Supplementary Figure 1.

The phenotype of MDSC in the lung and spleen from lung cancer challenged mice. Increased MDSC infiltration following establishment of tumor burden via an intra-cardiac (i.c.) route in a murine model of lung cancer is associated with reduced survival. FACS plots showing percentages and phenotypic characterization of (a) lung and (b) spleen MDSC in tissues harvested from LLC-challenged mice at 10 days after tumor challenge. (c) Flow cytometry data for percentages of MDSC and CD8+ T cell infiltration in the tumor nodules of mice challenged with LLC via i.c. route. (**p<0.01 vs Day 3, n=11 animals/group, 3 replicate experiments). (d) Kaplan-Meier curve showing survival of mice injected with LLC via i.c. route following therapy with SODmim, Gem and SODmim + Gem. Mantel-Cox log-rank test showed (p<0.001 for Gem vs SOD mim +Gem, p<0.001 for SODmim vs Gem, p<0.0001 for SOD mim vs combination, p=0.05 PBS vs Gem, p<0.0001 for PBS vs combination (n=6 mice/group).
Reduced tumor burden in mice from Gem and SODmim+Gem therapy groups. (a) Representative FACS plots showing inhibition of MDSC infiltration in lung and spleen tissues from tumor bearing mice treated with combination therapy compared to individual treatment and untreated control groups. (b) Comparison of tumor weights between Gem and SOD mim + Gem groups at the indicated times (p<0.001) (n=3 mice/group) (c) A representative photograph depicting the tumor burden in a mouse from both the Gem and SODmim + Gem groups.
Supplementary Figure 3.

Combination therapy reduced infiltration of neutrophils but not macrophages in tumor. MDSC depletion using intraperitoneal administration of anti-Gr-1 antibody and SOD mimetic together enhanced anti-tumor immunity. In LLC tumor cells, combination therapy did not elicit chemosensitization (a) Percent neutrophils in tumor in SOD mim+Gem vs Gem (*p<0.05), Gem & SOD mim+Gem vs PBS(**p<0.01), SOD versus PBS(*p<0.05). Percent cells were determined from flow cytometry and data represent n=9 mice/group from n=3 experiments. (b) Percent MDSC in tumor following treatment with combination of SOD mim and anti-Gr-1 Ab (**p<0.01 vs controls), anti-Gr-1 Ab (**p<0.01 vs controls) or control treatments. (c) Stacked histograms showing expansion of memory CD8+ T cell populations in tumor with the indicated treatments (n=3 mice/group). (c) Percent tumor cell viability determined using MTS assay from LLC cells treated for 24 hours with indicated doses of SOD mim and then treated with Gem for 24 hours at the indicated concentrations.(**p<0.01 & *p<0.05 vs untreated, n=5 replicates/group).
Combination therapy was efficient in reducing tumor growth in catalase deficient mice by decreasing MDSC infiltration and improving the memory CD8⁺ T cell response. (a) Representative FACS plots showing MDSC levels in lung, spleen and tumor tissues from catalase deficient LLC-challenged mice after combination therapy, individual treatments and controls. (b) FACS plots showing increased T_{SCM} CD8⁺ T cell infiltration in LLC-challenged catalase deficient mice. (c) Stacked histogram plots of percentages of subsets within total CD8⁺ populations in the tumor tissues. (d) Tumor weights in catalase deficient mice.
Persistent memory CD8⁺ T cell subsets expand following re-encounter with i.v. challenged LLC cells. Circulating CD8⁺ T cell subsets in adoptive transfer recipients analyzed by flow cytometry (a) Baseline at day 93 after adoptive transfer, (b) on days 9 and 15 after rechallenge with tumor. (c) Representative FACS plots showing characterization of circulating memory subsets.
Adoptively transferred memory cells persist and reduce MDSC infiltration and tumor burden in LLC-rechallenged recipient mice. (a) Representative FACS plots of circulating Gr-1⁺ CD11b⁺ MDSC at day 15 after rechallenge with LLC in adoptive transfer recipients compared to tumor challenged controls. (b) Representative photomicrographs of adoptive transfer recipient mice showing reduced tumor burden in lung tissue compared to controls. (c) Tumor weights in adoptive transfer recipients on day 15 following LLC rechallenge.
Adoptively transferred memory cells expand upon encounter with tumor cells. Representative FACS plots of collagenase extracted CD8\(^+\) cells from tumor tissue of CD45.1 mice which were recipients of adoptive transfer of CD8\(^+\) T\(_{CM}\) and T\(_{EM}\) cell subsets purified from tumor bearing CD45.2 donors treated with combination therapy or gemcitabine therapy. CD8\(^+\) cells were gated and then analyzed for CD45.2 and CD45.1 to determine the percentage of persisting donor cells as compared to recipient CD8\(^+\) T cells.