Methods

*Data Source*

Data for the current study comes from previously reported prospective studies of human herpesvirus 6 (HHV-6) natural history and human bocavirus-1 shedding events from a group of neonates1,2. Incidence of HHV-6 has been reported between 64-83% in the United States and in some studies up to 90% prevalence has been reported3. For this reason, the entire cohort of children carried HHV-6 at one point in the 2-year study period.

*Study Population*

Inclusion criteria for enrollment included pregnancy of the mother, receipt of care at a seattle area-based obstetrical practice, and provision of informed consent prior to participation. Children were then followed from birth for up to two years between April 1997 and August 2003. No further exclusion criteria were applied within the birth cohort.

*Clinical Data Collection*

Demographic data were collected from the mother at enrollment through administered surveys. Symptom data were collected using a daily diary recorded by the mother and/or father. Symptoms included: fever (temperature > 38˚C), roseolla, cough, rhinnorhea, vomiting, diarrhea, rash, fussiness above the baseline level for the child, seizure, and physician visit for illness. Data was further collected on breast-feeding and breast-feed duration, group child-care, daycare attendance and duration and playgroup activity.

*Statistical Methods*

For the purposes of this analyses, acute respiratory infection



Results

Table 1. Infant Demographics and Characteristics (n = 86)

|  |  |  |  |
| --- | --- | --- | --- |
| Characteristic | No. | % | Total No. |
| Female | 32 | 37.2 | 86 |
| Race |  |  |  |
| Asian | 9 | 10.5 | 81 |
| Black | 3 | 3.5 | 81 |
| White | 62 | 72.1 | 81 |
| Other | 7 | 8.1 | 81 |
| Unknown | 5 | 5.8 | 81 |
| Siblings | 41 | 47.7 | 86 |
| Total Family Members, Median (IQR) | 2.5 | 2.0 – 3.0 | 86 |
| Family Members < 18 years old, Median (IQR) | 0 | 0 – 1 | 86 |
| Breast Fed | 84 | 97.7 | 86 |
| Breast Feed Duration (weeks), Mean (SD) | 47.4 | 25.0 | 83 |
| Attended Daycare | 34 | 39.5 | 86 |
| Daycare Duration (weeks), Median (IQR) | 65.1 | 41.0 – 80.6 | 34 |
| Age at First Daycare Attendance (weeks), Median (Range) | 24 | 6 – 87 | 34 |
| Participated in Playgroup Activity | 55 | 64.7 | 85 |
| Playgroup Duration (weeks), Mean (SD) | 62.7 | 30.0 | 55 |
| Mother’s Age Infant Birth (years), Mean (SD) | 34 | 4.3 | 76 |
| Family Income ≥ 50,000 per year | 69 | 97.2 | 71 |

Table 2. Recorded Symptoms and ARI episodes from 86 infants

|  |  |  |
| --- | --- | --- |
| Full Sample | N | % |
| Fever | 583 / 8244 | 7.1 |
| Cough | 1538 / 8244 | 18.7 |
| Rhinnorhea | 2707 / 8244 | 32.8 |
| ARI | 1410 / 8244 | 17.1 |
|  |  |  |
| Per Infant | Median | Range |
| Fever | 6 | 1 – 20 |
| Cough | 17 | 1 – 51 |
| Rhinorrhea | 30.5 | 7 – 85 |
| ARI | 14.5 | 1 – 46 |

Figure 1. ARI Episodes and Daycare Attendance



|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Model | HR  (95% CI) | Estimated Q-matrix  (95% CI) | Sojourn Time (Weeks)  (95% CI) | Mean Sojourn Difference in ARI episodes  (95% CI)†  ‡ |
| Female | 0.93 (0.78, 1.10)  0.99 (0.83, 1.18) |  | 6.64 (6.11, 7.22)  1.41 (1.30, 1.53) | 0.02 (-0.22, 0.27) |
| Siblings | 1.08 (0.91, 1.28)  1.15 (0.97, 1.36) |  | 6.37 (5.64, 7.18)  1.31 (1.16, 1.47) | -0.19 (-0.40, 0.04) |
| Daycare Attendance\* | 1.76 (1.47, 2.11)  0.72 (0.60, 0.86) |  | 4.25 (3.67, 4.93)  1.72 (1.49, 1.99) | 0.48 (0.20, 0.77) |
| HBoV-1\* | 1.11 (0.90, 1.36)  0.89 (0.72, 1.09) |  | 6.47 (5.89, 7.11)  1.44 (1.32, 1.58) | 0.16 (-0.02, 0.38) |
| Playgroup Activity\* | 1.24 (1.05, 1.47)  1.00 (0.85, 1.18) |  | 5.84 (5.15, 6.63)  1.41 (1.24, 1.59) | -0.01 (-0.18, 0.20) |

Table 3. Univariate markov models assessing ARI free and ARI incident states between (n = 86) neonates followed from birth up to 2 years.

Abbreviations: HR, hazard ratio; CI, confidence interval; ARI, acute respiratory infection; HBoV-1, human bocavirus type 1

qrs indicates transition from state r to s (1: ARI (-), 2: (ARI (+)). i.e., q12 indicates transition from ARI free to ARI episode state, q22 indicates sojourn times in ARI episode states.

\* Modelled as time-varying covariates

† Confidence interval calculated through non-parametric bootstrap procedure.

‡ Sojourn time differences in ARI episodes between levels of covariates, e.g. between females and males.



**Markov model output assessing acute respiratory infection (ARI) in (n = 86) neonates from birth to 1 year of life.** (A) Estimated transition probability by daycare attendance. is the probability of occupying state *s* at time *t + u* conditionally on occupying state *r* at time *u*. ARI (-) – ARI (+) denotes the transition from an ARI free state to an ARI episode. (B) Hazard ratios and 95% confidence intervals from 5 different models, daycare attendance alone, daycare vs. female sex, daycare vs. household siblings, daycare vs. HBoV-1 shedding, and daycare vs. playgroup activity. Grey block denotes the model with daycare alone as the covariate. (C) Estimated sojourn (wait) time in weeks in ARI episodes, displayed are point estimates and non-parametric bootstrap 95% confidence intervals.



**Structural changepoint analysis of acute respiratory infection (ARI), before and during daycare attendance.** Estimates generated from bayesian mixed-effects logistic regression with daycare as the changepoint. Time has been standardized to time of entry into daycare. (A) Predictions from the generated model computed with random slope and intercept, time 0 is the time of daycare introduction. (B:E) Analysis stratified by covariates of interest, displayed are predictions from the model with 95% credible intervals.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | Est. | Est. Error | OR (exp(Est.)) | 95% CrI |
| Intercept\* | -1.493 | 0.211 | 0.225 | 0.148 – 0.339 |
| Daycare start time\* | 0.009 | 0.005 | 1.001 | 0.999 – 1.020 |
| Time before daycare\* | 0.056 | 0.010 | 1.057 | 1.038 – 1.079 |
| Time during daycare\* | 0.003 | 0.006 | 1.003 | 0.991 – 1.013 |
| Female | -0.020 | 0.238 | 0.980 | 0.606 – 1.554 |
| Siblings | -0.217 | 0.252 | 0.805 | 1.038 – 1.080 |
| HBoV-1 Positivity† | 0.193 | 0.252 | 1.212 | 0.750 – 2.013 |
| Playgroup Activity† | 0.425 | 0.176 | 1.530 | 1.090 – 2.170 |

Table 4. Fixed effects generated from logistic mixed effects regression modelling ARI incidence.

Abbreviations. Est., coefficient estimate; Est. Error, estimated coefficient error; OR, odds ratio; CrI, credible interval; HBoV-1, human bocavirus type 1

\* Terms included in all models (Female, Siblings, HBoV-1, Playgroup). Estimates and errors vary little, so they are omitted from the table for brevity.

† Modelled as time-dependent covariates.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Model | HR  (95% CI) | Estimated Q-matrix  (95% CI) | Sojourn Time (Weeks)  (95% CI) | Mean Sojourn Difference in ARI episodes  (95% CI)†  ‡ |
| Female | 0.90 (0.70, 1.16)  1.01 (0.78, 1.30) |  | 5.97 (4.87, 7.33)  1.63 (1.33, 2.00) | -0.02 (-0.46, 0.49) |
| Siblings | 0.85 (0.67, 1.09)  1.30 (1.02, 1.65) |  | 5.98 (5.02, 7.14)  1.43 (1.20, 1.70) | -0.43 (-0.85, -0.02) |
| Daycare Attendance\* | 1.93 (1.48, 2.52)  0.80 (0.61, 1.04) |  | 5.61 (4.94, 6.36)  1.56 (1.38, 1.77) | 0.34 (0.03, 0.68) |
| HBoV-1\* | 0.88 (0.67, 1.17)  0.68 (0.52, 0.90) |  | 5.73 (4.97, 6.61)  1.80 (1.56, 2.08) | 0.58 (0.26, 0.91) |
| Playgroup Activity\* | 1.07 (0.83, 1.39)  1.02 (0.79, 1.32) |  | 5.32 (4.30, 6.58)  1.62 (1.32, 1.99) | -0.04 (-0.44, 0.4) |

Table X. Subset Analysis: Univariate markov models assessing ARI free and ARI incident states between (n = 34) neonates followed from birth up to 2 years.

Abbreviations: HR, hazard ratio; CI, confidence interval; ARI, acute respiratory infection; HBoV-1, human bocavirus type 1

qrs indicates transition from state r to s (1: ARI (-), 2: (ARI (+)). i.e., q12 indicates transition from ARI free to ARI episode state, q22 indicates sojourn times in ARI episode states.

\* Modelled as time-varying covariates

† Confidence interval calculated through non-parametric bootstrap procedure.

‡ Sojourn time differences in ARI episodes between levels of covariates, e.g. between females and males.