

1 **Supplementary Figure and Table Legends**

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3 Table S1: Expression levels of immunogenic tumor antigens as reported by Cheevers *et al.*<sup>23</sup> translated to  
4 murine gene names (Mouse Genome Informatics) in KPC3, AE17 and B16 with a minimal threshold of 1  
5 RPKM (reads per kilo base per million mapped reads).

6 Fig. S1: Venn diagram illustrating overlapping and non-overlapping genes of the tumor cell lines KPC3  
7 (pancreatic cancer), AE17 (mesothelioma), B16F10 (melanoma) and MC38 (colon adenocarcinoma).

8 Fig. S2: MFI of CD11c, CD40, PD-L1, MHCII, CD80 and CD86 on cultured DCs for vaccination. Control non-  
9 loaded DCs were not stimulated with CpG.

10 Fig. S3: Percentage of CD69+, Ki67+ and CD44+CD62L- subsets of circulating CD4+ and CD8+ T cells at  
11 day -3, 4 and 20. N=8 per group. Significance was determined using the non-parametric Mann-Whitney  
12 U test. Data presented as the mean  $\pm$  s.e.m. \*P<0.05.

13 Fig. S4: Relative production of IFN $\gamma$ , IL-2 and TNF $\alpha$  by CD8+ splenocytes of AE17 lysate-DC/ $\alpha$ CD40-  
14 treated and untreated after stimulation with DCs loaded with KPC3, AE17 or B16F10, or non-loaded DCs,  
15 normalized for untreated mice. N=6-10 per group. Significance was determined using the non-  
16 parametric Mann-Whitney U test. Data presented as the mean $\pm$ s.e.m. \*\*P<0.01, \*\*\*P<0.001,  
17 \*\*\*\*P<0.0001.

18 Fig. S5: Lysate-DC is not effective as monotherapy in tumor-bearing mice. (A) CD3+, CD4+ and CD8+  
19 circulating T cells as a percentage of alive CD45+ cells, four days after DC vaccination. (B) Percentage of  
20 CD44+CD62L- and Ki67+ subsets of CD4+ and CD8+ circulating T cells, four days after DC vaccination. (C)  
21 Tumor volume measured over time, and tumor size at the day of sacrifice (day 22). (D) CD3+, CD4+ and  
22 CD8+ TILs as a percentage of alive CD45+ cells. N=5-9 per group. Significance was determined using the  
23 non-parametric Mann-Whitney U test. Data presented as the mean  $\pm$  s.e.m. \*P<0.05, \*\*P<0.01,  
24 \*\*\*P<0.001.

25 Fig. S6: (A) Study setup (B) Tumor volume measured over time, and tumor size at day of sacrifice (day  
26 18). (C) Percentage of CD69+, Ki67+, PD-1+ and CD44+CD62L- subsets of CD4+ and CD8+ circulating T  
27 cells, four days after treatment initiation. (D) Memory status of CD4+ and CD8+ circulating T cells at day  
28 9 and day 16. (E) Number of CD3+, CD4+, CD8+, CD4+CD25+FoxP3+ TILs per mg tumor. (F) MFI of PD-1  
29 and Lag-3 of CD4+ and CD8+ TILs. N=7-8 per group. Significance was determined using the non-  
30 parametric Mann-Whitney U test. Data presented as the mean  $\pm$  s.e.m. \*P<0.05, \*\*P<0.01, \*\*\*P<0.001.

31 Fig. S7: (A) Study setup in mesothelioma model. (B) Kaplan-Meier analysis of treated and untreated  
32 animals. (C) Percentage of PD-1+ and Ki67 subsets of CD4+ and CD8+ circulating T cells, on day 16. N=5  
33 per group. Significance was determined using the non-parametric Mann-Whitney U test. Data presented  
34 as the mean  $\pm$  s.e.m. \*P<0.05, \*\*P<0.01.

35 Fig. S8: Tumor outgrowth curves of treated and untreated tumor-bearing mice.

36 Fig. S9: (A) Interim blood analysis on day 14. Percentage of CD4+ and CD8+ of CD3+ T cells. (B) Absolute  
37 number of CD3+, CD4+, CD8+, CD335+ and CD19+ cells per  $\mu$ L blood drawn on day 14.

38 Fig. S10: Orthotopic tumors taken out on day 17.

39 Fig. S11: Fraction of non-myeloid (CD45-), monocyte (CD45+F4/80-CD11b+Ly6C+Ly6G-), granulocyte  
40 (CD45+F4/80-CD11b+Ly6C-Ly6G+), cDC1 (CD45+F4/80-CD11b+CD11b-CD11c+MHCI+CD103+), cDC2  
41 (CD45+F4/80-CD11b+CD11c+MHCI+), MDSC (CD45+F4/80-CD11b+Ly6CintLy6Gint) and TAM  
42 (CD45+F4/80+CD11b+) as part of a whole of treated and untreated tumors.

43 Fig. S12: Hierarchical clustering of individual tumor samples based on genes significantly different  
44 between groups.

45 Fig. S13: Vulcano plots of differentially expressed genes between DC vaccination vs combination therapy  
46 (A) or  $\alpha$ CD40 vs combination therapy (B). The X-axis is log<sub>2</sub> fold change and the Y-axis is -log<sub>10</sub> of the  
47 original *p*-value. Markers with *p*-values < 0.05 and log<sub>2</sub> fold change > 0.5 are marked in red, while  
48 markers with *p*-values < 0.05 and log<sub>2</sub> fold change < -0.5 are marked in green. The two vertical lines

49 indicate the log<sub>2</sub> fold change threshold of 0.5 and -0.5. The horizontal line indicates the original *p*-value  
50 threshold of 0.05.

51 Fig. S14: (A) GSEA of T-cell exhaustion gene sets in tumors of  $\alpha$ CD40 or DC therapy versus combination  
52 therapy treated mice, presented as the normalized enrichment score (NES). (B) GSEA of T-cell  
53 exhaustion and glycolysis gene sets in tumors of combination therapy versus  $\alpha$ CD40 treated mice,  
54 presented as the normalized enrichment score (NES).

55 Fig. S15: Hematoxylin and Eosin, Serius Red and Trichromic staining on tumor tissue.

56 Fig. S16: CD31 immunohistochemistry staining on tumor tissue of treated and untreated mice.

57 Fig. S17: (A) Number and percentage of PD-1+, Tim-3+, VISTA+, CD39+ and NKG2A+ subsets of CD4+ TILs.  
58 (B) Number and percentage of IFN $\gamma$ +, Granzyme B+, IL-10+ and Ki67+ subsets of CD4+ TILs. (C) Detection  
59 of co-expression of inhibitory receptors (PD-1, Tim-3 and CD39) on CD4+ and CD8+ TILs. Numbers within  
60 circles represent percentage of TILs with 0 inhibitory receptors. (D) Percentage of PD-1/TIM-3 double  
61 positive and negative cells of CD4+ and CD8+ TILs. N=7-8 per group. Significance was determined using  
62 the non-parametric Mann-Whitney U test. Data presented as the mean  $\pm$  s.e.m. \**P*<0.05, \*\**P*<0.01,  
63 \*\*\**P*<0.001.

64 Fig. S18: ELISA-based IL-12p40 detection in supernatant of bone-marrow derived DCs stimulated with  
65 FGK45 or isotype. Significance was determined using the Student's t-test. Data presented as the mean  $\pm$   
66 s.e.m. \*\*\**P*<0.001.

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