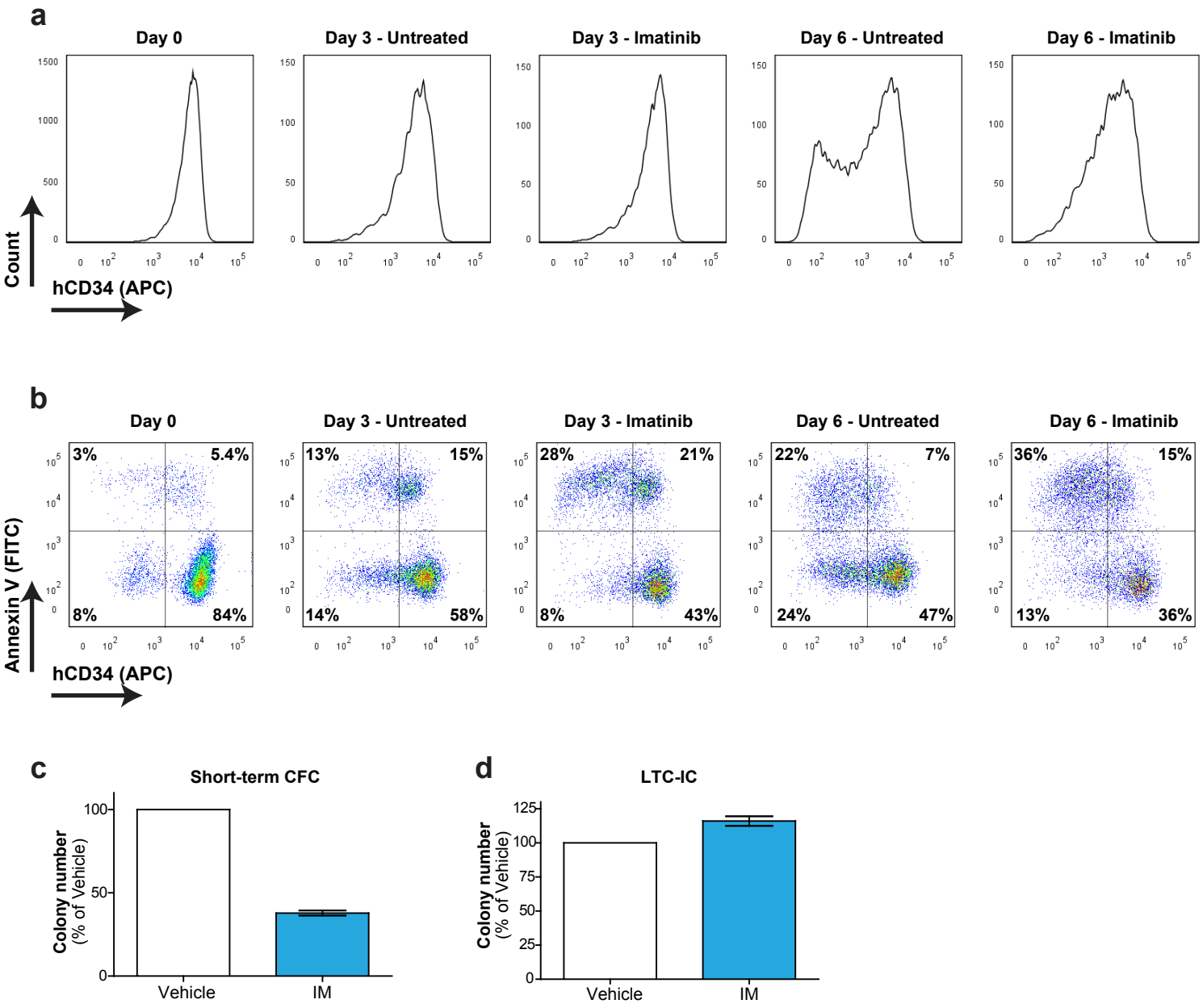
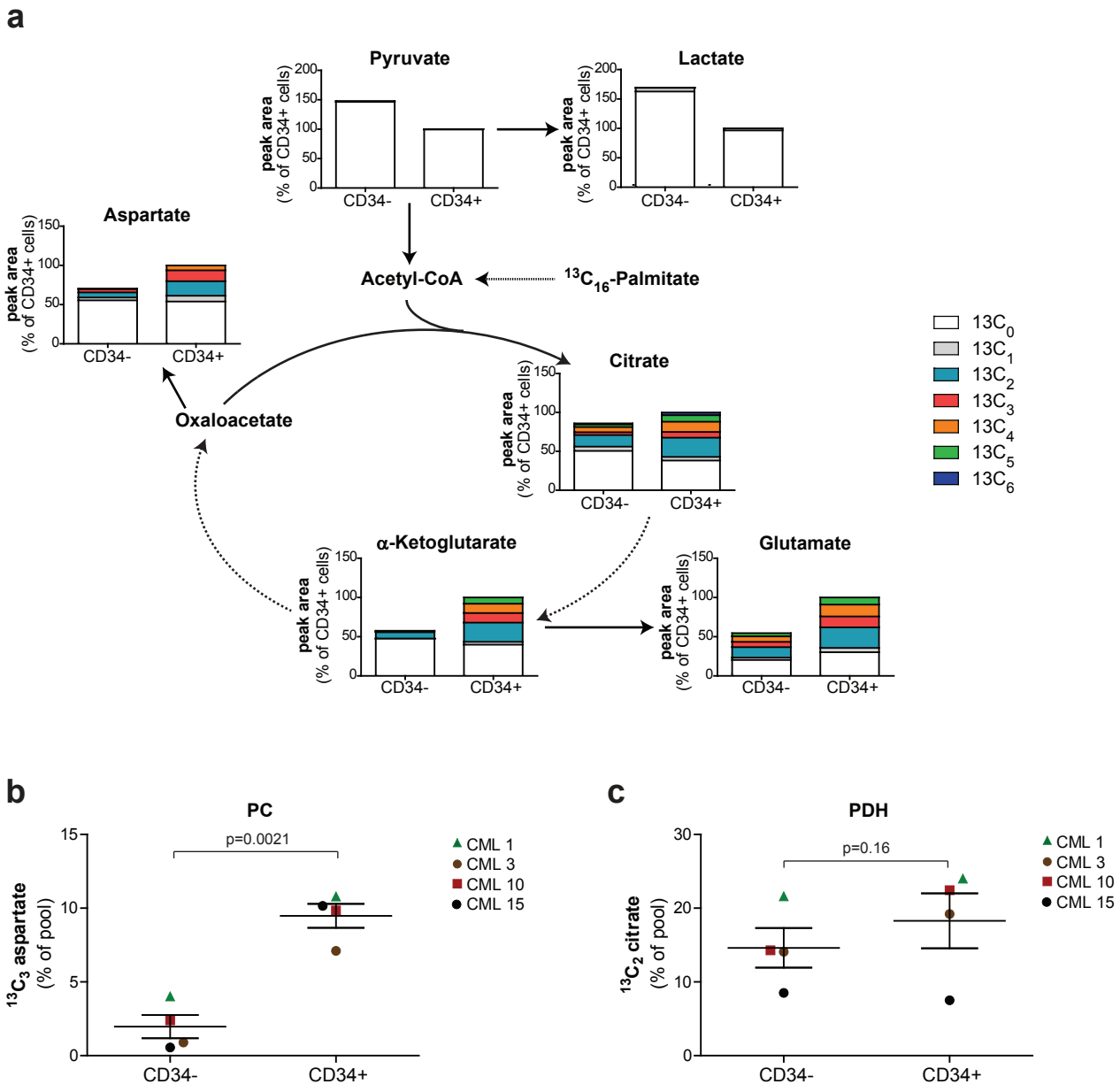
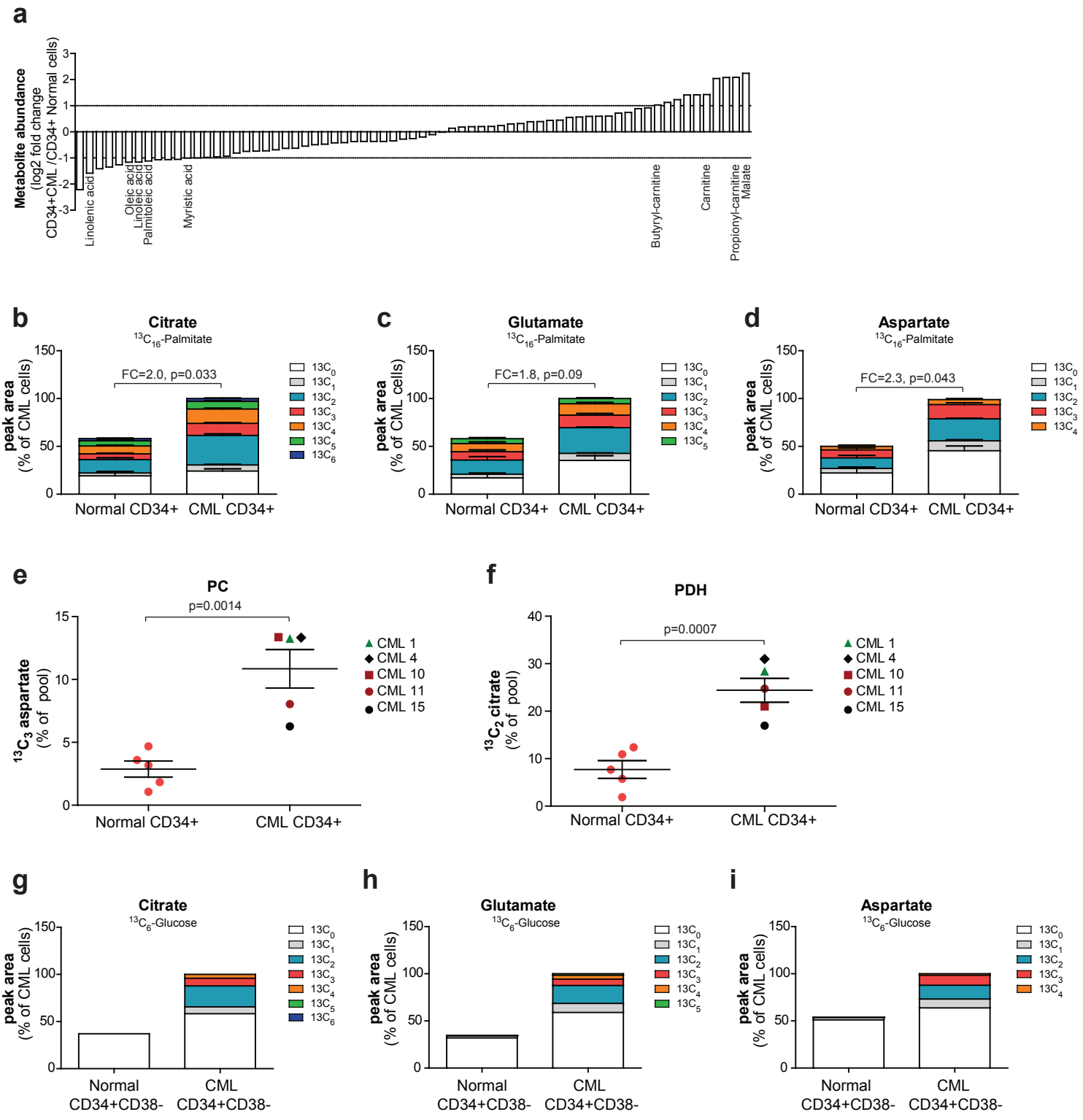


Supplementary Figure 1: Imatinib fails to eliminate LSCs.
(**a**) CD34 expression following *in vitro* culture of CD34$^+$ CML cells with imatinib (2 µM). (**b**) CD34 expression and cell death following *in vitro* culture of CD34$^+$ CML cells with imatinib (2 µM). (**c**) Number of colonies measured by CFC assay following 72 hours (h) drug treatment of CD34$^+$ CML cells with imatinib (2 µM). Mean ± S.E.M. n=4 patient samples. (**d**) Number of colonies measured by LTC-IC assay following single drug treatment of CD34$^+$ CML cells with imatinib (2 µM). Mean ± S.E.M. n=3 patient samples.



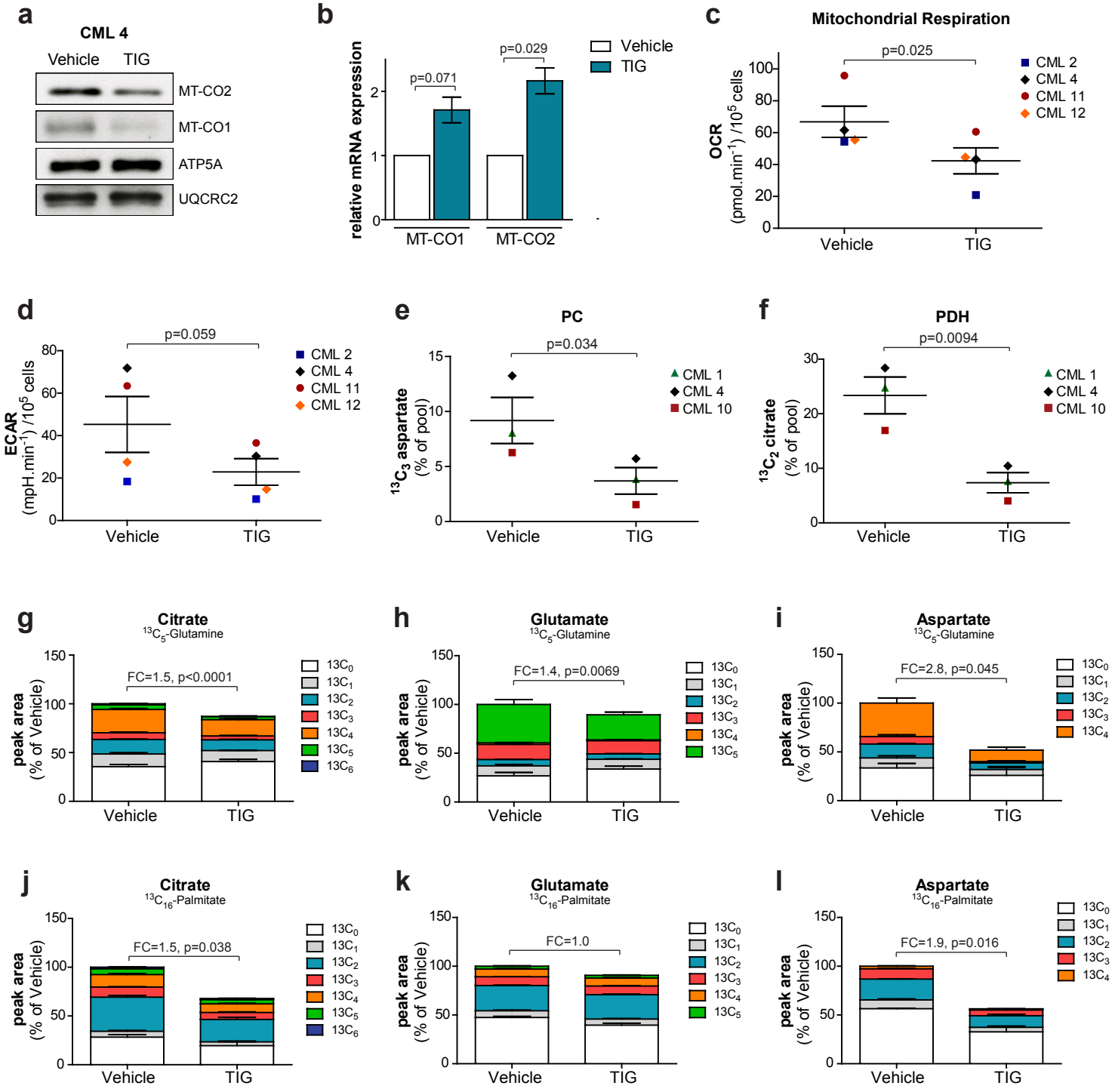
Supplementary Figure 2: Enhanced fatty acid oxidation in primitive CML cells compared to differentiated CML cells.

(**a**) Relative isotopologue distribution of indicated metabolites in CD34$^-$ and CD34$^+$ CML cells measured by LC-MS following 24 h incubation with $^{13}C_{16}$-labeled palmitate. n=1 patient sample. (**b-c**) Relative abundance of (**b**) $^{13}C_3$-aspartate and (**c**) $^{13}C_2$-citrate in CD34$^-$ and CD34$^+$ CML cells following 24 h incubation with $^{13}C_6$-labeled glucose. Mean ± S.E.M. n=4 patient samples. P-values were calculated by paired Student's t-test.

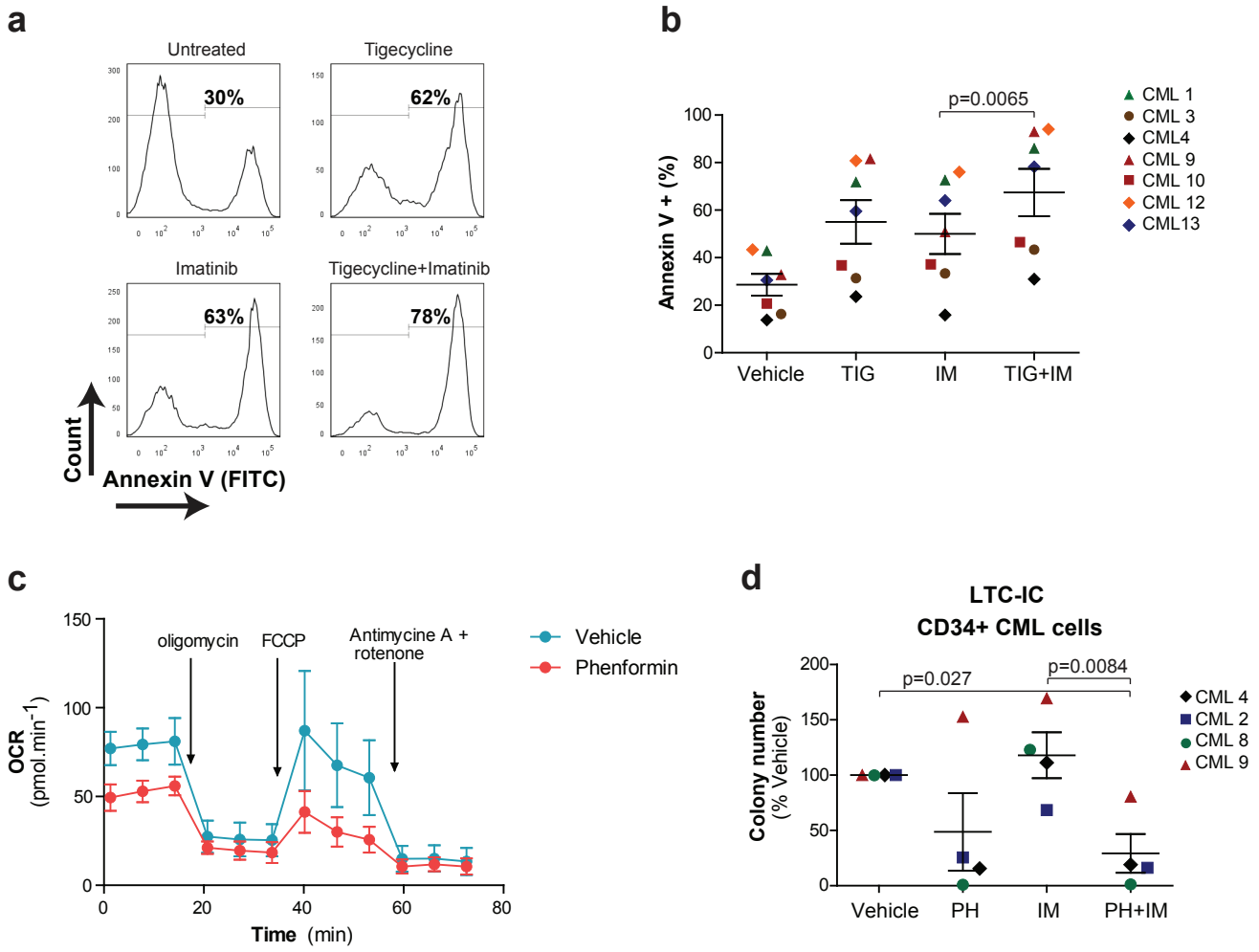


Supplementary Figure 3: Enhanced mitochondrial metabolic activity in primitive CML cells compared to normal undifferentiated hematopoietic cells.

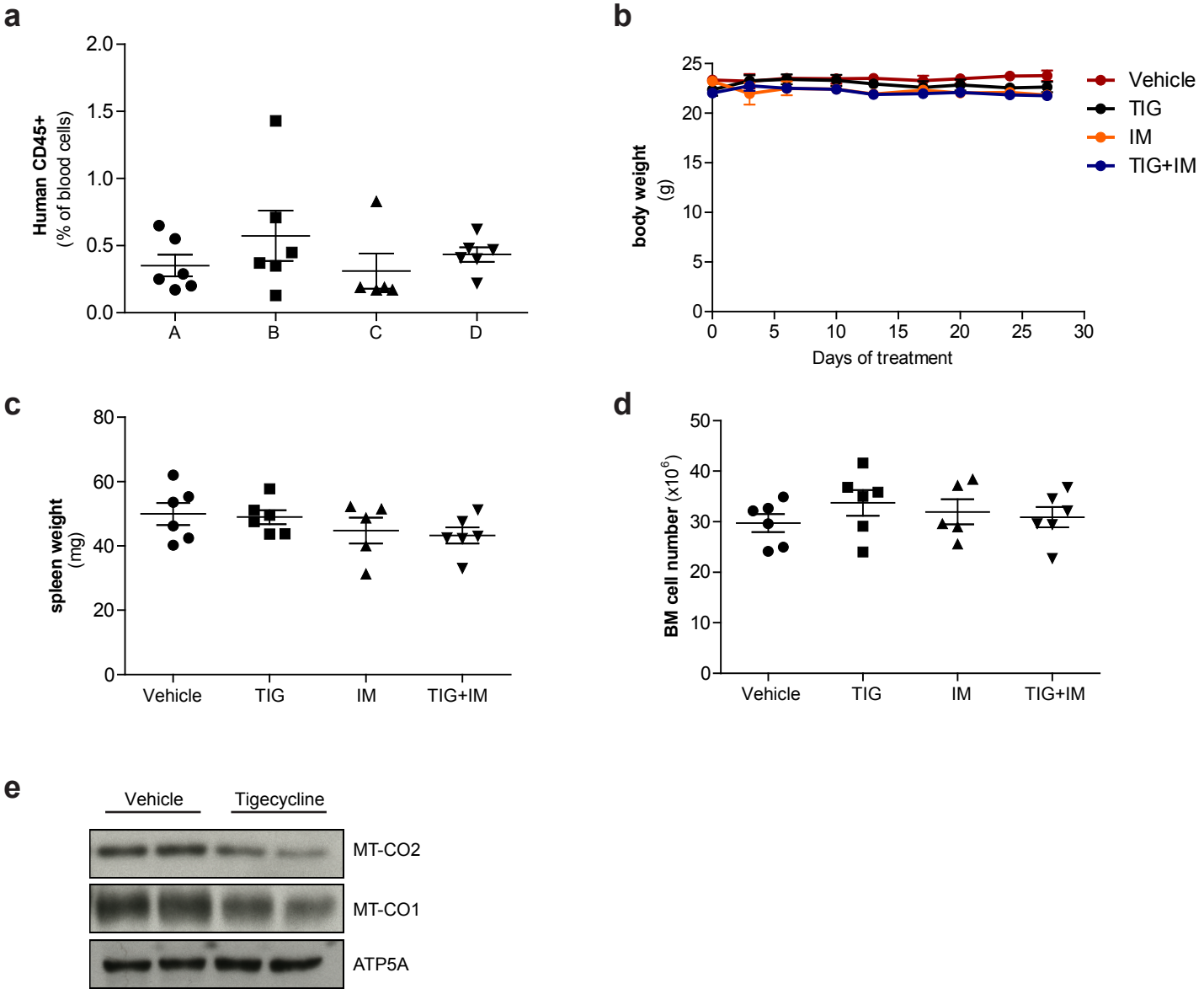
(**a**) Comparative steady-state metabolomics analysis of CD34$^+$ CML and CD34$^+$ normal cells measured by LC-MS. n=4 patient samples. (**b-d**) Relative isotopologue distribution of (**b**) citrate, (**c**) glutamate and (**d**) aspartate in CD34$^+$ CML and CD34$^+$ normal cells measured by LC-MS following 24 h incubation with $^{13}C_{16}$-labeled palmitate. Mean ± S.E.M. n=3 patient and normal samples. FC, Fold change of palmitate-derived ($^{13}C_{\geq 2}$) metabolite abundance relative to CD34$^+$ normal cells. (**e-f**) Relative abundance (**e**) $^{13}C_3$-aspartate and (**f**) $^{13}C_2$-citrate in CD34$^+$ CML and CD34$^+$ normal cells following 24 h incubation with $^{13}C_6$-labeled glucose. n=5 patient and normal samples. (**g-i**) Relative isotopologue distribution of (**g**) citrate, (**h**) glutamate and (**i**) aspartate in CD34$^+$CD38$^-$ CML and CD34$^+$CD38$^-$ normal cells measured by LC-MS following 24 h incubation with $^{13}C_6$-labeled glucose. n=1 patient sample. P-values were calculated by unpaired Student's t-test.



Supplementary Figure 4: Tigecycline inhibits oxidative metabolism in CD34$^+$ CML cells.
(**a**) Protein expression in CD34$^+$ CML cells following 72 h *in vitro* treatment with tigecycline (2.5 µM). n=1 patient sample (**b**) mRNA levels following 72 h i*n vitro* treatment with tigecycline (2.5 µM). n=3 patient samples. (**c**) Basal mitochondrial OCR in CD34$^+$ CML cells following treatment with tigecycline (2.5 µM). n=4 patient samples. (**d**) Basal Extracellular Acidification Rate (ECAR) in CD34$^+$ CML cells following treatment with tigecycline (2.5 µM). n=4 patient samples. (**e-f**) Relative abundance of (**e**) $^{13}C_3$-aspartate and (**f**) $^{13}C_2$-citrate in CD34$^+$ CML cells following 24 h incubation with $^{13}C_6$-labeled glucose in presence or absence of tigecycline (2.5 µM). n=3 patient samples. (**g-i**) Relative isotopologue distribution of (**g**) citrate, (**h**) glutamate and (**i**) aspartate in CD34$^+$ CML cells measured by LC-MS following 24 h incubation with $^{13}C_5$-labeled glutamine in presence or absence of tigecycline (2.5 µM). n=3 patient samples. (**j-l**) Relative isotopologue distribution of (**j**) citrate, (**k**) glutamate and (**l**) aspartate in CD34$^+$ CML cells measured by LC-MS following 24 h incubation with $^{13}C_{16}$-labeled palmitate in presence or absence of tigecycline (2.5 µM). n=3 patient samples. P-values were calculated by paired Student's t-test. All data are represented as Mean ± S.E.M.

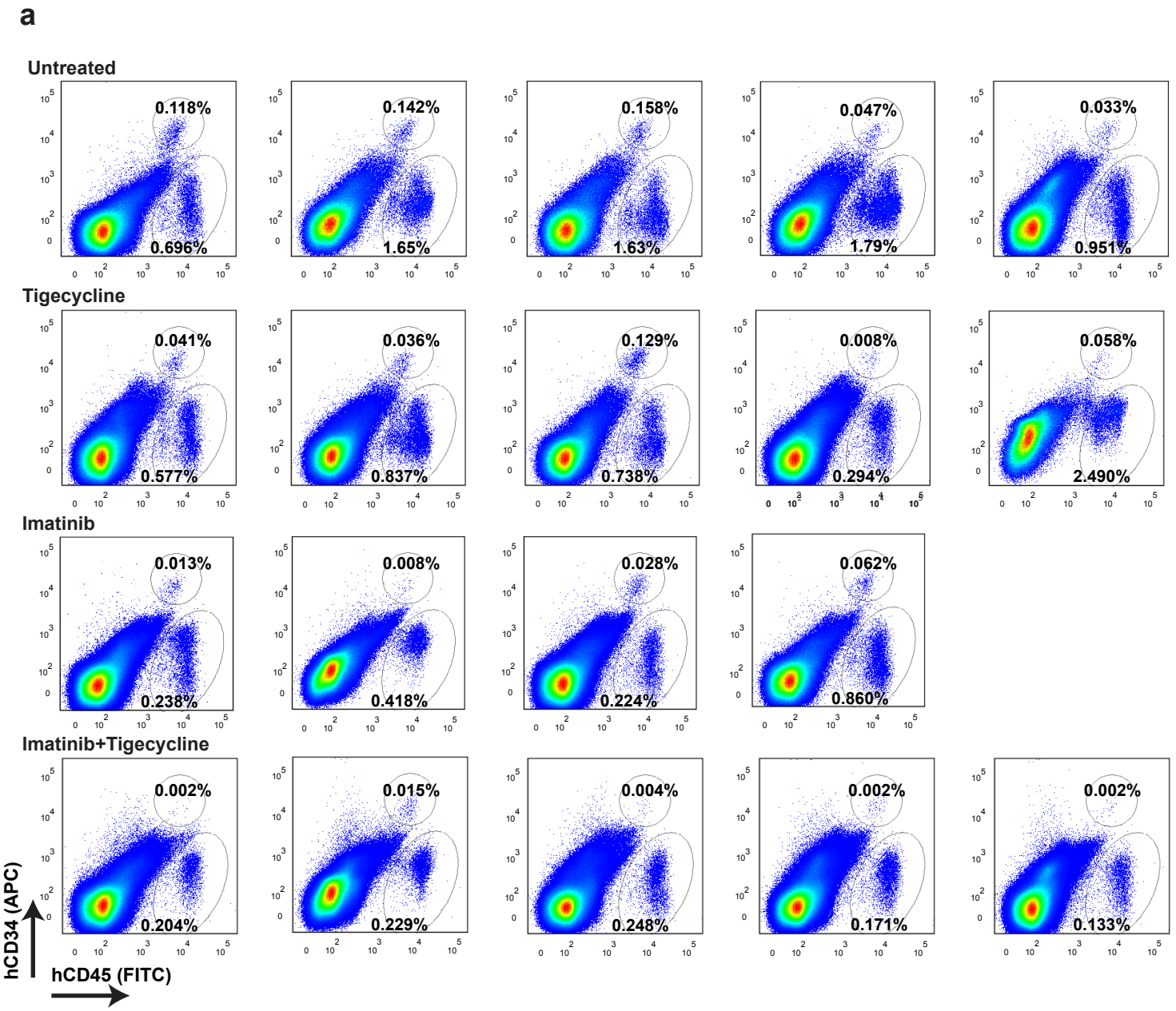


Supplementary Figure 5: Inhibition of mitochondrial activity targets primitive CML cells.
(**a**) Representative flow cytometry histograms and (**b**) percentage of Annexin V positive cells after 72 h treatment with tigecycline (2.5 µM), imatinib (2 µM) and combination (2.5 µM + 2 µM) in $CD34^+$ CML cells. Mean ± S.E.M. n=7 patient samples. (**c**) Representative respirometry output in $CD34^+$ CML following 24 h treatment with phenformin (10 µM). Mean ± S.D. n=1 patient sample. (**d**) Number of colonies measured by LTC-IC assay in $CD34^+$ CML cells. Mean ± S.E.M. n=4 patient samples. P-values were calculated by paired Student's t-test.

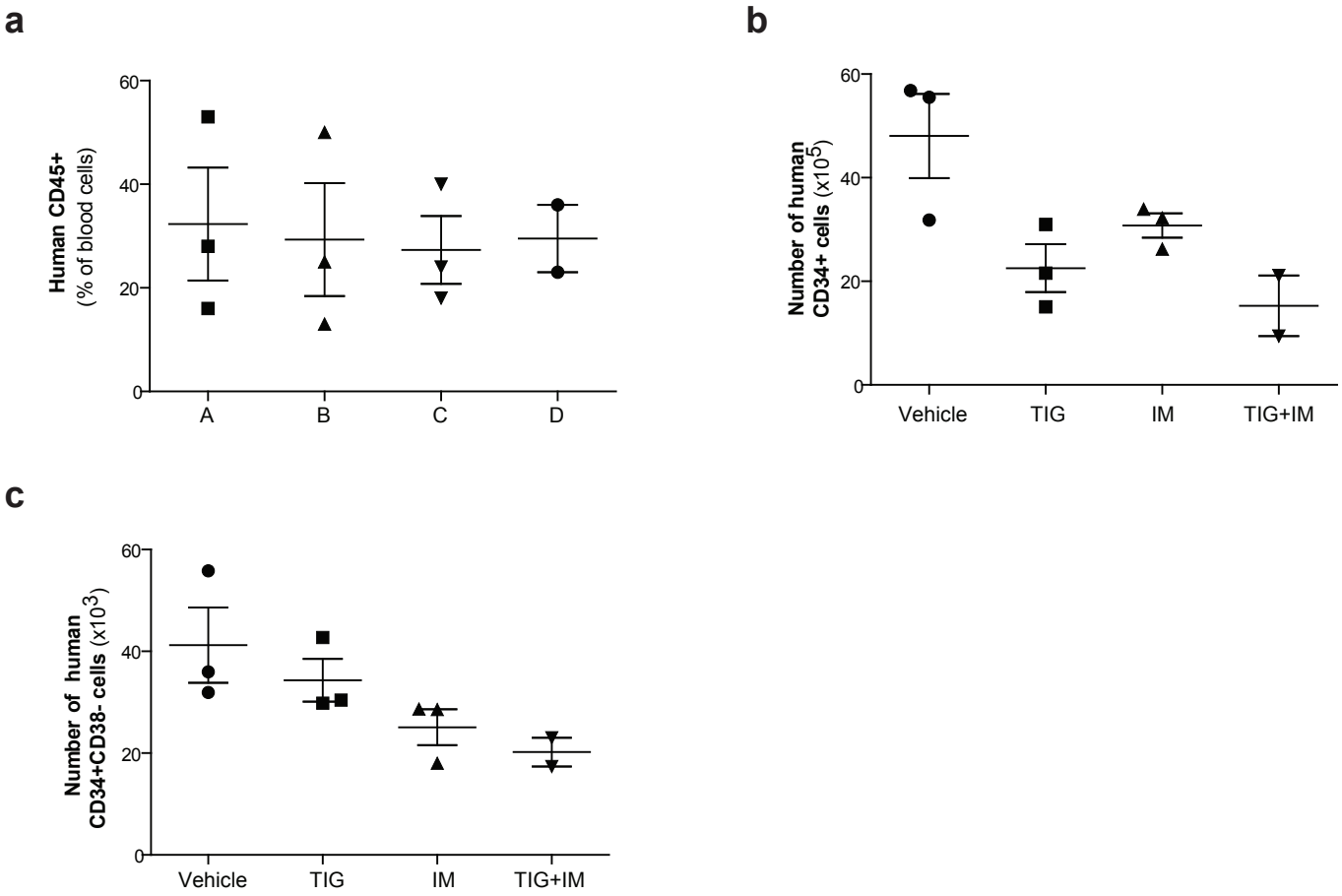


Supplementary Figure 6: *In vivo* tigecycline and/or imatinib treatment is not toxic towards normal murine cells.

(**a**) Percentage of human CD45$^+$ cells in the blood of immuno-compromised mice prior to drug treatment. Group A, B, C and D were later on treated with Vehicle, TIG, IM and TIG+IM respectively. (**b**) Change in mice body weights upon drug treatment. (**c**) Spleen weight and (**d**) bone marrow cellularity of mice engrafted with CD34$^+$ CML cells after 4 weeks of treatment with the indicated drugs. (**e**) Protein expression in FACS-sorted CD34$^+$ CML cells following 4 weeks *in vivo* drug treatment. Mean ± S.E.M.



Supplementary Figure 7: The combination of imatinib and tigecycline targets CML stem-cells *in vivo*. Mice were transplanted with CD34$^+$ CML cells and analysis of human cell engraftment was performed following 4 weeks of drug treatment. (**a**) Percentage of human CD45$^+$ and human CD34$^+$ cells from total bone marrow cells. (**b**) Number of human CD45$^+$ cells engrafted in the bone marrow of immuno-compromised mice. Mean ± S.E.M. P-values were calculated by unpaired Student's t-test on logarithmically transformed variables to meet the assumption of normality.



Supplementary Figure 8: Tigecycline marginally affects normal HSCs *in vivo*.
(**a**) The pre-treatment engraftment levels of cord blood cells in immuno-compromised mice were assessed by monitoring the percentage of human $CD45^+$ circulating leukocytes. Group A, B, C and D were later on treated with Vehicle, TIG, IM and TIG+IM respectively. (**b-c**) Number of (**b**) human $CD34^+$ and (**c**) human $CD34^+CD38^-$ cord blood cells engrafted in the bone marrow following *in vivo* drug treatment. n≥2 mice per treatment arm. TIG, tigecycline; IM, imatinib.