

Targeting Pancreatic Ductal Adenocarcinoma Acidic Microenvironment

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Supplementary Information

Supplementary Table 1S. Mean (and SEM) of luminescence signal in pancreas and fluorescence signal of Alexa647-WT-pHLIP in pancreas, kidney, liver and lung obtained after image analysis of organs from athymic nude mice without and with tumor (orthotopic model of PDAC with luciferase-labeled Capan-2 cells. The data are shown on Figure 1.

	Normal, + WT <i>n</i> = 5	Tumor, - WT <i>n</i> = 5	Tumor, + WT <i>n</i> = 5
Pancreas luminescence $\times 10^7$, p/s/cm ² /sr	0.013 ± 0.004	3.493 ± 1.718	4.634 ± 2.889
Pancreas fluorescence $\times 10^{-4}$	0.209 ± 0.079	0.055 ± 0.010	0.747 ± 0.129
Kidney fluorescence $\times 10^{-4}$	0.307 ± 0.065	0.035 ± 0.011	1.283 ± 0.151
Liver fluorescence $\times 10^{-4}$	0.416 ± 0.220	0.060 ± 0.007	0.857 ± 0.249
Lung fluorescence $\times 10^{-4}$	0.061 ± 0.022	0.031 ± 0.005	0.084 ± 0.005

Table 2S. Mean (and SEM) of fluorescence signal of Alexa546-WT-pHLIP and Alexa546-2K-WT-pHLIP in pancreas, kidney, liver and lung obtained after image analysis of organs from GEMMs with normal pancreas (non-tumor bearing mice) or PDAC (tumor bearing mice). The data are shown on Figure 3.

	Normal, WT <i>n</i> = 5	Tumor, WT <i>n</i> = 5	Normal, 2K-WT <i>n</i> = 5	Tumor, 2K-WT <i>n</i> = 5
Pancreas fluorescence x10 ⁻⁴	0.136 ± 0.042	2.396 ± 0.392	0.039 ± 0.010	0.555 ± 0.265
Kidney fluorescence x10 ⁻⁴	0.123 ± 0.021	0.524 ± 0.024	0.031 ± 0.003	0.085 ± 0.026
Liver fluorescence x10 ⁻⁴	0.272 ± 0.022	0.670 ± 0.099	0.415 ± 0.105	0.837 ± 0.307
Lung fluorescence x10 ⁻⁴	0.018 ± 0.003	0.029 ± 0.004	0.012 ± 0.001	0.047 ± 0.013

Table 3S. Mean (and SEM) of fluorescence signal of Alexa546-WT-pHLIP in pancreas, kidney, liver and lung obtained after image analysis of organs from non-tumor bearing mice, tumor bearing mice, mice containing PanIN lesions and mice with pancreatitis. The data are shown on Figure 5.

	Normal, - WT <i>n</i> = 5	Tumor, - WT <i>n</i> = 5	Normal, + WT <i>n</i> = 5	Pancreatitis, + WT <i>n</i> = 5	PanIN, + WT <i>n</i> = 5	Tumor, + WT <i>n</i> = 5
Pancreas fluorescence x10 ⁻⁴	0.032 ± 0.013	0.074 ± 0.003	0.046 ± 0.016	0.111 ± 0.020	0.421 ± 0.127	1.789 ± 0.467
Kidney fluorescence x10 ⁻⁴	0.033 ± 0.011	0.015 ± 0.003	0.267 ± 0.021	0.271 ± 0.060	0.297 ± 0.045	0.687 ± 0.193
Liver fluorescence x10 ⁻⁴	0.060 ± 0.006	0.059 ± 0.009	0.839 ± 0.126	0.682 ± 0.101	1.075 ± 0.055	0.802 ± 0.270
Lung fluorescence x10 ⁻⁴	0.024 ± 0.002	0.024 ± 0.004	0.084 ± 0.010	0.096 ± 0.006	0.091 ± 0.015	0.129 ± 0.030

Table 4S. Mean (and SEM) of fluorescence signal of Alexa647-Var3-pHLIP and Alexa546-Var7-pHLIP in pancreas, kidney, liver and lung obtained after image analysis of organs from GEMMs with normal pancreas (non-tumor bearing mice) or PDAC (tumor bearing mice). The data are shown on Figure 6.

	Normal, Var3 <i>n</i> = 5	Tumor, Var3 <i>n</i> = 5	Normal, Var7 <i>n</i> = 5	Tumor, Var7 <i>n</i> = 5
Pancreas fluorescence x10 ⁻⁴	0.078 ± 0.009	0.967 ± 0.280	0.023 ± 0.002	0.524 ± 0.081
Kidney fluorescence x10 ⁻⁴	0.517 ± 0.061	1.865 ± 0.577	0.110 ± 0.016	0.401 ± 0.078
Liver fluorescence x10 ⁻⁴	0.359 ± 0.026	0.474 ± 0.117	0.064 ± 0.007	0.095 ± 0.017
Lung fluorescence x10 ⁻⁴	0.079 ± 0.003	0.130 ± 0.024	0.010 ± 0.001	0.013 ± 0.001

Supplementary Figure S1: WT-pHLIP efficiently detects human pancreatic cancer xenografts. Representative *in vivo* bioluminescent (A, B) and fluorescent (C, D) images of athymic nude mice without tumor and with tumor (orthotopic model of PDAC with luciferase-labeled Capan-2 cells) injected with 40 μM of Alexa647-WT-pHLIP in 100 μl of PBS (+WT-pHLIP) or PBS only (-WT-pHLIP).

