# SUPPLEMENTAL FIGURES

## FOR

# PHARMACOKINETICS AND TRANSGENE EXPRESSION OF IMPLANTED POLYTHEYLENIMINE-BASED PDNA COMPLEXES

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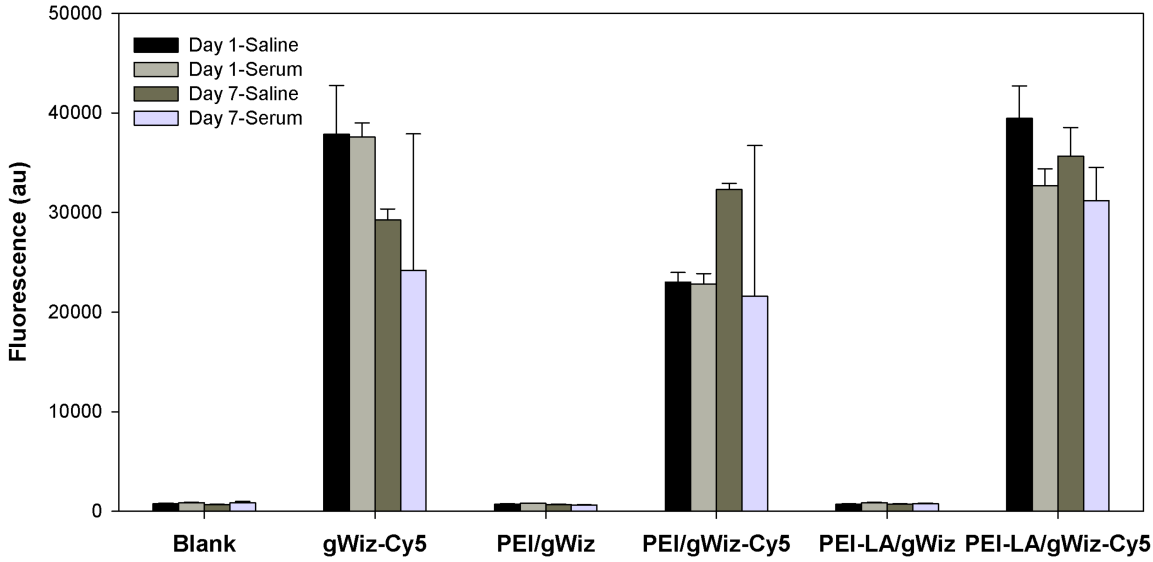
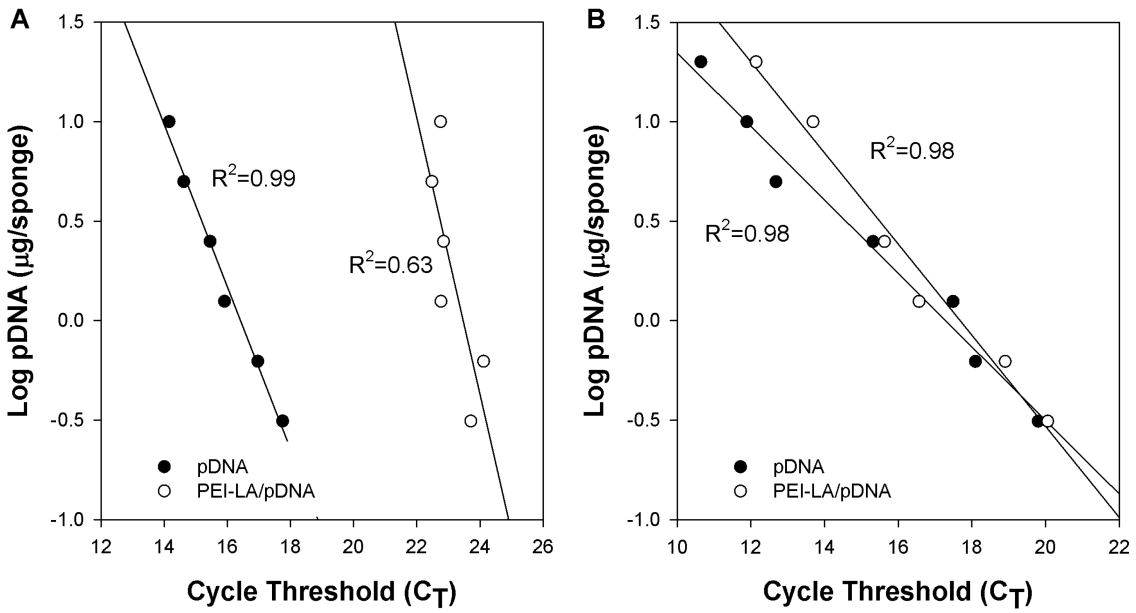
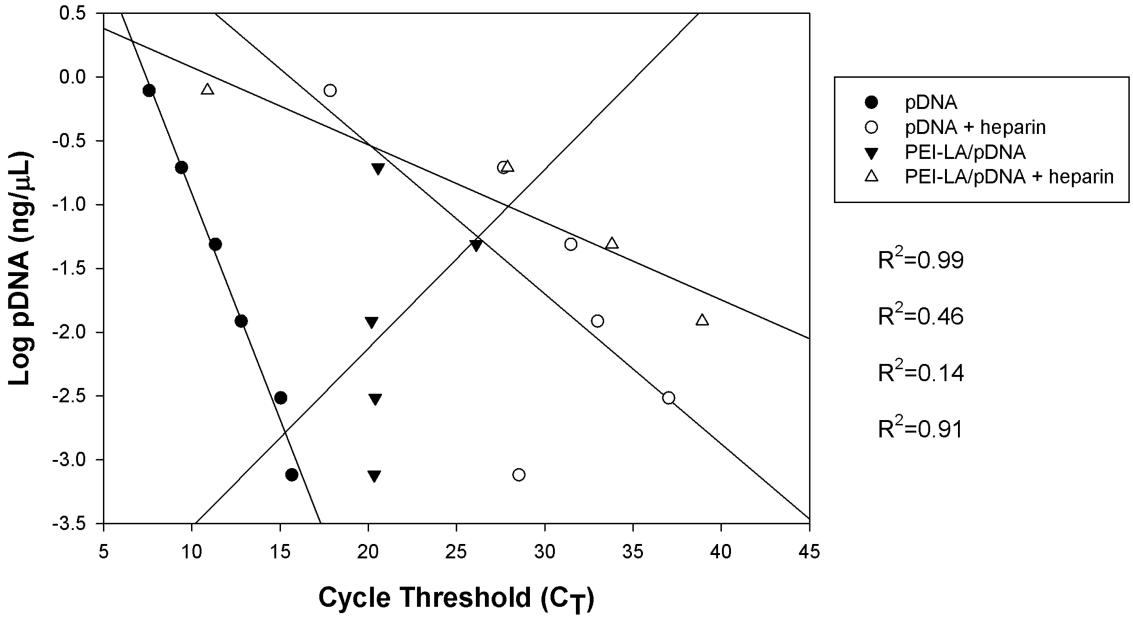


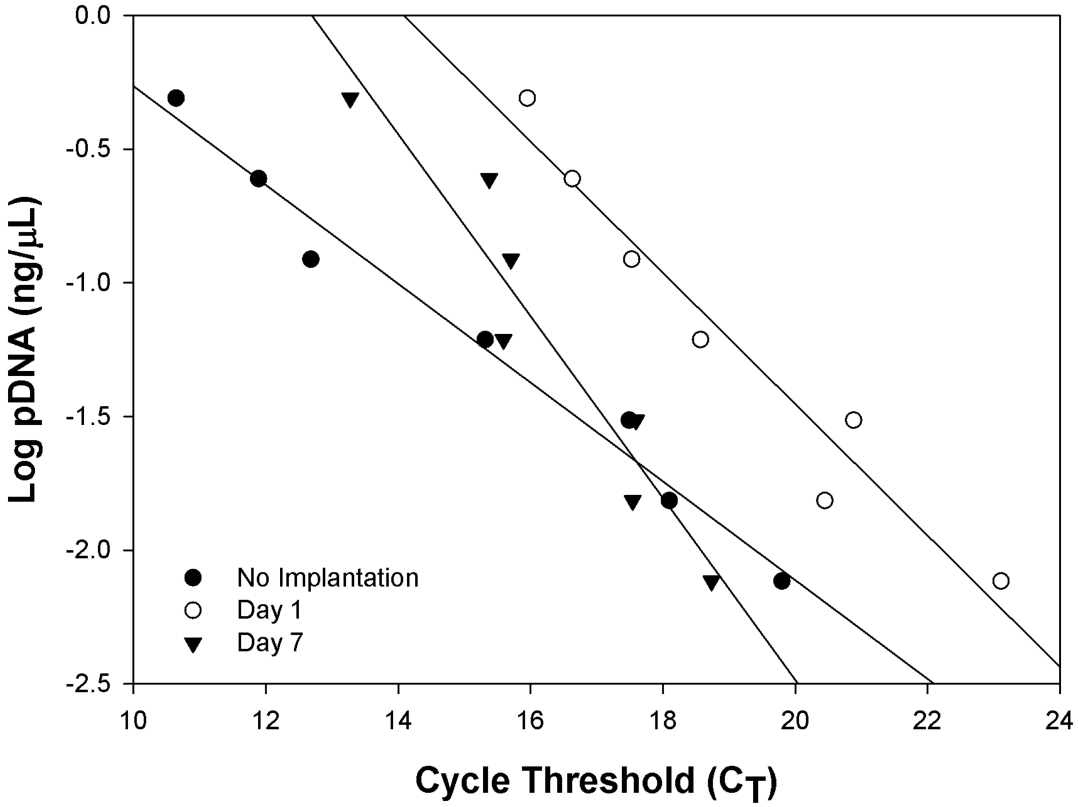

**Supplementary Figure 1: Changes in mean Cy5 fluorescence over time *in vitro*.** Sponges were loaded with saline (blank sponges), naked gWiz-Cy5, and PEI and PEI-LA complexes of gWiz or gWiz-Cy5, and incubated at 37 °C in either saline or full serum. The Cy5 fluorescence in sponges was imaged after 1 or 7 days and expressed as mean + SD.



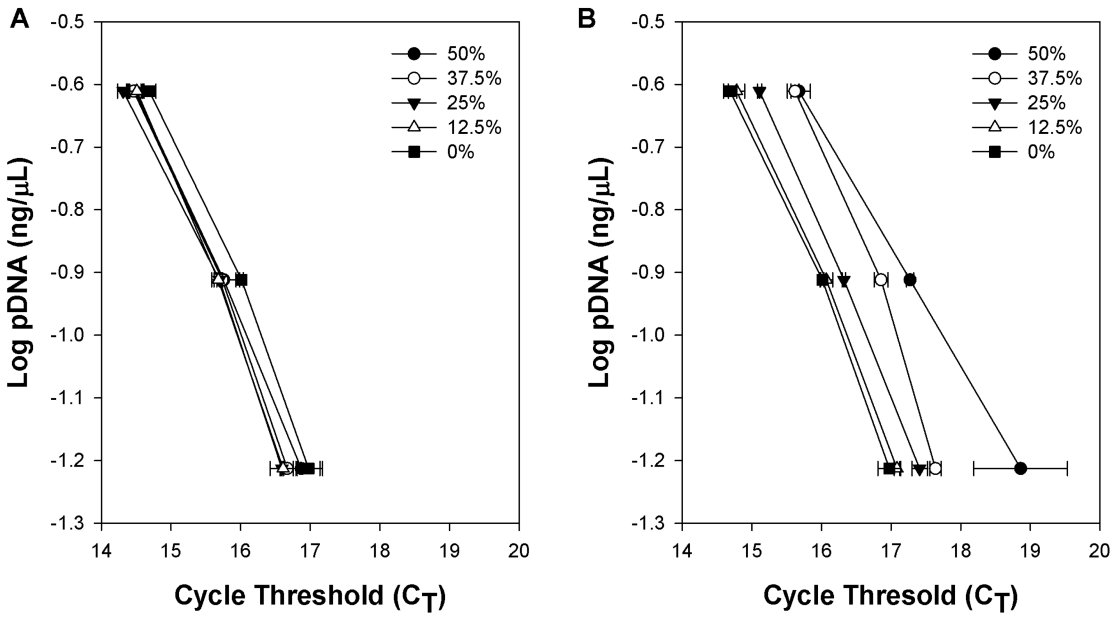

**Supplemental Figure 2: Comparison of pDNA extraction methods.** Sponges were loaded with pDNA (pCAG-dsRED) alone or pDNA bound to PEI-LA at a range of concentrations to give standard curves. pDNA was extracted from the sponges using either a column-based commercial kit (**A**) or a protocol using poly-acrylic acid to dissociate the complexes (**B**). The qPCR was run similarly in both protocols.



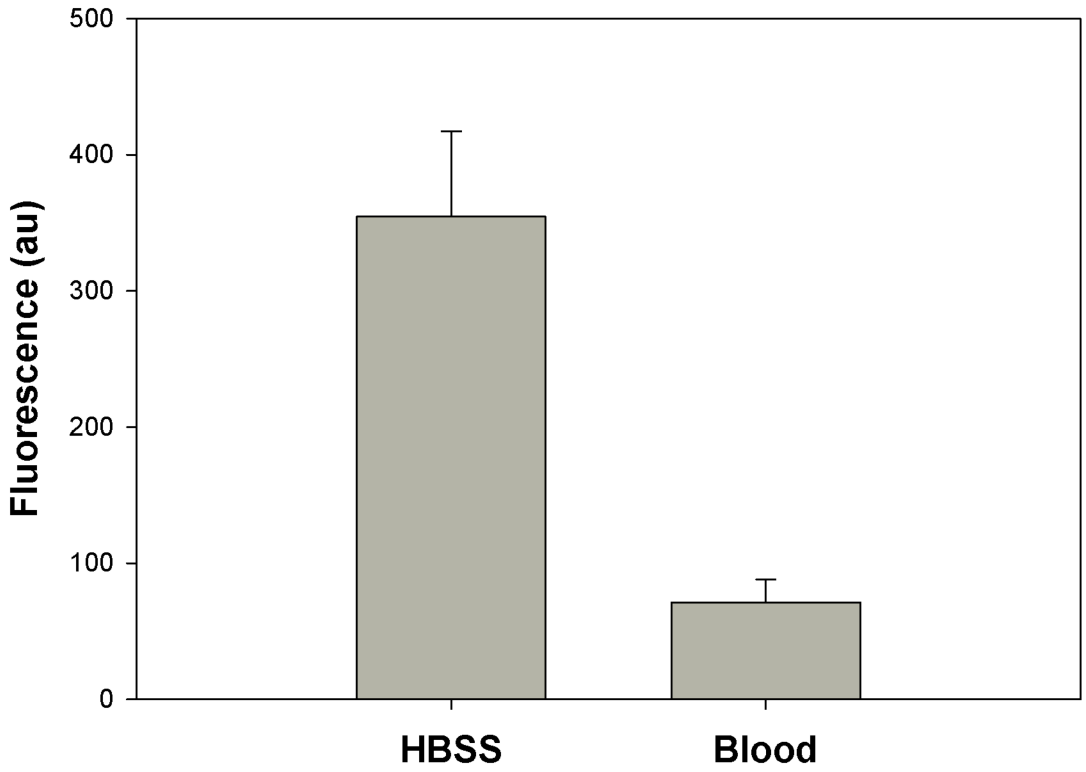

**Supplemental Figure 3: qPCR of heparin-dissociated complexes**. pDNA (pCAG-dsRED) with or without PEI-LA was incubated with or without heparin to dissociate the pDNA from the PEI-LA. The qPCR was then run to determine the $C_T$ for each preparation.



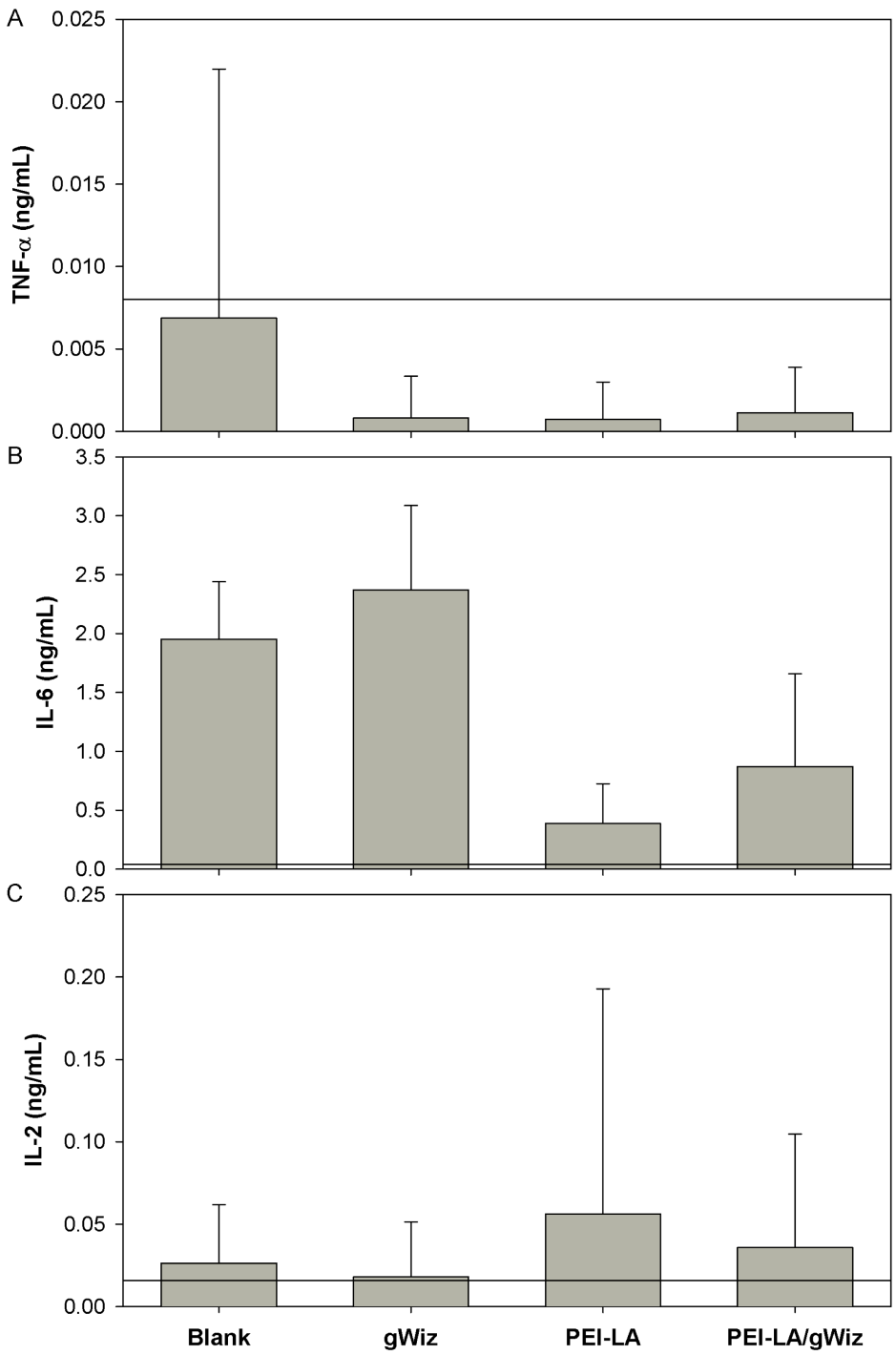

**Supplemental Figure 4: qPCR standard curves as a function of explant time point**. Naked pDNA (pCAG-dsRED) was soaked onto sponges that were either not implanted, or onto homogenates of blank sponges that were implanted for 1 and 7 days.




**Supplemental Figure 5: Blood spiked qPCR analysis**. Standard curves of naked pCAG-DsReRED were spiked with varying amounts of blood components (0 to 50% by volume). An equal amount of either rat serum (**A**) or rat blood clot (**B**) was extracted, diluted, and then added to the pDNA standard curve. Addition of extraction buffer alone (0% sample) served as the reference.




**Supplemental Figure 6: Imaging DsRed fluorescence with blood**. 293T cells expressing DsRed were trypsinized and then diluted with either HBSS or rat blood before being added to sponges. The DsRed fluorescence signal was measured from the sponges and normalized to a sample of HBSS-soaked sponge alone.




**Supplemental Figure 7: Levels of cytokines TNF-α, IL-6 and IL-2 in implanted sponges**. Sponges loaded with saline (blank), gWIZ (25 μg), PEI-LA (125 μg) and PEI-LA/gWIZ complexes (25/125 μg) were implanted subcutaneously for 10 days

and then harvested and cultured in cell culture media for 5 days. The supernatant was collected and assessed for tumor necrosis factor-α (**A**), interleukin-6 (**B**), and interleukin-2 (**C**) levels by commercially avalaible ELISA kits (R&D Systems).