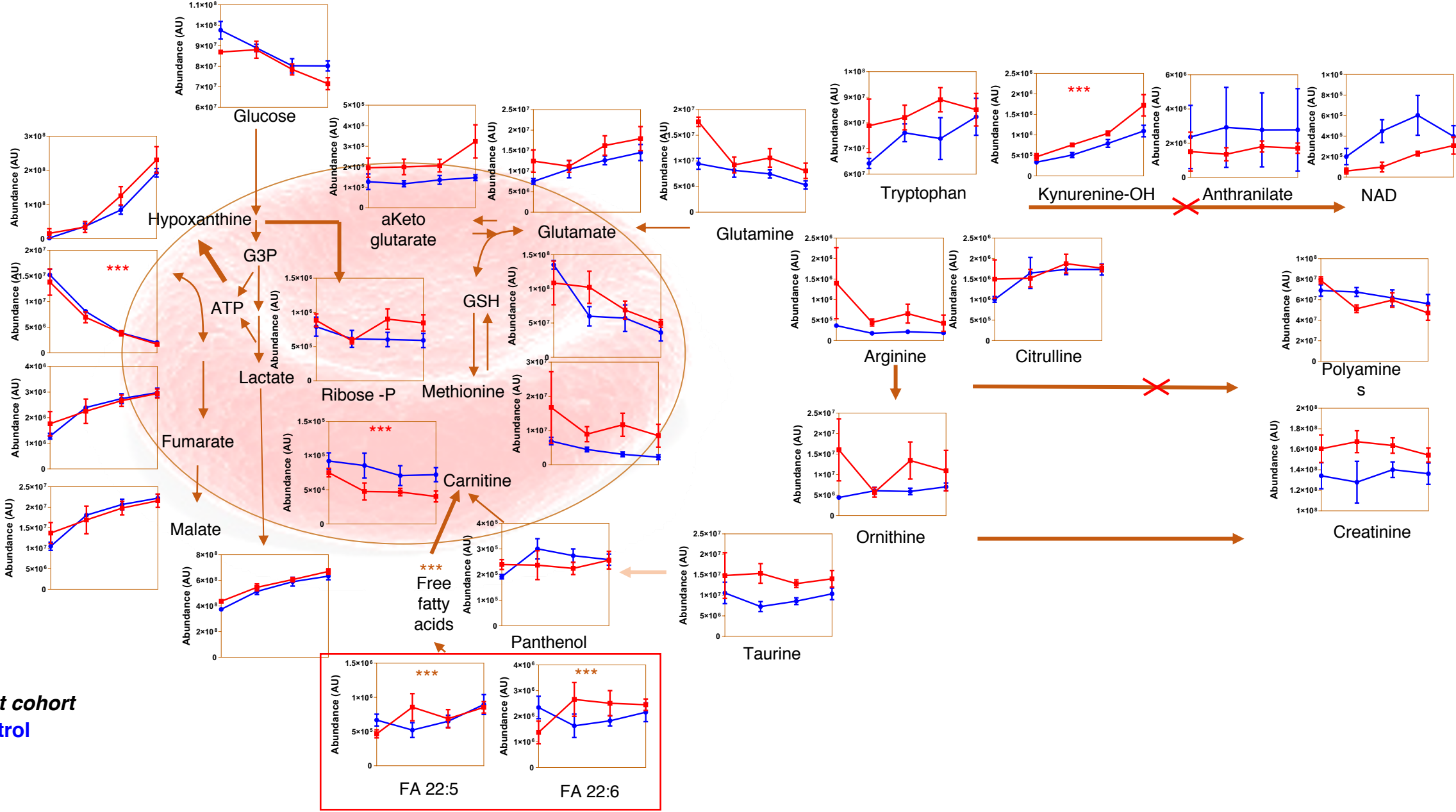
Supplementary Table S4: Multivariate linear regression was performed to test the effect of testosterone replacement therapy (TRT) for all three hemolysis measures at each time point of RBC cold storage. Testosterone intake (TRT) and donation site (cohort) were included as covariates in the model. Since only cohort 2 had data at storage week 1, this time point was excluded from the regression analysis. TRT was significantly ($p < 0.05$) associated with increased osmotic hemolysis except at week 4. This model also identified cohort as a significant modifier of hemolysis.

Hemolysis	Storage week	p-value of TRT
Spontaneous (cold storage)	2	0.100
	3	0.250
	4	0.119
	5	0.064
	6	0.058
Osmotic	2	0.010*
	3	0.011*
	4	0.181
	5	0.022*
	6	0.003*
Oxidative	2	0.211
	3	0.211
	4	0.233
	5	0.583
	6	0.271

Supplementary Figure S1: Bar graphs displaying alterations in RBC metabolic pathways from intact male, female and orchietomized FVB/NJ mice. Figure highlights metabolic pathways altered by intact males versus females, and intact males versus orchietomized mice. Bar plots (mean \pm SEM) display most impacted metabolites along related pathways. Significant (*, $p < 0.05$, **, $p < 0.01$) metabolite changes were determined by two-tailed t-test.



Supplementary Figure S2. An overview of the major red blood cell metabolic pathways, which were modified by storage duration (six weeks) and/or by testosterone replacement therapy (TRT). Line plots represent Mean±SEM in each storage week. n=5 TRT donors (red curves) and n=3 matched controls (blue curves). *, p<0.05 by two-way ANOVA.



Supplementary Figure S3. An overview of the major red blood cell metabolic pathways, which were modified by storage duration (six weeks) and/or by testosterone replacement therapy (TRT). Line plots represent Mean±SEM in each storage week. n=3 TRT donors (red curves) and n=3 matched controls (blue curves). *, p<0.05 by two-way ANOVA.

