Figure S1

Figure S1. Specific recruitment of engineering T cell into the collagen gel region. T cells injected in media channel without the presence of HepG2-Env do not migrate into the collagen gel region after 15 hrs (scale bar 400μm).

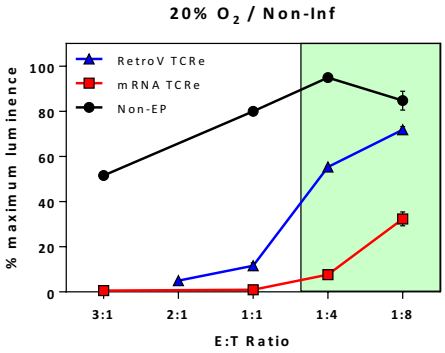
20% O2 / Non-Inf
RetroV TCRe
mRNA TCRe
Non-EP
% maximum luminence
100
80
60
40
20
0
3:1
2:1
1:1
1:4
1:8
E:T Ratio

Figure S2

Figure S2. Analysis of different engineered T cell preparates using 2-Dimensional well-based cytotoxicity assay. Different engineered HBV-specific T cell preparates (RetroV TCRe; mRNA TCRe; Non-electroporated controls) were co-cultured for 15hrs with luciferase expressing HepG2-Env targets and their cytotoxicity were assessed at different effector to target ratios under normal *in vitro* assay conditions (20% Oxygen and no inflammatory cytokines). Maximum luminence was defined using wells where only luciferase+ HepG2-Env targets were seeded. Green shaded areas denote E:T ratios where targets were not completely killed and data from these E:T ratios were further analyzed.

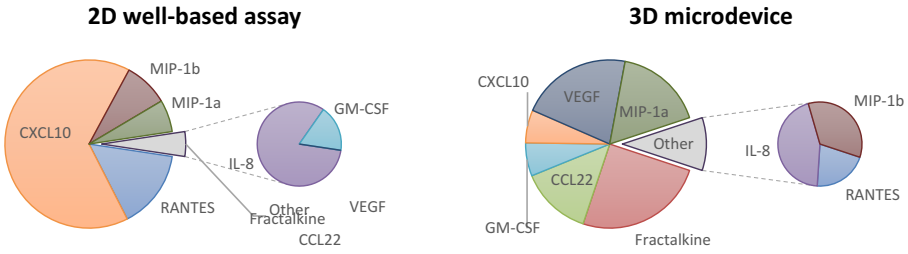
2D well-based assay
MIP-1b
MIP-1a
GM-CSF
CXCL10
IL-8
RANTES
Other
Fractalkine
VEGF
CCL22
3D microdevice
CXCL10
VEGF
MIP-1a
MIP-1b
Other
IL-8
CCL22
RANTES
GM-CSF
Fractalkine

Figure S3

Figure S3. P**rofile of secreted factors present in the supernatants obtained from classical 2D well-based and 3D microdevice assays.** Concentration of soluble factors in the supernatants obtained from mRNA electroporated TCR-T cells co-cultured in 3D microdevices or in classical 2D assay with HBV envelope expressing HepG2 targets were analyzed. The top 5 soluble factors detected in each system were collated and representative pie charts are displayed. The experiment was performed twice.

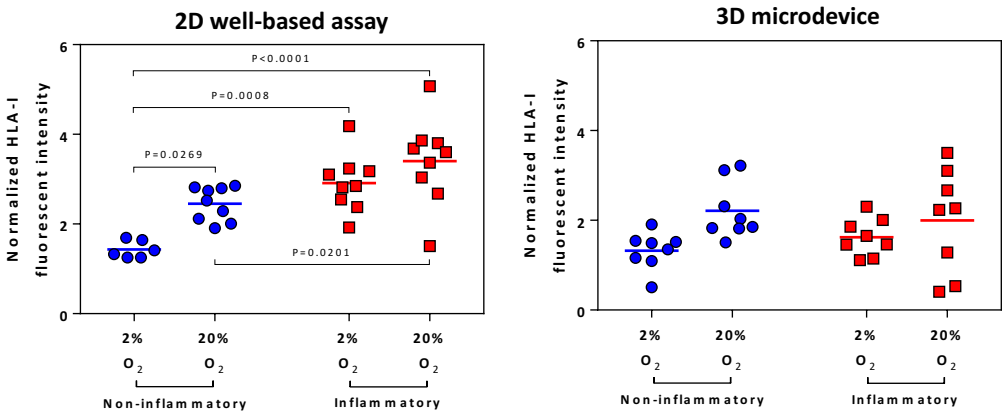
2D well-based assay
3D microdevice
Normalized HLA-I fluorescent intensity
P<0.0001
P=0.0008
P=0.0269
P=0.0201
2% O2
20% O2
Non-inflammatory
Inflammatory

Figure S4

Figure S4. HLA-I expression on targets cultured in classical 2D well-based and 3D microdevice assays. Cell surface expression of HLA-I molecules on HBV envelope expressing HepG2 targets cultured in a 3D microdevice or on microwell plates at the indicated conditions are shown. Each dot represents a single experiment. HLA class I fluorescent intensity was normalized by cell number and differences in HLA expression among different treatments were evaluated by Two-way ANOVA with Tukey's multiple comparison test.