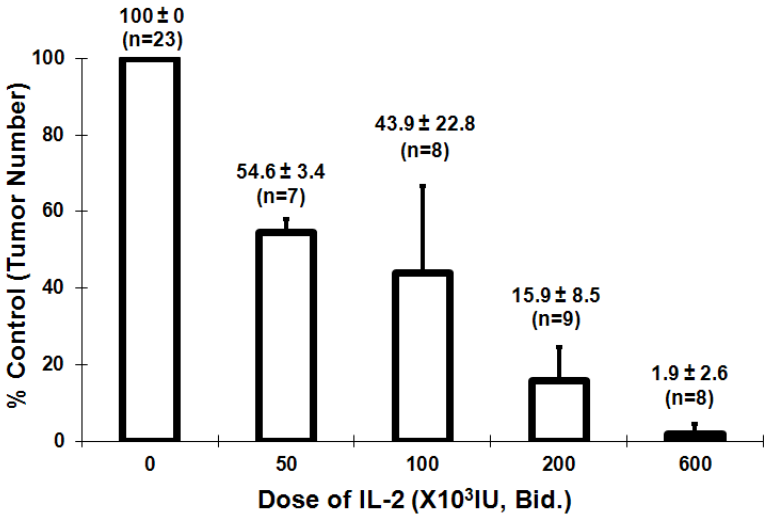
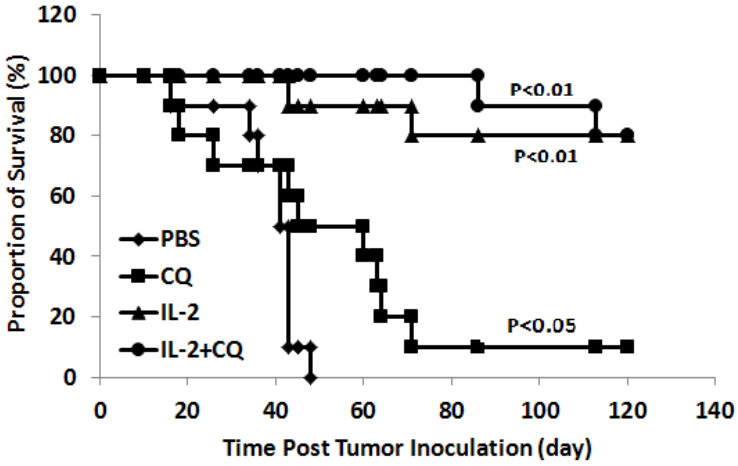
Supplemental Figure 1.



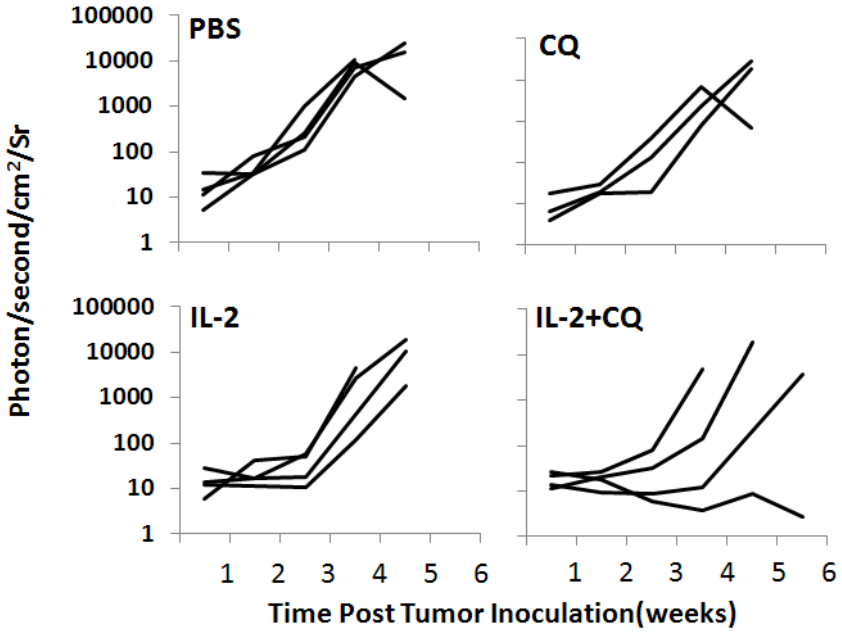
Supplemental Figure 1. Administration of rIL-2 inhibits tumor growth in murine hepatic metastasis model in a dose dependent fashion. 2X10^5 MC38 tumor cell were injected into C57BL/6 mice via portal vein injection. Mice receiving rIL-2 treatment at the concentrations indicated were sacrificed 14 days following tumor inoculation; individual tumor nodules were counted and compare to UT controls.

Supplemental figure 2.



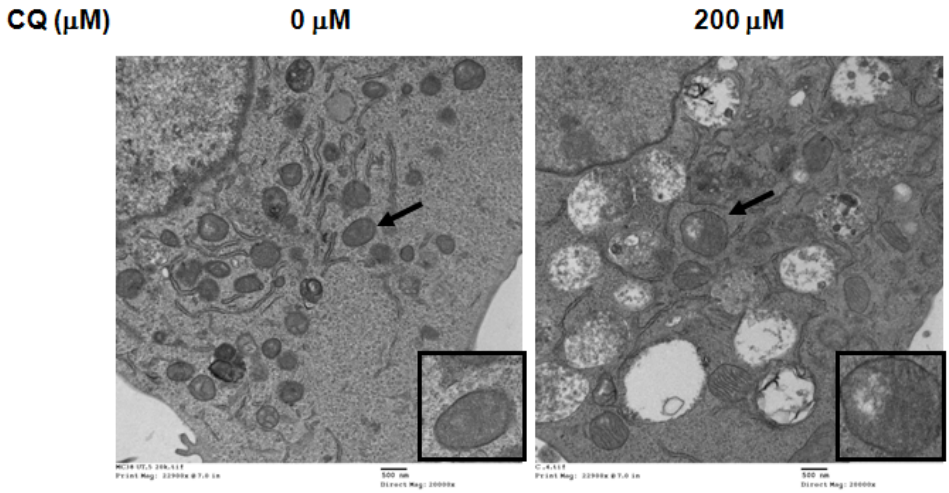
Supplemental Figure 2. HDIL-2 alone or combined with CQ significantly inhibits Panc02 tumor growth, prolonging survival time in a hepatic metastasis model. C57BL/6 mice received 1X10^6/mouse luciferase expressing Panc02 tumor cell via portal vein injection, and were randomly divided into four groups, 10 mice per group. On day 7, mice received HDIL-2 600K IU, twice a day for 5 day or combined with CQ 50mg/kg/day for 30 days. Mice receiving CQ alone or UT were used as control.

Supplemental Figure 3.



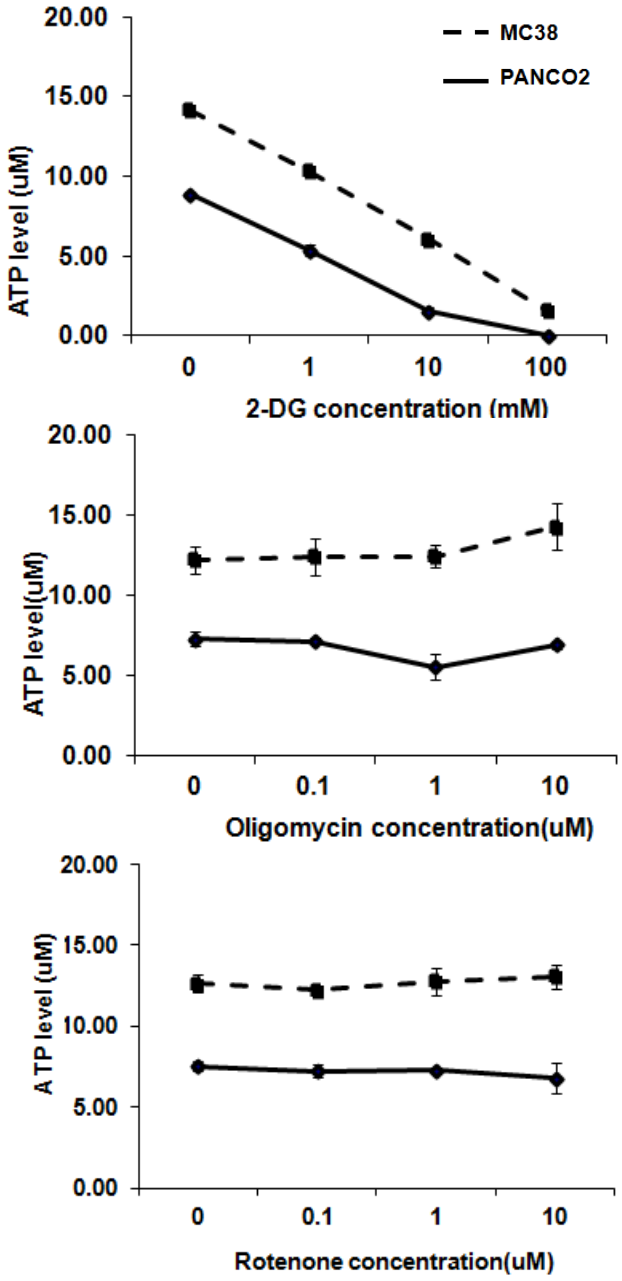
Supplemental Figure 3. **HDIL-2 combined with CQ inhibits Renca tumor growth in an hepatic metastasis model.** Balb/c mice received 2X10^5/mouse luciferase expressing Renca tumor cell via portal vein injection, and were randomly divided into four groups. On day 7, mice received HDIL-2 600K IU, twice a day for 5 day or combined with CQ 50mg/kg/day for 30 days. Mice receiving vehicle or CQ alone were used as controls. Experiment is repeated once and received similar results.

Supplemental Figure 4.



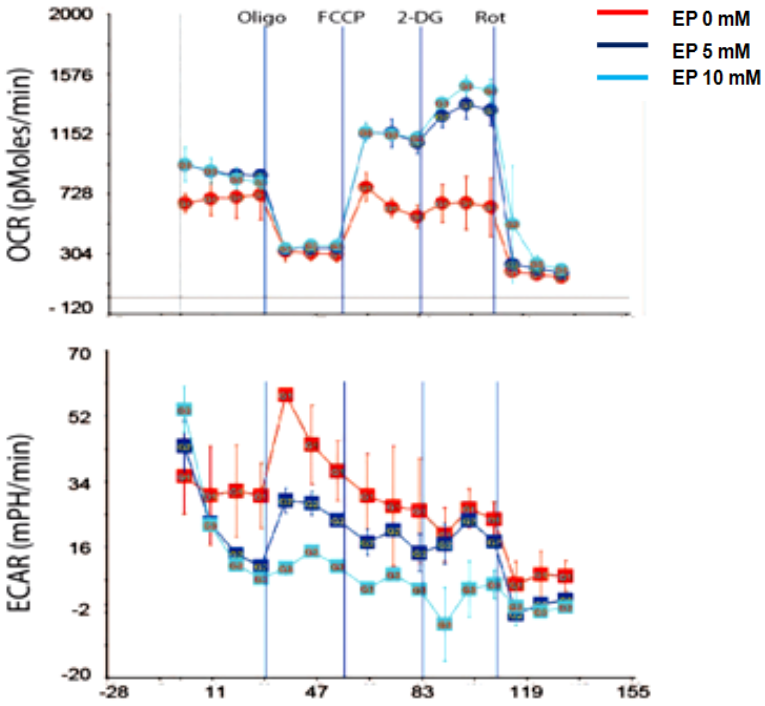
Supplemental Figure 4. CQ treatment results in mitochondrial morphologic changes in MC38 cells. MC38 cells were exposed to 200μM CQ for 4 hrs. Ultrastructure was assessed by TEM. Damaged mitochondria which exhibit disrupted membranes, absent and/or disordered mitochondrial christae (arrow) are shown.

Supplemental Figure 5.



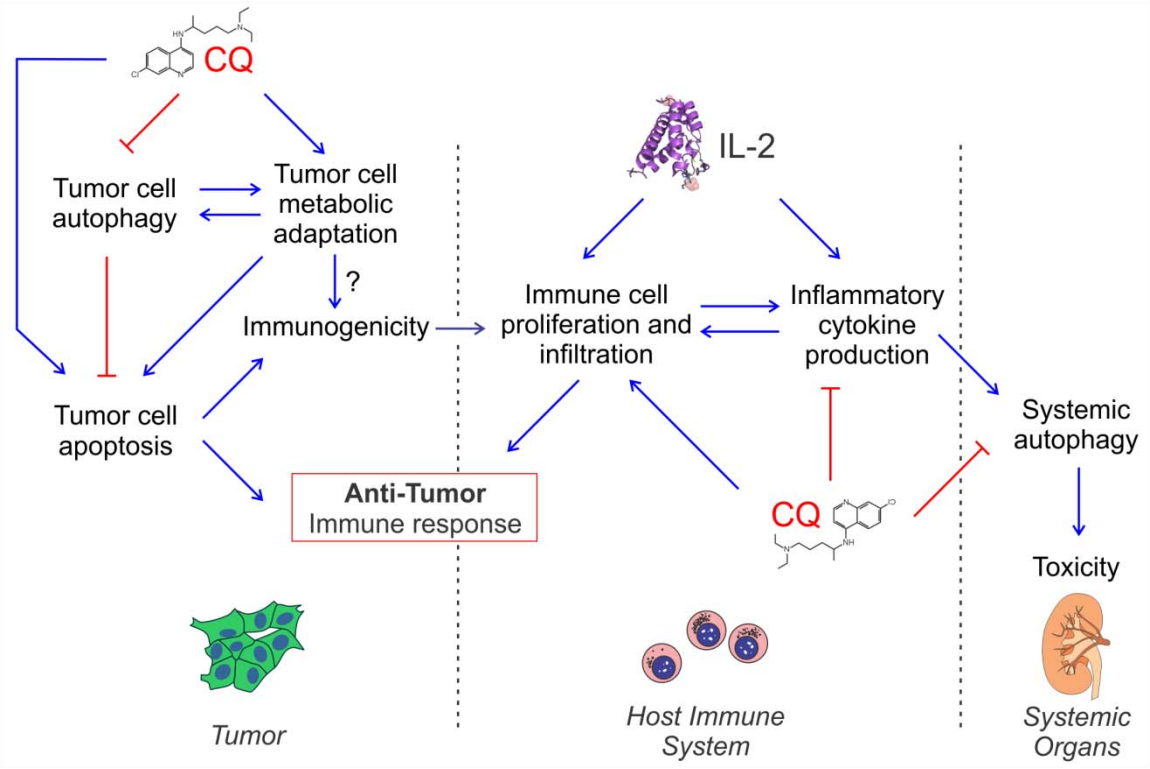
Supplemental Figure 5. Murine tumor cells are dependent on glycolysis. 2X10^4/well of MC38 and Panc02 cells were exposed to oligomycin, rotenone or 2-DG respectively at the concentration as indicated for 1 hr. ATP levels were measured by using a luminescence ATP detection assay system.

Supplemental Figure 6.



Supplemental Figure 6. Ethyl pyruvate alters tumor cell metabolism. (A). Measurement of tumor cell metabolism. $2X10^4$ cells/well were seeded into Seahorse culture plate. Cells were pre-treated with 10mM EP for 1 hour, then washed out with non-buffer medium and OCR and extracellular acidification rate (ECAR) measured in the Seahorse XF bioenergetic assay. Individual compounds were added sequentially as shown. Data presented are representative of three experiments.

Supplemental Figure 7.



Supplemental Figure 7. Proposed mechanisms of autophagy inhibition during HDIL-2 immunotherapy promoting long term tumor regression. Administration of HDIL-2 stimulates the host immune system, induces immune cell proliferation and infiltration and mediates an anti-tumor response. HDIL-2 increases inflammatory cytokine production from immune cells which can act to enhance the anti-tumor response. Meanwhile, the cytokine storm produced from excess cytokines induces 'systemic autophagy', decreasing normal organ function in favor of autophagy induced 'survival mode'. When CQ is combined, it first reduces inflammatory cytokine production, inhibits systemic autophagy and therefore limits toxicity of HDIL-2. Second, CQ directly inhibits tumor cell autophagy, induces apoptosis, resulting in a direct anti-tumor effect. Third, CQ enhances immune cell responsiveness, either by increasing the efficacy of HDIL-2 immunotherapy via immune compartment effects or by altering tumor metabolic adaptation, enhancing tumor cell immunogenicity.