Supplemental Information

SUPPLEMENTARY METHODS

Surveillance Methods

We identified eligible AI/AN children < 5 years old via facility-based screening and enrolled via parental informed consent. Study staff conducted on-site surveillance and enrollment 5 to 6 days per week; potential participants admitted during hours when staff were not on site were contacted for enrollment at the earliest opportunity. We documented all potentially eligible individuals, including those discharged or transferred before study staff could make contact. Patients with a nonrespiratory primary cause for hospitalization were excluded.

Vanderbilt University Medical Center PCR Assays for Respiratory Viral Detection

Total nucleic acid (TNA) extracts (100 mL TNA extracted from 100 mL specimen) were prepared from respiratory specimens using the MagNA Pure LC Total Nucleic Acid Isolation Kit and MagNA PURE LC 2.0 automated extraction platform (Roche). Extracts were tested by RT-qPCR for viral pathogen targets (see below) using StepOnePlus, QuantStudio 3, and QuantStudio 6-Flex real-time PCR systems (Applied Biosystems).

RT-qPCR for respiratory syncytial virus universal and A and B antigenic groups was performed with the Superscript III Platinum One-Step Quantitative RT-PCR System with ROX passive reference dye (Invitrogen). All extracts were additionally tested for RNAse P (RNP) as an endogenous indicator of specimen quality. A pathogen-free human specimen control was included with each extraction as an indicator of extraction success and sentinel of contamination. Each PCR plate contained positive controls corresponding to tested targets and negative (water) blanks to detect cross-contamination. RNP-negative specimens were retested in duplicate using the original TNA extract, followed by duplicate testing of a fresh extract in the event of persistently undetectable RNP. Detection of RNP in the second extract triggered retesting for all pathogen targets. Any level of pathogen target detection accompanied by a characteristic exponential amplification curve was deemed positive.

Virus Target	F Primer (5′–3′)	R Primer (5′–3′)	Probe (5′–3′)	Source	Reference
Respiratory syncytial virus, universal	GGCAAATAT GGAAACATA CGTGAA	TCTTTTTCT AGGACATTG TAYTGAACA G	FAM-CTGTGT ATGTGGAGC CTTCGTGAA GCT-BHQ-1	ThermoFisher (primers); biosearch (probe)	https://journals.asm.org/doi/10.1128/JCM.02270-10
Respiratory syncytial virus, A	AATACAGCCAAATCTAACCAACTTTACA	GCCAAGGAAGCATGCAATAAA	FAM-TGCTATTGTGCACTAAAG-MGBNFQ	ThermoFisher	Suman Das Laboratory, Vanderbilt University Medical Center
Respiratory syncytial virus, B	AATACAGCCAAATCTAACCAACTTTACA	GCCAAGGAAGCATGCAATAAA	VIC-CACTATTCCTTACTAAAGATGTC-MGBNFQ	ThermoFisher	Suman Das Laboratory, Vanderbilt University Medical Center

BHQ, black hole quencher; FAM, VIC, fluorophores; MGBNFQ, minor groove binder nonfluorescent quencher.

DISCORDANT SPECIMEN RESULTS BETWEEN STUDY SPECIMEN AND CLINICAL SPECIMEN

For incidence and cumulative incidence calculations of RSV-associated ARI, individuals with either a study specimen positive for RSV or a clinical specimen positive for RSV were considered RSV positive. Study-specific mid--turbinate swabs were available for 163 (96.4%) children enrolled in Alaska and 151 (97.4%) in the Southwest; all participants without a study-specific swab had a clinical RSV PCR result available.

We compared PCR results between study swabs tested by Vanderbilt and clinical swabs tested at IHS facilities for children < 5 who had both swabs collected. For the Southwest sites, we found a discordance rate of 10.4% for RSV (12 of 115 tests). Nine of the 12 discordant RSV results were negative by the clinical test and positive by the study test. For the Alaska sites, we found a discordance rate of 8.3% for RSV (12 of 145 tests). Three of the 12 discordant RSV results were negative by the clinical test and positive by the study test.

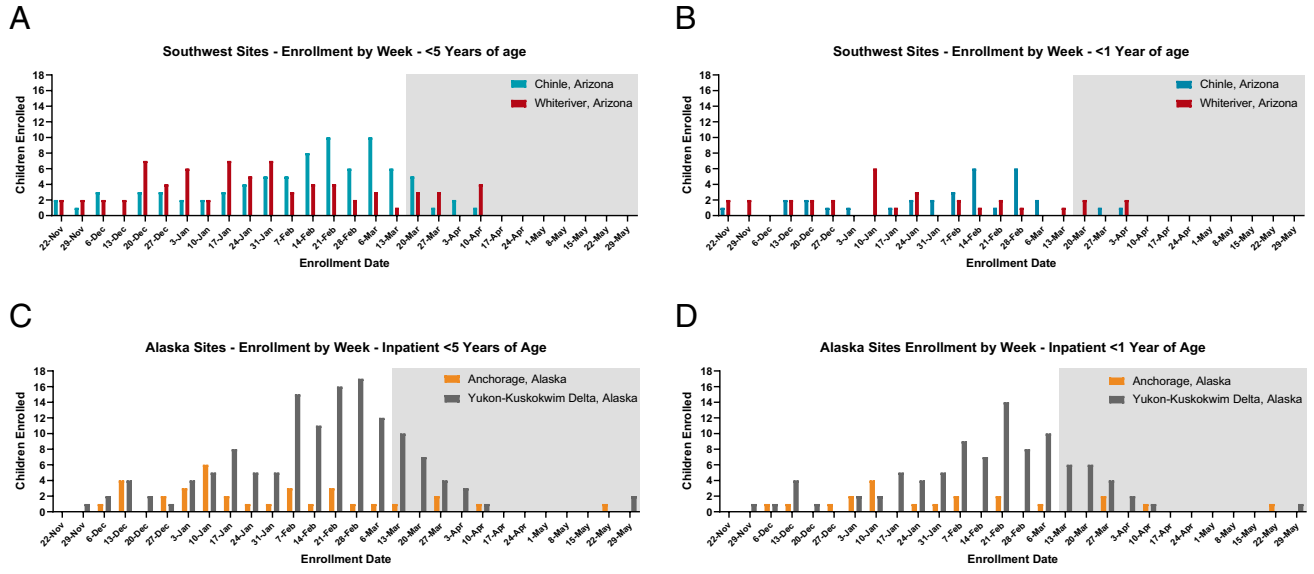
Careful review of these discrepancies did not identify any relationships between discordant results and month of collection, study staff member who collected or processed the specimens, time between clinical swab collection and VUMC swab collection, transport conditions, storage conditions, or any other discernable factors.

Additional Incidence Rate Methods

Numerators of enrolled children meeting the RSV ARI case definition were adjusted by age group to account for eligible but not enrolled cases, under the assumption that the proportion positive among enrolled individuals would be the same as the proportion positive among individuals not enrolled (Supplemental Table 2). Incidence rates were calculated as annual rates under the assumption that all respiratory cases for the year occurred during the enrollment period, which coincided with the typical respiratory season.

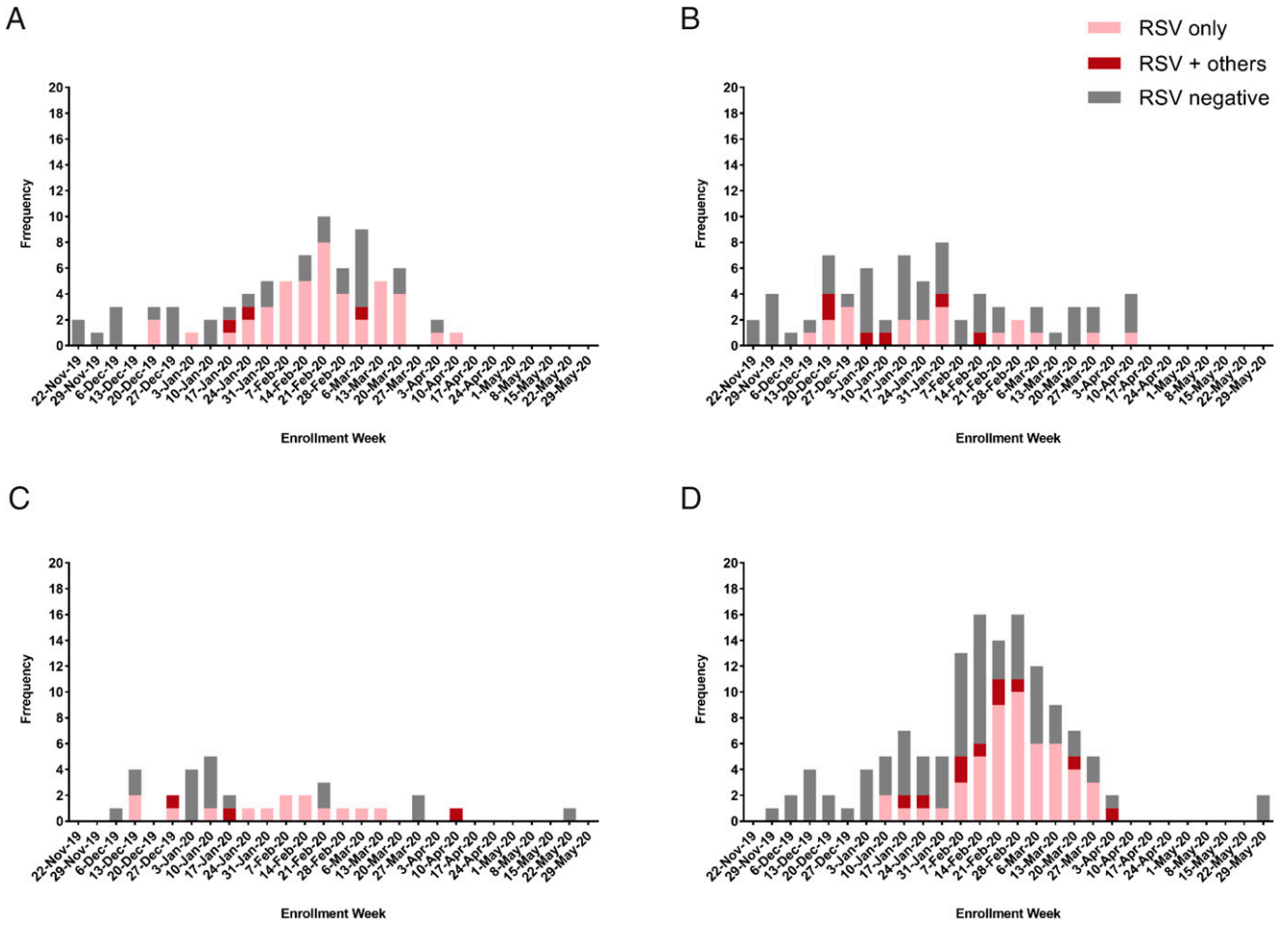
Ethics Approvals

This study was reviewed and approved by the Johns Hopkins Bloomberg School of Public Health IRB (No. 00009605), and the ethics review board for each participating Tribal population (Southwest sites: Navajo Nation Human Research Review Board [NNR-19.350] and Phoenix Area IHS IRB [19.08]; Alaska sites: Alaska Area IRB [2019-05-040-29], Alaska Native Tribal Health Consortium [2019-05-040-29], Southcentral Foundation [2019-05-040-29], and Yukon-Kuskokwim Health Corporation [19.06.01]).



SUPPLEMENTAL FIGURE 2

Enrollment by week by region and age, November 2019 to May 2020. (A) Enrollment of participants < 5 years of age for Southwest sites, Chinle, Arizona and Whiteriver, Arizona. (B) Enrollment of participants < 1 year of age for Southwest sites, Chinle, Arizona and Whiteriver, Arizona. (C) Enrollment of participants < 5 years of age in Alaska sites, Anchorage, Alaska and Yukon-Kuskokwim Delta, Alaska. (D) Enrollment of participants < 1 year of age for Alaska sites, Anchorage, Alaska and Yukon-Kuskokwim Delta, Alaska. The grey box designates the period of time when COVID-19 cases were circulating in the region.



SUPPLEMENTAL FIGURE 3

Detection of respiratory viruses by PCR among AI/AN children < 5 years of age hospitalized with acute respiratory infection (ARI), November 2019 to May 2020. (A) Chinle, Arizona, (B) Whiteriver, Arizona, (C) Anchorage, Alaska, (D) Yukon-Kuskokwim Delta, Alaska.

SUPPLEMENTAL TABLE 2 Crude and Adjusted Number of Cases of All-cause ARI and RSV-associated ARI Hospitalizations Among AI/AN Children < 5 y of Age, November 2019 to May 2020

	Chinle			Whiteriver			Anchorage			Yukon-Kuskokwim Delta		
Age, mo	Crude All-cause ARI	Crude RSV-ARI	Adjusted RSV-ARI[a]	Crude All-cause ARI	Crude RSV-ARI	Adjusted RSV-ARI[a]	Crude All-cause ARI	Crude RSV-ARI	Adjusted RSV-ARI[a]	Crude All-cause ARI	Crude RSV-ARI	Adjusted RSV-ARI[a]
0–2	10	4	8	11	2	2	16	7	8	44	16	23
3–5	12	8	9	7	4	5	6	3	3	35	14	20
0–5	22	12	17	18	6	7	22	10	11	79	30	43
6–11	21	10	13	18	8	11	3	0	0	46	22	29
0–11	43	22	30	36	14	18	25	10	11	125	52	72
12–23	36	15	20	23	11	12	13	8	11	43	11	18
24–35	19	7	9	14	4	5	5	0	0	17	5	8
36–47	6	3	4	6	2	4	1	0	0	5	1	1
48–59	3	3	3	3	0	0	4	1	4	2	1	2
24–59	28	13	16	23	6	9	10	1	4	24	7	11
0–59	107	50	66	82	31	39	48	19	26	192	70	101

[a] Adjusted to account for eligible but not recruited children by applying the proportion of enrolled children who met the case definition divided by total enrolled children to the total documented ARI admissions (enrolled plus eligible but nonenrolled cases) to estimate the adjusted number of children experiencing ARI by site.

SUPPLEMENTAL TABLE 3 Characteristics of AI/AN Children < 5 y of Age Hospitalized With All-cause ARI by Site, November 2019 to May 2020

	Chinle, Arizona Total (*N* = 82)		Whiteriver, Arizona Total (*N* = 73)		Anchorage, Alaska Total (*N* = 33)		Yukon-Kuskokwim Delta, Alaska Total (*N* = 136)	
Variable	**Mean (SD)**	**Range**	**Mean (SD)**	**Range**	**Mean (SD)**	**Range**	**Mean (SD)**	**Range**
Age in months	17.6 (12.7)	0–54	15.9 (12.1)	0–50	10.1 (12.2)	0–54	10.7 (10.7)	0–56
Gestational age at birth in weeks	37.9 (2.4)	27–41	37.0 (3.4)	25–41	37.8 (2.1)	31–41	36.1 (4.2)	24–41
Birth weight in grams	3211.0 (666.7)	828–5150	2819.4 (784.9)	970–4117	3108.7 (701.5)	1370–4280	3024.7 (961.5)	700–5290
Household density (no. persons in home per room)	1.8 (1.6)	0.5–12.0	1.4 (0.7)	0.2–3.7	1.5 (0.9)	0.7–5.0	2.4 (1.6)	0.5–8.5
No. of children < 5 y in home	1.6 (0.8)	0–4	1.6 (1.1)	0–5	1.7 (0.8)	0–4	1.4 (1.1)	0–5
	N	% (95% CI)	*N*	% (95% CI)	*N*	% (95% CI)	*N*	% (95% CI)
Male	52	63.4 (52.3–73.2)	29	39.7 (29.0–51.5)	17	51.5 (34.3–68.4)	81	59.6 (51.0–67.5)
Age group (months)								
0 to <3	5	6.1 (2.5–14.0)	9	12.3 (6.5–22.2)	14	42.4 (26.4–60.2)	30	22.1 (15.8–29.9)
3 to <6	10	12.2 (6.6–21.4)	5	6.8 (2.8–15.6)	5	15.2 (6.2–32.4)	25	18.4 (12.7–25.9)
6 to <12	17	20.7 (13.2–31.0)	18	24.7 (16.0–36.0)	1	3.0 (0.4–19.8)	36	26.5 (19.7–34.6)
12 to <24	28	34.1 (24.6–45.2)	24	32.9 (23.0–44.6)	9	27.3 (14.5–45.4)	29	21.3 (15.2–29.1)
24 to <36	14	17.1 (10.3–27.0)	12	16.4 (9.5–27.0)	3	9.1 (2.8–25.6)	10	7.4 (4.0–13.2)
36 to <48	5	6.1 (2.5–14.0)	3	4.1 (1.3–12.2)	0	3.0 (0.4–19.8)	5	3.7 (1.5–8.6)
48 to <60	3	3.7 (1.2–10.9)	2	2.7 (0.7–10.5)	1	3.0 (0.4–19.8)	1	0.7 (0.1–5.1)
Received influenza vaccine if ≥6 mo at enrollment	54	80.6 (69.1–88.5)	31	52.5 (39.6–65.1)	10	66.7 (38.2–86.6)	37	46.3 (35.5–57.3)
≥1 dose of PCV13 and/or DTaP vaccine if ≥2 mo at enrollment	77	97.5 (90.2–99.3)	66	100.0	21	100.0	100	84.7 (77.0–90.2)
Chronic cough	8	9.8 (4.9–18.5)	9	12.3 (6.5–22.2)	3	9.1 (2.8–25.6)	17	12.5 (7.9–19.3)
Chronic wheeze	1	1.2 (0.2–8.4)	2	2.7 (0.7–10.5)	1	3.0 (0.4–19.8)	8	5.9 (3.0–11.4)
Chronic shortness of breath	1	1.2 (0.2–8.4)	3	4.1 (1.3–12.2)	1	3.0 (0.4–19.8)	4	2.9 (1.1–7.6)
Uses supplemental oxygen at home	2	2.4 (0.6–9.4)	3	4.1 (1.3–12.2)	2	3.0 (0.4–19.8)	5	2.9 (1.1–7.6)
Underlying medical condition[a]								
Any	44	53.7 (42.7–64.3)	29	39.7 (29.0–51.5)	27	81.8 (64.2–91.9)	97	73.5 (65.2–80.4)
Prematurity[b]	9	11.0 (5.8–20.2)	21	28.8 (19.4–40.3)	10	30.3 (16.7–48.5)	37	27.2 (20.3–35.4)
Recurrent wheeze or reactive airway disease or asthma	9	11.0 (5.8–19.9)	0	0.0	7	21.2 (10.2–39.1)	19	14.0 (9.1–20.9)
Chronic lung disease	2	2.4 (0.6–9.4)	0	0.0	1	3.0 (0.4–19.8)	23	16.9 (11.5–24.2)
Congenital heart disease	9	11.0 (5.8–19.9)	7	9.6 (4.6–19.0)	1	3.0 (0.4–19.8)	13	9.6 (5.6–15.8)
Obesity[c]	5	6.1 (2.5–14.0)	0	0.0	1	3.0 (0.4–19.8)	0	0.0
Gastroesophageal reflux disease	0	0.0	5	6.8 (2.8–15.6)	0	0.0	2	1.5 (0.4–5.8)
Developmental delay	18	22.0 (14.2–32.4)	13	17.8 (10.5–28.5)	2	6.1 (1.4–22.2)	11	8.1 (4.5–14.1)
Seizure disorder	4	4.9 (1.8–12.5)	0	0.0	1	3.0 (0.4–19.8)	7	5.1 (2.5–10.5)
Other neurologic or neuro-muscular disorder	0	0.0	0	0.0	1	3.0 (0.4–19.8)	6	4.4 (2.0–9.5)

SUPPLEMENTAL TABLE 3 Continued

Variable	Chinle, Arizona Total (*N* = 82)		Whiteriver, Arizona Total (*N* = 73)		Anchorage, Alaska Total (*N* = 33)		Yukon-Kuskokwim Delta, Alaska Total (*N* = 136)	
	Mean (SD)	Range	Mean (SD)	Range	Mean (SD)	Range	Mean (SD)	Range
Genetic or metabolic disorder[d]	3	3.7 (1.2–10.9)	1	1.4 (0.2–9.4)	0	0.0	3	2.2 (0.7–6.7)
Other	16	19.5 (12.2–29.7)	18	24.7 (16.0–36.0)	22	66.7 (48.5–80.9)	65	47.8 (39.5–56.2)
Previous hospitalization for respiratory illness	31	37.8 (27.9–48.9)	39	53.4 (41.8–64.7)	7	21.2 (10.2–39.1)	46	33.8 (26.3–42.2)
No. of times (mean, SD) range	1.3 (0.6)	1–3	2.1 (1.2)	1–5	3.1 (3.5)	1–10	1.5 (0.8)	0–4
Running water in the home	65	79.3 (69.0–86.8)	72	98.6 (90.6–99.8)	31	93.9 (77.8–98.6)	88	64.7 (56.2–72.3)
Tobacco smoker in the home	5	6.1 (2.5–14.0)	10	13.7 (7.5–23.8)	12	36.4 (21.5–54.4)	54	39.7 (31.8–48.2)
Maternal education of high school or less	50	61.0 (49.9–71.0)	62	84.9 (74.6–91.5)	26	78.8 (60.9–89.8)	114	83.8 (76.6–89.1)
Fuel used most frequently to heat home[e]								
Improved	22	26.8 (18.3–37.6)	19	26.0 (17.1–37.5)	26	78.8 (60.9–89.8)	109	80.1 (72.5–86.1)
Not improved	59	72.0 (61.1–80.7)	53	72.6 (61.1–81.7)	0	0.0	16	11.8 (7.3–18.4)
Other	1	1.2 (0.2–8.4)	1	1.4 (0.2–9.4)	2	6.1 (1.4–22.2)	9	6.6 (3.5–12.3)
Missing	0	0.0	0	0.0	5	15.2 (6.2–32.4)	2	1.5 (0.4–5.8)
Exhaust vent present if heating source is not improved	57	96.6 (87.1–99.2)	44	83.0 (70.1–91.1)	NA	NA	14	87.5 (58.3–97.2)
For children <3 y old	Total (*n* = 74)		Total (*n* = 68)		Total (*n* = 32)		Total (*n* = 130)	
Breastfeeding history								
Ever breastfed	54	73.0 (61.6–82.0)	45	66.2 (54.0–76.5)	28	87.5 (70.2–95.4)	90	69.2 (60.7–76.6)
Currently breastfeeding	23	31.1 (21.5–42.7)	13	19.1 (11.3–30.4)	12	37.5 (22.2–55.8)	38	29.2 (22.0–37.7)
Received Palivizumab during the months of Sept 2019–May 2020	1	1.4 (0.2–9.2)	2	2.9 (0.7–11.3)	0	0.0	16	12.3 (7.6–19.2)

NA, not available.

[a] No enrolled individuals had HIV or AIDS, active TB, immunosuppressed, bone marrow or solid organ transplant, indwelling catheter, blood clot or coagulopathy, sickle cell disease, thalassemia, or hemoglobinopathy, goiter or thyroid disease, obstructive sleep apnea, chronic liver disease, chronic endocrine condition, cancer, or history of Guillain-Barre Syndrome. Fewer than 5 enrolled individuals across all sites had Down syndrome, cerebral palsy, heart failure, high blood pressure, stroke or other cerebral disease, chronic kidney disease, or atopic or allergic conditions; therefore, these conditions were not included in the table.

[b] Prematurity was defined as <37 wk gestational age, but in some cases, only a qualitative indication of "premature yes or no" was available (e.g., no specific gestational age in weeks).

[c] Obesity was considered an underlying condition if it was listed as an underlying condition on the patient's medical chart.

[d] Excluding Arctic CPT1a variant.

[e] Improved: electricity, natural gas, LPG, kerosene, stove oil; not improved: pellet, wood, coal, charcoal.

SUPPLEMENTAL TABLE 4 RSV-Associated Hospitalized Children < 3 Years of Age by Gestational Age at Birth and Age Group

Southwest Sites	0–2 mo	3–5 mo	6–11 mo	12–23 mo	24–35 mo
<29 wk, *n* (%)	0 (0)	0 (0)	1 (5.3)	0 (0)	0 (0)
29–31 wk, *n* (%)	0 (0)	0 (0)	2 (10.5)	0 (0)	0 (0)
32–34 wk, *n* (%)	0 (0)	1 (8.3)	1 (5.3)	0 (0)	1 (9.1)
35–36 wk, *n* (%)	0 (0)	1 (8.3)	1 (5.3)	2 (7.7)	0 (0)
≥37 wk, *n* (%)	5 (83.3)	10 (83.3)	12 (63.2)	20 (76.9)	10 (90.9)
Total[a], *n*	6	12	19	26	11
Alaska sites					
<29 wk, *n* (%)	0 (0)	0 (0)	0 (0)	1 (5.3)	1 (20.0)
29–31 wk, *n* (%)	0 (0)	0 (0)	2 (9.1)	1 (5.3)	0 (0)
32–34 wk, *n* (%)	0 (0)	3 (17.6)	0 (0)	1 (5.3)	0 (0)
35–36 wk, *n* (%)	4 (17.4)	1 (5.9)	2 (9.1)	3 (15.8)	0 (0)
≥37 wk, *n* (%)	15 (65.2)	10 (58.8)	9 (40.9)	10 (52.6)	3 (60.0)
Total[b], *n*	23	17	22	19	5

[a] Includes 7 children with unknown gestational age at birth.
[b] Includes 20 children with unknown gestational age at birth.

SUPPLEMENTAL TABLE 5 Incidence Per 1000 of All-cause ARI Hospitalization and RSV-associated Hospitalization Among AI/AN Children < 5 y of Age, November 2019 to May 2020

Age, mo	Chinle, Arizona	Whiteriver, Arizona	Anchorage, Alaska	Yukon-Kuskokwim Delta, Alaska	
All-cause ARI hospitalization					
0–2	94.3 (50.8–175.3)	177.4 (98.3–320.4)	93.6 (57.3–152.7)	268.3 (199.7–360.5)	NA
3–5	113.2 (64.3–199.3)	112.9 (53.8–236.8)	35.1 (15.8–78.1)	213.4 (153.2–297.4)	NA
0–5	103.8 (68.3–157.6)	129.0 (79.0–210.6)	61.4 (40.0–94.2)	237.8 (190.5–296.9)	NA
6–11	98.6 (64.3–151.2)	146.3 (92.2–232.3)	8.8 (2.8–27.2)	140.7 (105.4–187.8)	NA
0–11	101.2 (75.0–136.4)	138.2 (98.8–193.4)	35.1 (23.5–52.3)	186.3 (156.0–222.4)	NA
12–23	75.8 (54.7–105.1)	74.0 (49.1–111.3)	17.5 (10.2–30.2)	62.3 (46.2–84.0)	NA
24–35	39.3 (25.0–61.5)	40.3 (23.9–68.1)	7.0 (2.9–16.7)	26.3 (16.4–42.3)	NA
36–47	11.2 (5.0–24.9)	16.4 (7.4–36.6)	1.2 (0.2–8.6)	8.8 (3.7–21.3)	NA
48–59	5.3 (1.7–16.3)	8.8 (2.8–27.4)	5.4 (2.0–14.3)	3.6 (0.9–14.4)	NA
24–59	17.6 (12.1–25.5)	21.9 (14.5–32.9)	4.4 (2.3–8.1)	13.6 (9.1–20.3)	NA
0–59	42.9 (35.5–51.9)	49.1 (39.4–61.2)	12.6 (9.5–16.8)	60.4 (52.3–69.7)	NA
RSV-associated hospitalization					
Age, mo	**Chinle, Arizona**	**Whiteriver, Arizona**	**Anchorage, Alaska**	**Yukon-Kuskokwim Delta, Alaska**	**NVSN (Curns et al) for comparison**
0–2	75.5 (37.7–150.9)	44.4 (13.6–144.6)	50.4 (25.8–98.2)	141.6 (94.3–212.7)	NA
3–5	90.6 (48.1–170.5)	90.3 (39.5–206.8)	21.1 (7.5–59.1)	122.9 (79.4–190.2)	NA
0–5	83.0 (52.0–132.5)	70.4 (36.3–136.6)	35.7 (20.4–62.6)	132.3 (98.2–178.1)	21.6 (20.0–23.3)
6–11	61.6 (35.9–105.8)	90.1 (50.0–162.3)	0.0 (0.0–10.8)	91.6 (64.0–131.0)	8.2 (7.1–9.3)
0–11	71.8 (50.4–102.4)	80.6 (51.9–125.2)	19.2 (11.2–33.0)	112.2 (89.3–141.0)	14.9 (13.9–16.0)
12–23	42.1 (27.2–65.3)	38.7 (22.0–68.1)	15.6 (8.7–27.7)	26.4 (16.6–41.8)	4.5 (3.9–5.2)
24–35	19.6 (10.4–37.1)	14.7 (6.2–35.0)	0.0 (0.0–5.1)	13.2 (6.7–25.8)	NA
36–47	8.4 (3.3–21.1)	11.0 (4.1–29.2)	0.0 (0.0–4.5)	1.8 (0.2–12.6)	NA
48–59	5.3 (1.7–16.3)	0.0 (0.0–10.9)	5.4 (2.0–14.3)	3.6 (0.9–14.4)	NA
24–59	10.9 (6.8–17.4)	8.2 (4.2–16.0)	1.1 (0.3–3.8)	5.9 (3.2–10.9)	1.2 (1.2–1.5)
0–59	27.2 (21.4–34.4)	25.4 (18.7–34.5)	7.7 (5.3–11.1)	32.7 (26.9–39.7)	4.6 (4.3–4.8)

95 % confidence intervals are in parentheses. ARI, acute respiratory infection; NA, not available; NVSN, New Vaccine Surveillance Network; RSV, respiratory syncytial virus. Incidence of RSV-associated hospitalization in 2019 to 2020 from NVSN, as reported by Curns et al[9] included for comparison.