Supplementary Table 1. Association between Nasopharyngeal/Oropharyngeal Cycle Threshold (Ct) Values and Duration of Illnessa among CXR+b Cases

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | **1-2 days since onset** | |  | **3-6 days since onset** | |  | **> 7 days since onset** | |  | **p-valuec** | | |
| **Virus** | **nd** | **Ct Value**  **Mean (95% CI)** |  | **nd** | **Ct Value**  **Mean (95% CI)** |  | **nd** | **Ct Value**  **Mean (95% CI)** |  | **1-2 vs.**  **3-6 days** | **3-6 vs.**  **>7 days** | **1-2 vs.**  **>7 days** |
| **Adenovirus** | 58 | 29.5 (28.2-30.8) |  | 77 | 26.8 (25.5-28.0) |  | 28 | 26.4 (24.7-28.2) |  | .008 | .96 | .02 |
| **Coronavirus 229** | 4 | 32.9 (30.1-35.8) |  | 11 | 29.8 (26.4-33.2) |  | 3 | 33.1 (26.6-39.5) |  | .15 | .26 | . |
| **Coronavirus 43** | 13 | 28.0 (24.7-31.3) |  | 15 | 25.4 (22.1-28.6) |  | 9 | 25.4 (20.7-30.0) |  | .11 | .91 | .91 |
| **Coronavirus 63** | 13 | 26.9 (23.6-30.1) |  | 14 | 27.1 (24.8-29.3) |  | 9 | 27.0 (23.1-30.9) |  | .59 | .26 | .12 |
| **Coronavirus HKU** | 10 | 30.3 (25.6-35.1) |  | 15 | 29.0 (25.6-32.4) |  | 12 | 28.6 (24.7-32.4) |  | .86 | .75 | .54 |
| **Influenza A** | 13 | 30.3 (28.5-32.1) |  | 31 | 28.0 (26.9-29.0) |  | 17 | 28.2 (26.3-30.2) |  | .13 | .51 | .07 |
| **Influenza B** | 4 | 26.9 (23.1-30.8) |  | 8 | 27.3 (24.4-30.1) |  | 6 | 28.6 (24.9-32.4) |  | .44 | . | .08 |
| **Influenza C** | 1 | 24.5 (.-.) |  | 6 | 29.5 (24.2-34.7) |  | 2 | 27.6 (23.5-31.6) |  | .91 | .56 | . |
| **HBOV** | 80 | 30.6 (29.2-32.0) |  | 112 | 30.0 (28.8-31.3) |  | 38 | 31.2 (29.6-32.8) |  | .54 | .32 | .67 |
| **HMPV A/B** | 51 | 28.1 (27.1-29.0) |  | 114 | 27.9 (27.2-28.7) |  | 19 | 29.5 (28.1-30.9) |  | .43 | .29 | .52 |
| **Parainfluenza 1** | 31 | 26.0 (24.1-27.9) |  | 50 | 25.4 (23.9-26.8) |  | 8 | 30.8 (27.8-33.8) |  | .20 | .02 | .11 |
| **Parainfluenza 2** | 5 | 32.6 (26.7-38.6) |  | 10 | 31.9 (28.0-35.7) |  | 8 | 37.5 (36.1-38.8) |  | .55 | .32 | .70 |
| **Parainfluenza 3** | 31 | 23.9 (22.2-25.6) |  | 52 | 24.7 (23.5-25.9) |  | 21 | 27.1 (24.5-29.6) |  | .49 | .04 | .21 |
| **Parainfluenza 4** | 15 | 32.1 (29.5-34.8) |  | 21 | 31.0 (29.1-32.8) |  | 8 | 32.9 (29.4-36.5) |  | .62 | .51 | .59 |
| **PV/EV** | 51 | 30.3 (29.3-31.3) |  | 60 | 29.9 (28.9-30.9) |  | 20 | 30.3 (28.6-32.0) |  | .49 | .65 | .78 |
| **Rhinovirus** | 150 | 31.2 (30.6-31.7) |  | 154 | 31.9 (31.3-32.5) |  | 61 | 32.5 (31.8-33.1) |  | .18 | .98 | .16 |
| **RSV** | 108 | 21.6 (20.9-22.3) |  | 290 | 22.1 (21.7-22.6) |  | 62 | 23.4 (22.3-24.6) |  | .51 | .04 | .04 |

a Duration of illness determined by longest duration of symptoms among cough, shortness of breath or fever.

b Chest x-ray positive (CXR+) defined as having radiographic evidence of pneumonia

c P-values obtained comparing mean Ct values after adjusting for age and site. Missing p-values are a result of zero cells.

d Number positive in the nasopharynx for that virus.

Supplementary Table 2. Mean Nasopharyngeal/Oropharyngeal PCR Cycle Threshold (Ct) Values among Chest X-ray (CXR) positive cases by HIV status, South Africa and Zambia Sites

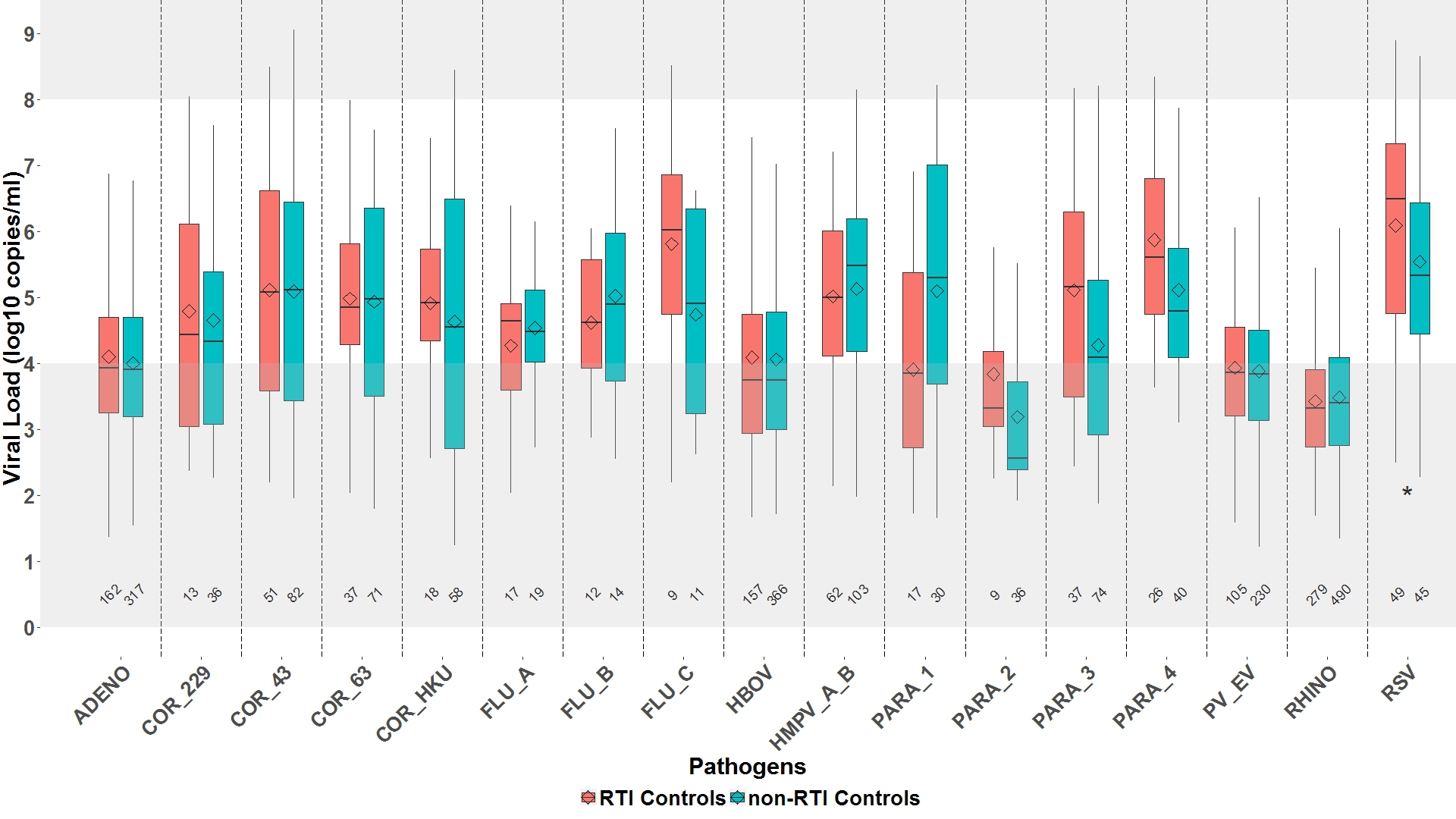
|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Virus** | **HIV Negativea, CXR+b Cases (N=622)** | | | **HIV Positive, CXR+b Cases (N=142)** | | |  |
| **N** | **%** | **Ct Value Mean (95% CI)** | **N** | **%** | **Ct Value Mean (95% CI)** | **p-value (adjusted)c** |
| **Adenovirus** | 82 | 9.9 | 27.2 (26.1-28.3) | 22 | 15.6 | 27.0 (24.8-29.2) | .63 |
| **Coronavirus 229** | 8 | 1.0 | 30.1 (27.1-33.2) | 1 | 0.7 | 35.0 (.) | .37 |
| **Coronavirus 43** | 18 | 2.2 | 27.8 (25.0-30.6) | 5 | 3.5 | 24.4 (18.3-30.6) | **.04** |
| **Coronavirus 63** | 18 | 2.2 | 26.3 (23.6-29.0) | 3 | 2.1 | 31.0 (26.6-35.5) | .55 |
| **Coronavirus HKU** | 20 | 2.4 | 28.3 (25.3-31.3) | 3 | 2.1 | 27.4 (24.1-30.8) | .67 |
| **Influenza A** | 31 | 3.7 | 28.4 (27.2-29.6) | 1 | 0.7 | 22.4 (.) | .18 |
| **Influenza B** | 9 | 1.1 | 27.7 (25.1-30.4) | 0 | 0.0 | . | . |
| **Influenza C** | 5 | 0.6 | 24.0 (22.5-25.6) | 2 | 1.4 | 23.4 (19.6-27.2) | .56 |
| **HBOV** | 113 | 13.7 | 31.1 (29.9-32.3) | 23 | 16.3 | 30.0 (27.3-32.7) | .44 |
| **HMPV A/B** | 73 | 8.8 | 28.0 (27.1-28.9) | 5 | 3.5 | 31.8 (27.8-35.8) | **.02** |
| **Parainfluenza 1** | 20 | 2.4 | 28.9 (27.5-30.3) | 4 | 2.9 | 30.5 (27.3-33.7) | .54 |
| **Parainfluenza 2** | 3 | 0.4 | 33.9 (27.1-40.7) | 1 | 0.7 | 32.1 (.) | . |
| **Parainfluenza 3** | 47 | 5.7 | 24.7 (23.3-26.2) | 13 | 9.2 | 28.1 (25.3-30.8) | **.01** |
| **Parainfluenza 4** | 21 | 2.5 | 31.4 (29.3-33.5) | 4 | 2.8 | 35.2 (32.5-37.9) | .11 |
| **PV/EV** | 50 | 6.0 | 29.6 (28.8-30.4) | 5 | 3.5 | 32.3 (28.8-35.7) | .32 |
| **Rhinovirus** | 195 | 23.6 | 31.2 (30.8-31.6) | 29 | 20.6 | 32.1 (31.1-33.2) | .19 |
| **RSV** | 236 | 28.5 | 22.0 (21.4-22.5) | 18 | 12.8 | 27.0 (23.7-30.3) | **<.001** |

a Includes 155 children without clear documentation of negative status but who are believed to be HIV-negative.

b CXR+ defined as having radiographic evidence of pneumonia.

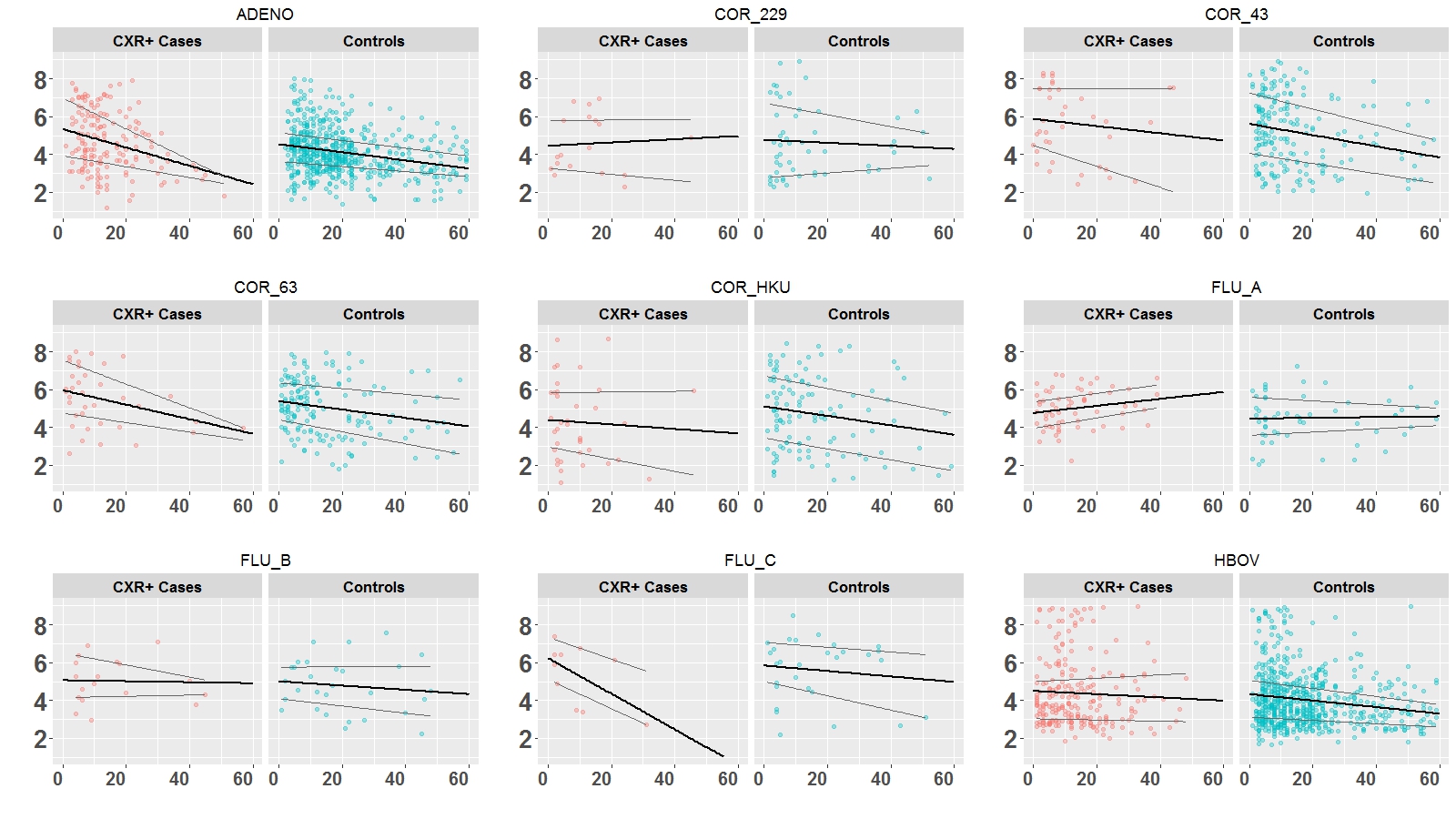
c Comparing mean Ct value in HIV positive vs. HIV negative cases adjusting for age and site. Missing p-values are a result of zero cell

Supplementary Figure 1. Nasopharyngeal/Oropharyngeal Viral Density (log10 copies/mL) for Controls with and without Symptoms of Respiratory Tract Illness (RTI) among those with Positive Densities

 Restricted to 5 of 7 PERCH sites (excluding Zambia and South Africa which had few RTI controls). Box and whiskers plot features include the following: central line in box is median, bottom line of box is first quartile (25%), top line of box is third quartile (75%), diamond is mean, top and bottom of whiskers represent 95% confidence intervals. Shaded areas indicate areas outside the linear range of the assay for calculation of viral load from Ct values, where there is a greater degree of uncertainty in viral density calculations. Numbers on x-axis indicate number of positive results for that virus.

\*p-value comparing means between RTI and non-RTI controls <.05 after adjusting for age and site.

Supplementary Figure 2. Linear Regression of Nasopharyngeal/Oropharyngeal Viral Density (log10 copies/mL) on Age for CXR+ Cases and Controls among those with Positive Densities, by Age



**Age (months)**

**Age (months)**

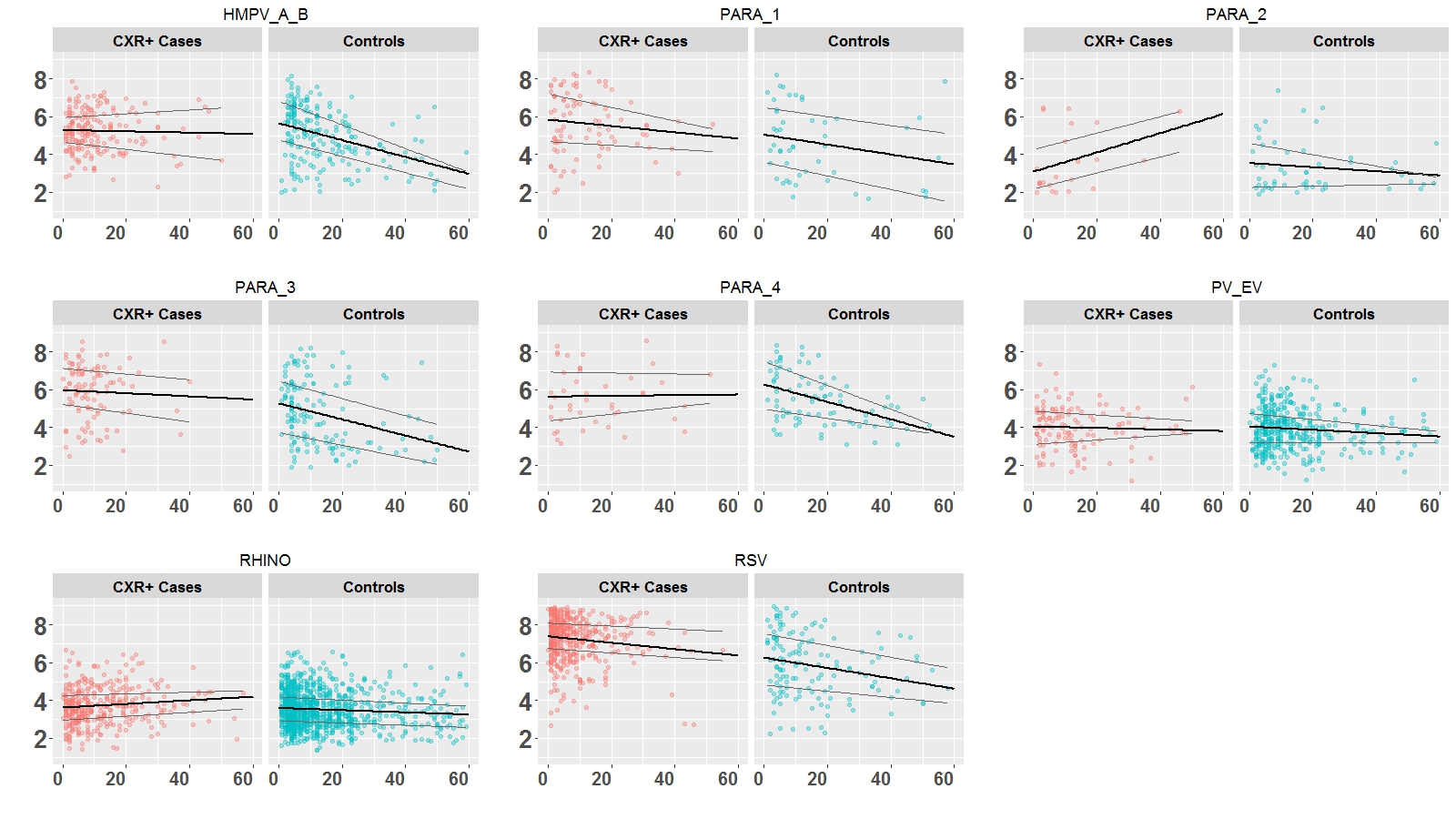
**Age (months)**

**Viral Density (log10 copies/ml)**

**Viral Density (log copies/ml)**

**Viral Density (log copies/ml)**

Supplementary Figure 2 (*continued*). Linear Regression of Nasopharyngeal/Oropharyngeal Viral Density (log10 copies/mL) on Age for CXR+ Cases and Controls among those with Positive Densities, by Age



**Age (months)**

**Age (months)**

**Age (months)**

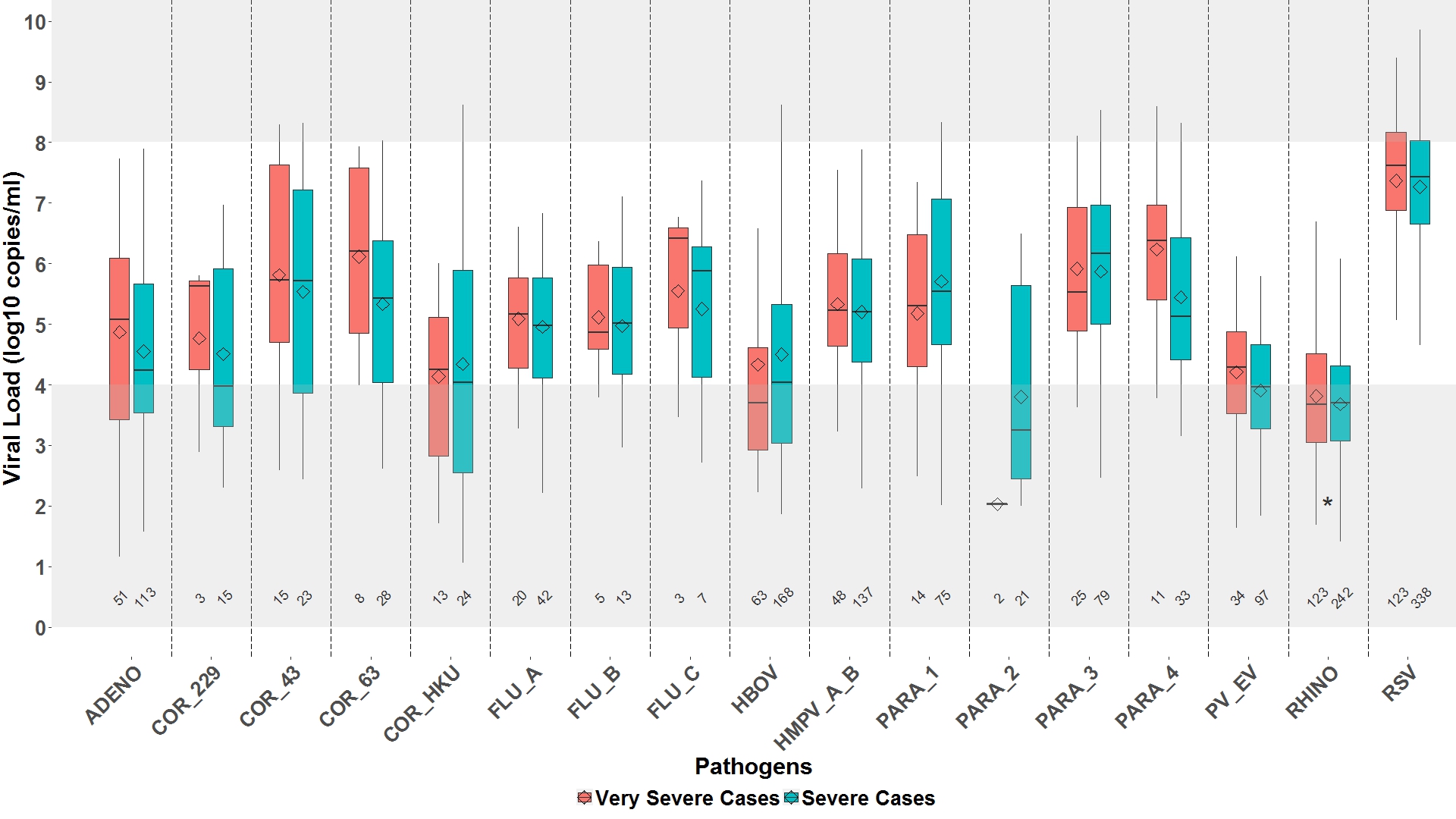
**Viral Density (log copies/ml)**

**Viral Density (log copies/ml)**

**Viral Density (log10 copies/ml)**

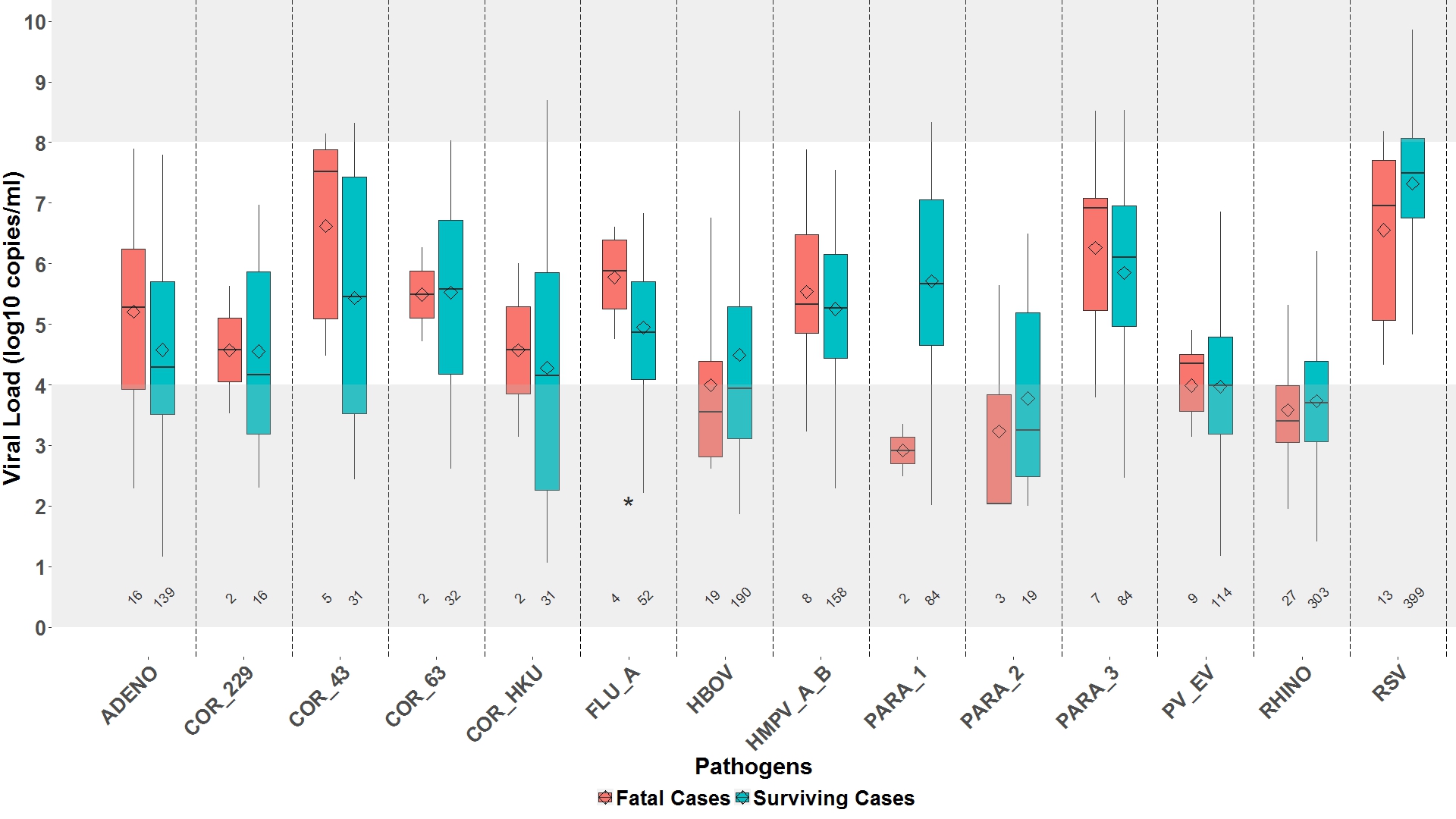
Chest x-ray positive (CXR+) defined as having radiographic evidence of pneumonia. Black line: simple linear regression of NP/OP viral density on age. Grey lines: fitted 25% and 75% quantiles of the viral density using quantile regression. For CXR + cases, a significant trend of decreasing viral load with increasing age was observed for adenovirus and RSV and a significant trend of increasing viral load with increasing age was observed for rhinovirus after adjusting for site. For controls, a significant trend of decreasing viral load with increasing age was observed for adenovirus, coronavirus 43, coronavirus HKU, influenza B, influenza C, parainfluenza viruses 3 and 4, parechovirus/enterovirus, rhinovirus, and RSV after adjusting for site.

Supplementary Figure 3. Nasopharyngeal/Oropharyngeal Viral Density (log10 copies/mL) among CXR+ Cases with Positive Densities, by Pneumonia Severity

Chest x-ray positive (CXR+) defined as having radiographic evidence of pneumonia. Box and whiskers plot features include the following: central line in box is median, bottom line of box is first quartile (25%), top line of box is third quartile (75%), diamond is mean, top and bottom of whiskers represent 95% confidence intervals. Shaded areas indicate areas outside the linear range of the assay for calculation of viral load from Ct values, where there is a greater degree of uncertainty in viral density calculations. Numbers on x-axis indicate number of positive results for that virus.

\*p-value comparing means between severe and very severe cases < .05 after adjusting for age and site.

Supplementary Figure 4. Nasopharyngeal/Oropharyngeal Viral Density (log10 copies/mL) for CXR+ Cases among those with Positive Densities, by Vital Status



Chest x-ray positive (CXR+) defined as having radiographic evidence of pneumonia. Box and whiskers plot features include the following: central line in box is median, bottom line of box is first quartile (25%), top line of box is third quartile (75%), diamond is mean, top and bottom of whiskers represent 95% confidence intervals. Shaded areas indicate areas outside the linear range of the assay for calculation of viral load from Ct values, where there is a greater degree of uncertainty in viral density calculations. Numbers on x-axis indicate number of positive results for that virus. Influenza B, parainfluenza 1, and parainfluenza 4 excluded due to no positive results in at least one of the comparison categories.

\*p-value comparing means between fatal and surviving cases <.05 after adjusting for age and site.

**Abbreviations for Supplementary Figures 1-4:**

ADENO, adenovirus; COR\_229, human coronavirus 229; COR\_43, human coronavirus 43; COR\_63, human coronavirus 63; COR\_HKU, human coronavirus HKU; FLU\_A, influenza A; FLU\_B, influenza B; FLU\_C, influenza C; HBOV, human bocavirus; HMPV\_A\_B, human metapneumovirus A/B; PARA\_1, parainfluenza virus 1; PARA\_2, parainfluenza virus 2; PARA\_3, parainfluenza virus 3; PARA\_4, parainfluenza virus 4; PV\_EV, parechovirus/enterovirus; RHINO, rhinovirus; RSV, respiratory syncytial virus.

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