

1 **Supplemental Materials and Methods**

2 **Determination of influenza specific ferret lung immunoglobulin relative levels**

3 Ferret lungs from day 0, 3, 7, and 14 pI were collected and frozen immediately on dry ice
4 and transferred to -80°C for storage before processing. Lung tissues were then
5 homogenized in the lysis buffer (50mM Tris-HCl pH 7.4, 150mM NaCl, 1% Triton-X
6 100) containing the CΦmplete protease inhibitors cocktail (Roche Diagnostics Ltd., Laval,
7 QC, Canada). All the procedures were carried out on ice. We performed ELISA with the
8 homogenates to measure influenza specific immunoglobulin levels in the lungs. We first
9 coated the live A/Mexico/4108/2009 H1N1pdm influenza virions to ELISA plates
10 (Thermo Fisher Scientific, Rochester, NY, USA) followed by appropriate blocking (1%
11 BSA in 0.05% PBST) and then incubation of the lung lysates at the protein concentration
12 of 50µg/mL. After sufficient washing, we incubated the plates with goat anti-ferret IgA,
13 goat anti-ferret IgG, or goat anti-ferret IgM (1:1000, Rockland Immunochemicals Inc.
14 Gilbertsville, PA, USA) followed by sufficient washing and anti-goat HRP antibody
15 incubation (1:10000, Santa Cruz Biotechnology Inc., Santa Cruz, CA, USA). After
16 washing, the plates were developed and read at 490nm and 570nm (for background
17 subtraction).

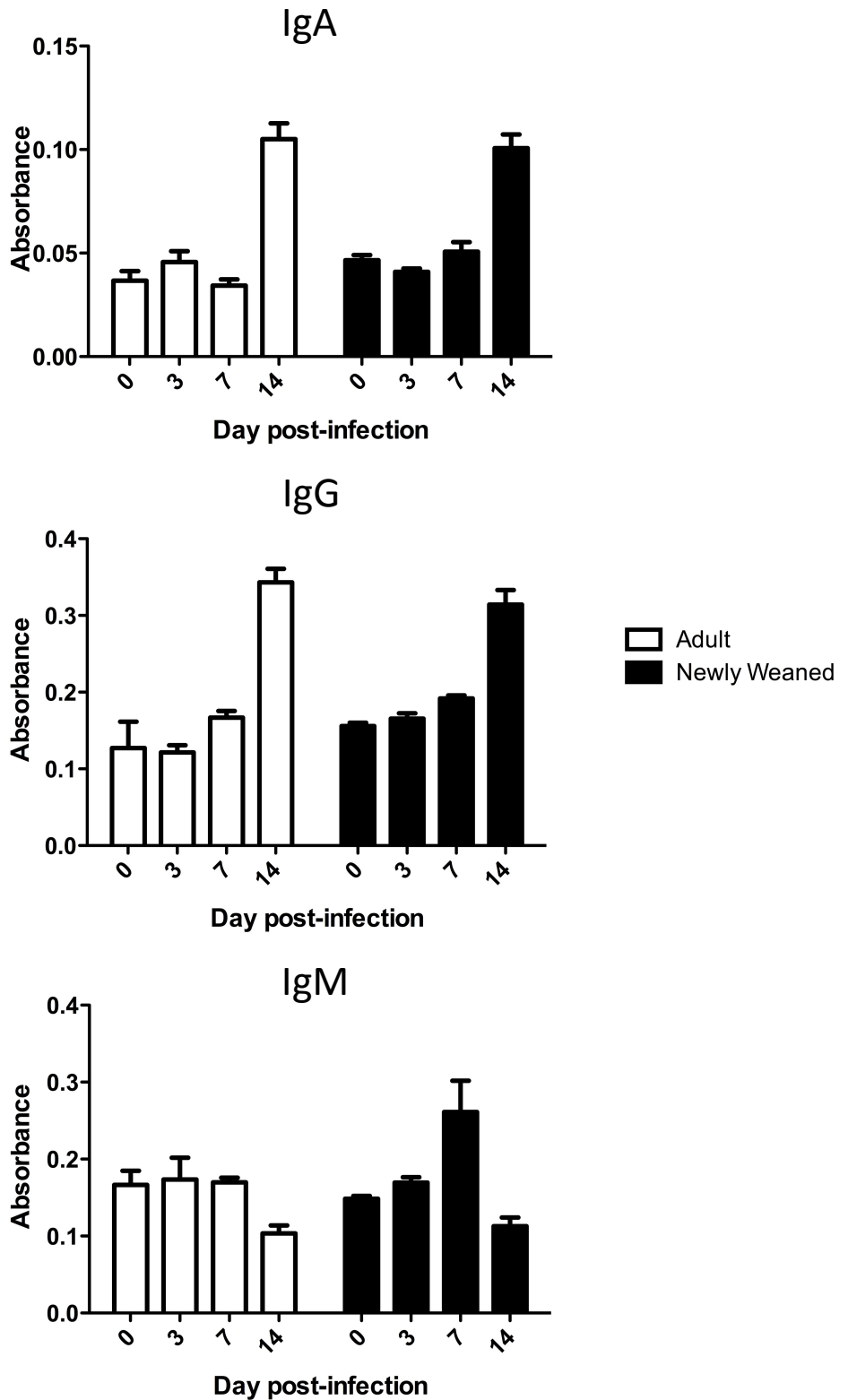


Figure S1. Equivalent influenza specific lung immunoglobulin levels in both infected adult and newly weaned ferrets

Time course of influenza specific lung IgA, IgG, and IgM levels from H1N1pdm infected adult and newly weaned ferrets. Ferret lungs collected at the indicated days pi were homogenized and the lysates were used to measure the relative immunoglobulin levels. Error bars represent standard error of the mean.