Supplemental Figure 1

m-MDSC gating strategy using various HLA-DR cutoffs based on expression of HLA-DR in lineage positive cells yields differing m-MDSC frequencies and is susceptible to inter-user variability.

Supplemental Figure 2.

PBMCs available in multiple aliquots were serially analyzed over 3 weeks to evaluate day-to-day reproducibility of $CV_{HLA\text{-}DR}$ measures. A standard error of measurement of 1.4% and 0.7%, respectively for QC sample 1 and 2 was detected.

Supplemental Figure 3.

PBMCs from metastatic melanoma patients treated with ipilimumab depleted of CD14-expressing cells were stimulated to proliferate with OKT-3 and IL-2. CFSE dilution of CD3+ T cells in the culture is measured in the absence of CD14+ cells or with CD14+ cells added back.

Supplemental Figure 4

Different summary statistics can be applied to measure HLA-DR expression on lineage negative, CD14+CD11b+ cells and assess relationship to overall survival (using maximum logrank statistics as described in Methods). CV was used as a self-normalizing measurement that was preferred to eliminate non-biological variation in clinical measurements (e.g. day-to-day variation, differences in sample handling and FACS acquisition).

Supplemental table 1. Univariate analysis of relationship between m-MDSC and overall survival at pre-treatment baseline and week 6 after ipilimumab treatment at 10mg/kg and 3mg/kg.