CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER:

214410Orig2s000
210854Orig1s005,s009

CLINICAL MICROBIOLOGY/ VIROLOGY REVIEW(S)

DIVISION OF ANTIVIRAL PRODUCTS (HFD-530)

VIROLOGY REVIEW

NDA: 214410 / 210854 SE-009 / 210854 SE-005 SDN (EDR): 92 (0091) / 518 (515) / 519 (516)
DATE REVIEWED: 5/12/2022
Virology Reviewer: William Ince, Ph.D.

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Applicant	**Contact**
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NDA/SDN	SDN	EDR	Date Received	Date Assigned	Description	Appendix
214410	089	0089	12/16/2021	12/20/2021	PMR 3961-4 Final Report: CSR for T0835	*NA*
210854	509	0507	12/16/2021	12/20/2021	PMR 3961-4 Final Report: Cross-reference to 214410 SDN 089	*NA*
214410	092	0091	2/16/2022	2/16/2022	Complete Response	*NA*
210854 SE-005	518	0515	2/16/2022	2/16/2022	Pediatric treatment/Cross reference to 214410	*NA*
210854 SE-009	519	0516	2/16/2022	2/16/2022	Pediatric PEP/Cross reference to 214410	*NA*
214410	094	0094	2/23/2022	2/23/2022	Response to IR: T0835 sequence analysis dataset	*NA*
210854 SE-005	522	0519	2/23/2022	2/23/2022	Response to IR: Cross-reference to 214410 SDN 094	*NA*
210854 SE-009	523	0520	2/23/2022	2/23/2022	Response to IR: Cross-reference to 214410 SDN 094	*NA*
214410	095	0095	3/8/2022	3/8/2022	Response to IR: Dataset flag definitions	*NA*
210854 SE-005/MI	525	0522	3/8/2022	3/8/2022	Response to IR: Cross-reference to 214410 SDN 095	*NA*
210854 SE-009/MI	526	0523	3/8/2022	3/8/2022	Response to IR: Cross-reference to 214410 SDN 095	*NA*
214410	111	0111	7/11/2022	7/12/2022	Response to IR: PMC/PMR	C
210854 SE-005/MI	550	0547	7/11/2022	7/12/2022	Response to IR: Cross-reference to 214410 SDN 111	C
210854 SE-009/MI	551	0548	7/11/2022	7/12/2022	Response to IR: Cross-reference to 214410 SDN 111	C
214410	112	0112	7/13/2022	7/14/2022	Response to IR: PMC/PMR	D
210854 SE-005/MI	552	0549	7/13/2022	7/14/2022	Response to IR: Cross-reference to 214410 SDN 112	D
210854 SE-009/MI	553	0550	7/13/2022	7/14/2022	Response to IR: Cross-reference to 214410 SDN 112	D

NA: Not applicable; submission content was reviewed and incorporated into the review body where appropriate.

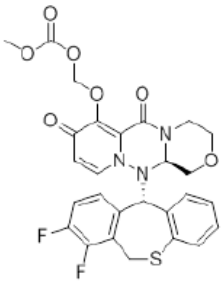
Related/Supporting Documents: IND 126653, Original NDA 210854 (SDN 000)

Product Name(s):

Proprietary Name: XOFLUZA®
Non-Proprietary/USAN: baloxavir marboxil (active metabolite: baloxavir)
Code Name/Number: S-033188 (prodrug), (active metabolite: S-033447 or RSC-033447)

Chemical Name: ({(12aR)-12-[(11S)-7,8-difluoro-6,11-dihydrodibenzo[b,e]thiepin-11-yl]-6,8-dioxo-3,4,6,8,12,12a-hexahydro-1H-[1,4]oxazino[3,4-c]pyrido[2,1-f][1,2,4]triazin-7-yl}oxy)methyl methyl carbonate

Structural formula:



S-033188

DIVISION OF ANTIVIRAL PRODUCTS (HFD-530)

VIROLOGY REVIEW

NDA: 214410 / 210854 SE-009 / 210854 SE-005 SDN (EDR): 92 (0091) / 518 (515) / 519 (516)
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Molecular Formula: $C_{27}H_{23}F_2N_3O_7S$
Molecular Weight: 571.55 Da (482 Da, active metabolite S-033447)
Drug category: Antiviral
Dosage Form(s): Tablet / granules for suspension
Route(s) of Administration: Oral
Indication(s): Treatment and postexposure prophylaxis of acute uncomplicated influenza in patients ≥ 5 years of age.
Dispensed: Rx _X_ **OTC** ___

Abbreviations: CDC, US Centers for Disease Control and Prevention; DDBJ, DNA Databank of Japan; EBI, European Bioinformatics Institute; HH, household; HHC, household contact; INSDC, International Nucleotide Sequence Database Collaboration; IP, index patient; NCBI, US National Center for Biotechnology Information; NIID, Japanese National Institute of Infectious Diseases; OwH, otherwise healthy; PEP, post exposure prophylaxis; RAS, resistance-associated substitution; RIDT, rapid influenza diagnostic test; TE, treatment-emergent; WHO, World Health Organization.

DIVISION OF ANTIVIRAL PRODUCTS (HFD-530)

VIROLOGY REVIEW

NDA: 214410 / 210854 SE-009 / 210854 SE-005 SDN (EDR): 92 (0091) / 518 (515) / 519 (516)
DATE REVIEWED: 5/12/2022
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BACKGROUND

This submission contains the Applicant's response to the Complete Response Letter sent 11/23/2020. Baloxavir marboxil (Xofluza®) was approved 10/24/2018 in the U.S. for the treatment of acute, uncomplicated influenza virus infection in patients 12 years of age and older (N210854.000). Baloxavir marboxil was subsequently approved for the treatment of patients at high-risk of influenza complications (10/16/2019; N210854.S001.077). On 11/23/2020, baloxavir marboxil was approved for postexposure prophylaxis (SE-0010); Integrated Review 214410.000/SE-004/005/009/010). The Applicant also sought to expand approval to the treatment of patients <12 years of age (SE-005), but the efficacy supplement was not approved based on the concern that the elevated frequency of treatment-emergent resistance to baloxavir observed in children could lead to increased transmission of resistant virus (Integrated Review 214410.000/SE-004/005/009/010). In adult/adolescent clinical trials, treatment-emergent resistance has been observed in 4.5% (6/134), 10.9% (53/485), and 0.9% (2/224) of influenza type A/H1N1, A/H3N2, and B virus infections, respectively, whereas in pediatric trials, treatment-emergent resistance was previously observed in 20% (4/20), 27.9% (34/122), and 0% (0/21) of influenza type A/H1N1, A/H3N2, and B virus infections, respectively, as reported in labeling (Integrated Review 214410.000/SE-004/005/009/010). Within each virus type and subtype, frequencies of baloxavir resistance indicated above were relatively consistent between trials conducted over multiple seasons; however, data from an additional trial submitted late in the review cycle for SE-004/5, indicated treatment-emergent resistance frequencies up to 75% among A/H3N2 infected pediatric subjects (study T0835 Integrated Review 214410.000/SE-004/005/009/010). Based on the concern regarding the increased frequencies of treatment-emergent resistance to baloxavir observed in pediatric subjects, a Complete Response Letter was sent 11/23/2020 pertaining to approval for treatment and postexposure prophylaxis (PEP) of patients <12 years of age. The PEP supplement (SDN 005) was split to allow approval for patients ≥12 years of age (assigned SE-010) while denying approval for PEP for patients <12 years of age (assigned SE-009).

The Complete Response Letter indicated the following deficiencies that the Applicant needed to address:

- Submit data from Trial MV40618 entitled, "*A Phase IIIb, Multicenter, Randomized, Double-Blind, Placebo-Controlled, Clinical Efficacy Study of Baloxavir Marboxil for the Reduction of Direct Transmission of Influenza from Otherwise Healthy Patients to Household Contacts*", in order to adequately evaluate the risk of transmission of baloxavir resistant influenza virus from treated index patients, including those less than 12 years of age.

- Additionally, the Applicant should submit the complete clinical study report and datasets from Study T0835 for full evaluation of resistance in that study.

The Applicant subsequently submitted a Type C meeting request to discuss elements of the Applicant's proposed resubmission regarding SE-005 (treatment) and SE-009 (PEP) including the data that would be needed to support an approval for patients aged 5 to <12 years of age in the absence of data from trial MV40618, for which enrollment has been delayed due to the SARS-CoV-2 pandemic. The FDA advised the Applicant that the Agency would consider data supporting approval for baloxavir treatment and postexposure prophylaxis for patients ≥5 years of age in the absence of data from MV40618 and provided an outline of the types of data and analyses that should be submitted to support approval (N214410.000.MP.067 with follow up in N214410.000.079; see also Meeting Minutes dated 8/30/2021). Key information requested to support an indication in subjects 5 to <12 years of age included pooled resistance analyses of all baloxavir trials conducted to date to identify risk factors associated with treatment-emergent resistance to support a potential age cutoff, an assessment of the impact of treatment-emergent resistance on outcomes, an assessment of the

potential for transmission of baloxavir-resistant variants based on trial T0834 (postexposure prophylaxis) and virus shedding modeling, and analyses of current surveillance data.
With their resubmission, the Applicant is seeking the following indications:

Proposed indications:

- SE-005: Treatment of influenza in patients aged 5 years and above who have been symptomatic for no more than 48 hours.
- SE-009: Postexposure prophylaxis of influenza in individuals aged 5 years and above.

Proposed doses:

- <20 kg: 2 mg/kg
(b) (4)

CLINICAL VIROLOGY REVIEW

Trials Supporting the Proposed Indications

Trials used to support the proposed indications and that were used in pooled resistance analyses include data from trials submitted and reviewed previously by the FDA (N210854.000, N210854.S001.077, Integrated Review 214410.000/SE-004/005/009/010) as well as data from completed pediatric trial T0835, the Complete Study Report and datasets for which were submitted to NDA 214410 on December 16, 2021 in fulfillment of PMR 3961-4 (NDA 214410 SDN 089; resistance analysis datasets were requested and subsequently submitted in SDN 094) (Table 1). Interim resistance data for 10 subjects from ongoing pediatric trial CP40559 (MINISTONE-1) were also included in pooled resistance analyses (Table 1).

Table 1: Supporting Clinical Trials

Study and Locations	Study Design	Study Population	Objectives	Dose Regimen	Number of Subjects Randomized	Study Dates (FDA virology reviews of trial data)
1518T0821/ CV40814 (T0821) 72 sites in Japan	Phase 2, multicenter, randomized, placebo- controlled, double-blind study with 4 arms: • Bxm 10 mg • Bxm 20 mg • Bxm 40 mg • Placebo	Otherwise healthy adult subjects (20 to 64 years) with acute uncomplicated influenza	Efficacy, PK, safety and tolerability	Baloxavir (10 and 20 mg tablets): single dose of 10, 20, or 40 mg.	400 subjects randomized (100 per dose group)	December 2015 to April 2016 (N210854.000)
1601T0831/ CV40815 (CAPSTONE-1) 297 sites in Japan, the US, and Canada [a]	Phase 3, multicenter, randomized, placebo / active control, double-blind study with 3 arms: • Baloxavir • Placebo • Oseltamivir (subjects ≥20 years only)	Otherwise healthy adult and adolescent subjects (12 to 64 years and ≥40 kg) with acute uncomplicated influenza	Efficacy, PK, safety and tolerability	Baloxavir (20 mg tablet): single dose by body weight: <80 kg: 40 mg ≥80 kg: 80 mg Oseltamivir: 75 mg BID for 5 days	1436 subjects randomized (baloxavir 612, placebo 310, oseltamivir 514)	December 2016 to April 2017 (N210854.000)

1602T0832/CV40818 (CAPSTONE-2) 551 sites (global)	Phase 3, multicenter, randomized, placebo / active control, double-blind study with 3 arms: Baloxavir Placebo Oseltamivir	Adults and adolescents (≥12 years and ≥40 kg) with acute uncomplicated influenza at high risk of developing influenza complications	Efficacy, PK, safety and tolerability	Baloxavir (20 mg tablet): single dose by body weight: <80 kg: 40 mg ≥80 kg: 80 mg Oseltamivir: 75 mg BID for 5 days	2184 subjects randomized (baloxavir 730, placebo 729, and oseltamivir 725) [b]	January 2017 to April 2018 (N210854.S001.077)
1618T0822/ CV40816 (T0822) 41 sites in Japan	Phase 3, multicenter, open-label, non-controlled study	OwH pediatric subjects (6 months to <12 years) with acute uncomplicated influenza	Safety and tolerability, PK, efficacy	Baloxavir (10 and 20 mg tablets): single dose according to body weight: 5 mg (5 to <10 kg), 10 mg (10 to <20 kg), 20 mg (20 to <40 kg), 40 mg (≥40 kg)	107 subjects received study drug	November 2016 to April 2017 (N210854.000; Integrated Review 214410.000/SE-004/005/009/010)
1705T0833/ CV40964 (T0833) 20 sites in Japan	Phase 3, multicenter, open-label, non-controlled study	OwH pediatric subjects (weighing <20 kg and aged <12 years) with acute uncomplicated influenza	Safety and tolerability, PK, efficacy	Baloxavir (2% granules): single dose according to body weight: <10 kg: 1 mg/kg 10 to <20 kg: 10 mg	33 subjects received study drug	December 2017 to February 2018 (N210854.000; Integrated Review 214410.000/SE-004/005/009/010)
CP40563 (MINISTONE-2) (global)	Phase 3, multicenter, randomized, double–blind, active-controlled study	OwH pediatric subjects (1 to <12 years) with influenza–like symptoms	Safety, PK, efficacy	Baloxavir single dose according to body weight: <20 kg: 2 mg/kg ≥20 kg: 40 mg	173 subjects received study drug (baloxavir 115, oseltamivir 58)	November 2018 to April 2019 (Integrated Review 214410.000/SE-004/005/009/010)
1813T0835/ XV41429 Japan	Phase 3, open-label	OwH pediatric subjects (from birth to <20 kg) with influenza	Safety and tolerability, PK, efficacy	Baloxavir single dose according to body weight: <10 kg and <3 months: 1 mg/kg <10 kg and ≥3 months: 2 mg/kg 10 to <20 kg: 20 mg	45 subjects enrolled.	January 2019 to March 2020 (CSR and ADAM datasets submitted to 214410 SDN 089; resistance datasets submitted to SDN 094; reviewed below)
CP40559 (MINISTONE-1) (global)	Phase 3, multicenter single-arm, open-label study	OwH pediatric subjects (<1 year) with influenza-like symptoms	Safety, PK, efficacy	Baloxavir single dose: <3 months: 1 mg/kg ≥3 months: 2 mg/kg	Planned enrolment: 30 subjects. 42 subjects received study drug to date.	Ongoing (data for 13 subjects were included in some pooled resistance analyses; interim resistance data reviewed in N210854.510)

BID: twice daily; F: female; ITTI: Intention-to-treat Infected; M: male; NA: neuraminidase inhibitor; OwH: otherwise healthy; PK: pharmacokinetic; QD: once daily.
a. Trial T0831 was conducted at sites in Japan, the US, and Canada. However, no patients were enrolled in Canada.
b. The actual number of patients randomized in Study T0832 was 2184 patients, including 2 patients who were randomized twice in error; both patients were re-assigned to the oseltamivir group before dosing. Thus, a total of 2182 unique patients were randomized to treatment.

Supportive Trials Not Previously Reviewed:

1813T0835: *An Open-label Study to Assess the Safety, Tolerability, Pharmacokinetics, and Efficacy of Baloxavir Marboxil 2% Granules after Administration of a Single Dose to Otherwise Healthy Pediatric Patients*

with Influenza. This trial was a non-IND trial conducted exclusively in Japan between January 25, 2019 and March 17, 2020.

Primary endpoints:
- Safety assessments
- PK assessments

Efficacy endpoints (relevant to Virology)
- Time to alleviation of influenza illness
- Longitudinal quantitative evaluations of virus and viral RNA shedding
- Time to cessation of individual symptoms

Inclusion criteria (relevant to Virology)
- Individuals weighing <20 kg and who are <12 years of age with a diagnosis of influenza by fever AND positive RIDT.

Exclusion criteria (relevant to Virology)
- Severe symptoms of influenza virus infection requiring inpatient treatment
- High risk factors for severe disease or complications
- Concurrent infections requiring systemic antimicrobial and/or antiviral therapy at Screening
- Receipt of any influenza antiviral (including baloxavir) within 30 days of Screening
- Exposure to an investigational drug within 30 days or 5 half-lives of the drug prior to Screening.

Study Design: Subjects were dosed with 1 mg/kg if <10 kg and <3 months of age, 2 mg/kg if <10 kg and ≥3 months of age, or 20 mg (flat dose) if weighing 10 to <20 kg. Nasopharyngeal swabs were collected pre-dose at Day 1, and on Days 2, 3/4, 6, and 9. Specimens were also collected at Day 15 and Day 22 if influenza symptoms persisted. Sequencing of the viral PA gene was performed on baseline samples (Day 1) and the last evaluable RT-PCR-positive sample for all subjects in the ITTI population. All virologic assays were carried out by (b) (4) as previously described (Integrated Review 214410.000/S-004/S-010).

Study Population Characteristics: A total of 45 subjects were enrolled and treated, with 43 subjects included in the ITTI population (Table 2). Among those evaluated, type A/H1N1, A/H3N2 and B virus comprised approximately 23% (10/43), 51% (22/43) and 23% (10/43) of infections (1 subject had both A/H1N1 and A/H3N2 virus). There were 35 subjects who were <5 years of age, and among these subjects, 8 were <10 kg and in the 2 mg/kg baloxavir dose group, whereas 27 were ≥10 kg and were in the 20 mg baloxavir dose group. No subjects were <3 months of age (qualifying for the 1 mg/kg dose group), and 8 additional subjects were 5 (n=6) and 6 (n=2) years of age, ≥10 kg, and received a 20 mg dose of baloxavir. Of note, the 8 subjects ≥5 years of age were infected with type A/H1N1 (n=3) or B (n=5), but not A/H3N2, which was dominant in the trial (Table 2); baloxavir has exhibited reduced activity against type B virus (N210854.000; N210854.S001.077; Integrated Review 214410.000/S-004/S-010)

Table 2: Age, Weight, and Virus Type/Subtype at Baseline in Trial T0835

	Total	**Age Cohort**		**Weight Cohort**	
Virus type/subtype		**<5 years**	**≥5 years**	**<10 kg**	**≥10 to <20 kg**
Total	43	35	8	8	35
A/H1N1	10	7	3	1	9
A/H3N2	22	22	0	6	16
B	10	5	5	0	10
A/H1N1+A/H3N2	1	1	0	1	0

Source: FDA analysis of ADSL dataset.

The overall median time to alleviation of illness (in the ITTI population) was 37.8 hours. The median times to alleviation of illness in subjects <5 years of age and ≥5 years of age were 43.98 (n=35) and 25.9 hours (n=8), respectively, and were 26.4 hours and 42.1 hours in the <10 kg (n=8) and 10 to <20 kg (n=35) weight cohorts, respectively (Table 3).

Median change from baseline in virus shedding at Day 2 was -4.4 log_{10} $TCID_{50}$/mL overall, -4.4 and -4.25 log_{10} $TCID_{50}$/mL in the <5 and ≥5 years age cohorts, respectively, and -3.8 and -4.5 log_{10} $TCID_{50}$/mL in the <10 kg and 10 to <20 kg weight cohorts, respectively (Table 3); however, the proportion of subjects who were virus-positive at Day 6 was 59.5% overall, 61.8% and 50% in the <5 and ≥5 year age cohorts, respectively, and 62.5% and 58.8% in the <10 kg and 10 to <20 kg weight cohorts, respectively, after most subjects (76%) were negative at Day 2, consistent with significant reductions in virus shedding followed by significant rates of virus rebound (Table 3; Figure 1).

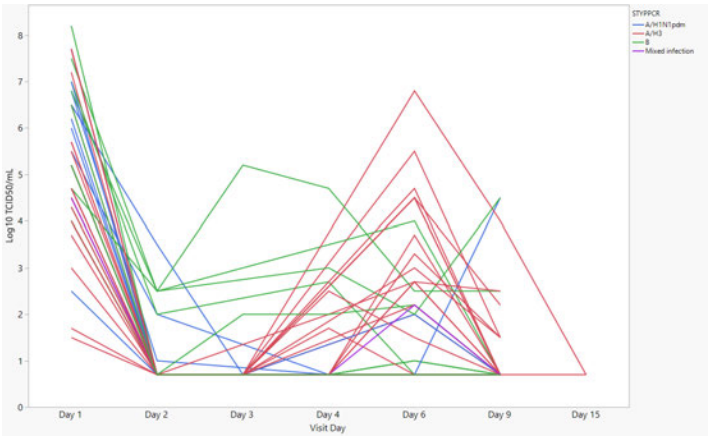
Table 3: Selected Endpoints by Age, Weight, and Virus Type/Subtype in Trial T0835

	Endpoint			
Subset	**Median time to alleviation of illness in hours (N; 95% CI)**	**Median change from baseline in virus shedding at Day 2 log_{10} $TCID_{50}$/mL (N; 95% confidence interval)**	**Percent of subjects who are virus-positive at Day 2 (positive/N)**	**Percent of subjects who are virus-positive at Day 6 (positive/N)**
Overall	37.8 (43; 27.5, 46.7)	-4.4 (42, -5.0, -3.8)	23.8% (10/42)	59.5% (25/42)
<5 years of age	43.98 (35; 29.1-74.1)	-4.4 (34; -5.0, -3.8)	14.7% (5/34)	61.8% (21/34)
≥5 years of age	25.93 (8; 7.9-317.8)	-4.25 (8; -7.5, -1.8)	62.5% (5/8)	50% (4/8)
<10 kg	26.4 (8, 12.1, 51.6)	-3.8 (8; -5.8, -2.3)	0% (0/8)	62.5% (5/8)
10 to <20 kg	42.1 (35; 28.6, 54.8)	-4.5 (34; -5.3, -3.6)	29.4% (10/34)	58.8% (20/34)
A/H1N1	39.98 (10; 24.4, 74.1)	-4.9 (10; -6.3, -3)	50% (5/10)	10% (1/10)
A/H3N2	41.99 (22; 20, 92.6)	-4 (21; -5, -3.3)	4.8% (1/21)	76.2% (16/21)
B	28.83 (10; 10.4, 176.0)	-4.4 (10; -5.8, -3.3)	40% (4/10)	70% (7/10)
A/H1N1+A/H3N2	14.33 (1)	-3.8 (1)	0% (0/1)	100 % (1/1)

Source: FDA analysis. ADTTE, ADLB1.

Figure 1: Virus (A) and viral RNA (B) shedding overall and virus shedding by weight (C) and age (D) in trial T0835. Curve shading indicates virus type/subtype: Blue = A/H1N1, red = A/H3N2, green = B. D) 1 = <10 kg, 2 10 to <20 kg. Source: FDA analysis of ALDB1 dataset.

A



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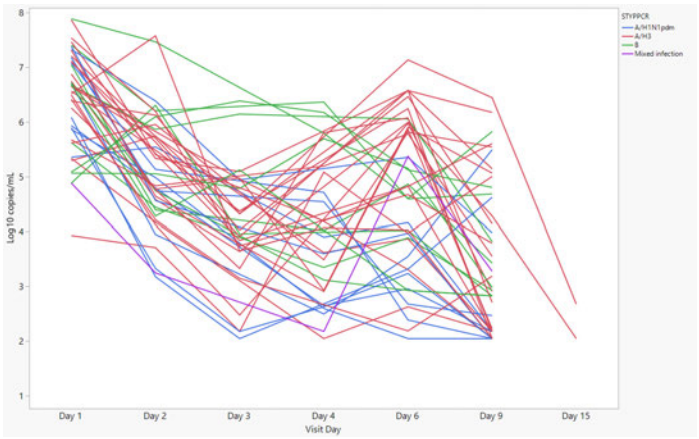

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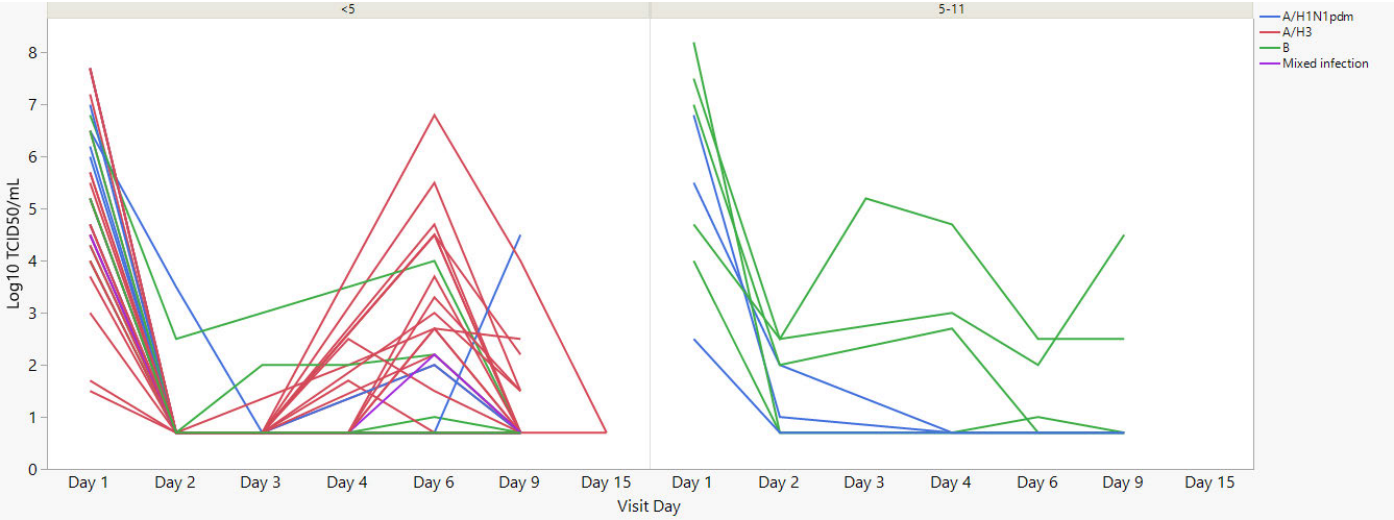

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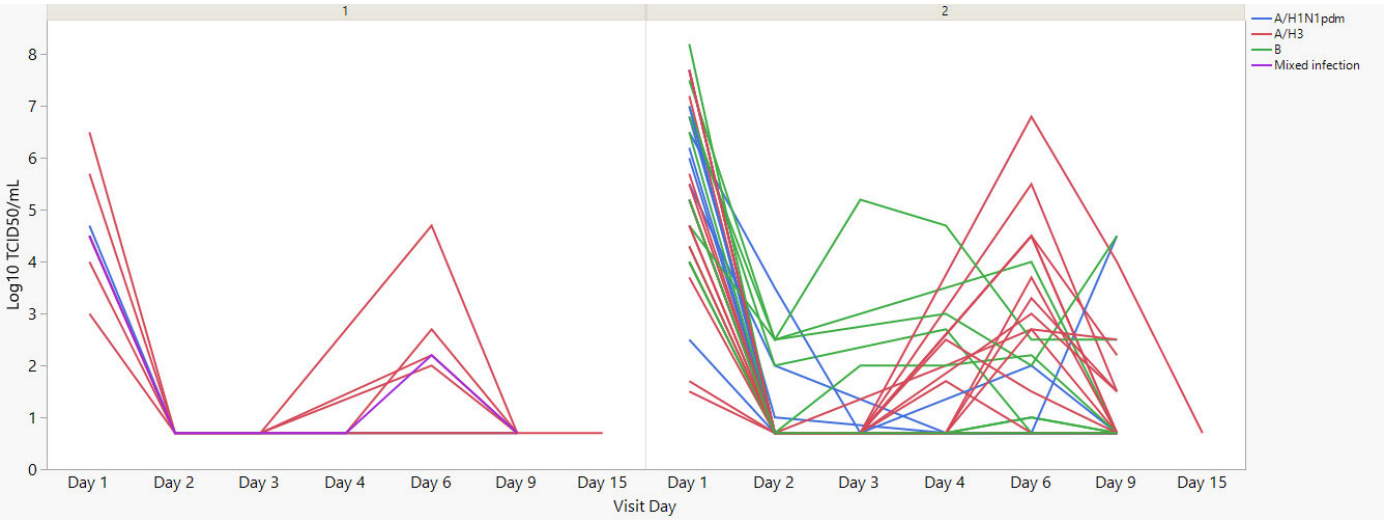


Resistance in Trial T0835

Of the 39 subjects with baseline and post-baseline sequence data, treatment-emergent substitutions were identified in 18 subjects (Table 4). Treatment-emergent resistance-associated substitutions (TE RAS; defined as substitution listed in the baloxavir USPI) were identified in 44% (17/39) of subjects overall, and 20% (2/10), 75% (15/20), and 0% (0/10) of subjects infected with type A/H1N1, A/H3N2, and B influenza virus, respectively (Table 5). Analysis of the frequency of TE RAS by age and weight/dose group trended higher in subjects <5 year of age (Tables 5) and who received a 20 mg flat dose (weighing ≥10 to <20 kg) (Table 6), although the number of subjects was too small to draw a conclusion based on these subsets (see pooled resistance analyses, below). Treatment-emergent resistance was significantly associated with virus rebound; 16 of the 17 subjects with a TE RAS (compared to 7 of 26 subjects without TE RAS) exhibited a rise in virus titer post-baseline after reaching titers at or below the limit of detection (0.7 $\log_{10}$ $TCID_{50}$/mL); (Table 4; Figure 2). In one subject with a mixed A/H1N1+A/H3N2 infection at baseline, only the A/H1N1 virus was able to be sequenced and was likely the majority of the rebound virus; however, no treatment-emergent substitution was identified in this subject (Figure 2).

Table 4: Treatment-Emergent PA Substitutions Observed in Trial T0835

Subject ID [a]	Influenza virus type/subtype	Substitution	Study day detected	Virus rebound?	Phenotypic status [b]
(b) (6)	A/H3N2	I38T/I	8	Yes	RAS
	A/H3N2	I38T	6	Yes	RAS
	A/H3N2	I38T	9	Yes	RAS
	A/H3N2	I38T/I	9	Yes	RAS
	A/H3N2	I38T	8	Yes	RAS
	A/H3N2	I38T	6	No	RAS
	A/H3N2	I38T	8	Yes	RAS
	A/H3N2	L28P/L	5	Yes	<2-fold; (N210854.507_214410.088)
	A/H3N2	I38K/T	5	Yes	RAS (I38T); I38K failed to be rescued; (N210854.507_214410.088)
	A/H3N2	I38M	9	Yes	RAS
	A/H1N1	I38T	10	Yes	RAS
	A/H1N1	A439V	10	Yes	Failed to be rescued; (N210854.507_214410.088)
	A/H3N2	I38T	9	Yes	RAS
	A/H3N2	I38I/M	5	Yes	RAS
	A/H3N2	I38T/I	8	Yes	RAS
	A/H3N2	I38T	7	Yes	RAS
	A/H1N1	A/V231V	7	Yes	Polymorphism; not evaluated in A/H1N1; <2-fold in A/H3N2 (Integrated Review 214410.000/S-004/S-010)
	A/H1N1	F314S/F	7	Yes	Not evaluated; F314 is >99.5% conserved
	A/H3N2	E23K/E	8	Yes	RAS
	A/H3N2	I38T/I	8	Yes	RAS
	A/H3N2	I38T	5	Yes	RAS
	A/H3N2	E23K	8	Yes	RAS

Source: FDA analysis of T0835 datasets.
a. Subjects with multiple substitutions are highlighted.
b. RAS: resistance-associated substitution identified in labeling; source data for substitutions are otherwise in the indicated review.
c. Mixed A/H1N1 and A/H3N2 infection; only A/H1N1 was evaluated.

Comment: A PMR will be proposed to evaluate PA substitution F314S, alone and in combination with A231V, for its impact on baloxavir susceptibility in A/H1N1 virus. This substitution was treatment-emergent in combination with polymorphism A231V in a subject who exhibited virus rebound (GSC004) in trial T0835.

Table 5: Frequency of TE RAS by Age and Virus Type/Subtype in Trial T0835

Age group	% TE RAS (TE RAS subjects/n)			
	A/H1N1	A/H3N2	B	Total
<5 years	28.6% (2/7) [a]	75% (15/20)	0% (0/5)	53% (17/32)
5-11 years	0% (0/2)	0 (0/0)	0% (0/5)	0% (0/7)
Total	22% (2/9)	75% (15/20)	0% (0/10)	44% (17/39)

Source: FDA analysis of T0835 datasets.
a. One subject with both A/H1N1 and A/H3N2 virus in the <5 years group was evaluated for TE RAS in A/H1N1 virus only; no RAS were detected.

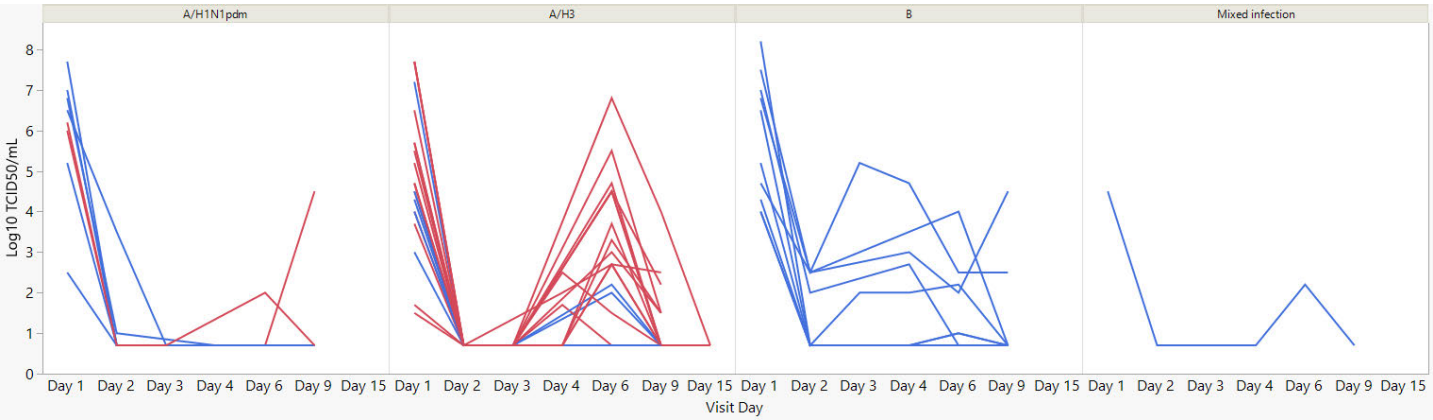
Table 6: Frequency of TE RAS by Dose and Virus Type/Subtype in Trial T0835

Weight-based dose group	% TE RAS (TE RAS subjects/n)			
	A/H1N1	A/H3N2	B	Total
1 mg/kg (<3 m and <10kg)	0% (0/0)	0% (0/0)	0% (0/0)	0% (0/0)
2 mg/kg (≥3 m and <10kg)	0% (0/1) [a]	50% (3/6)	0% (0/0)	42.9% (3/7)
20 mg (≥10 to <20 kg)	25% (2/8)	86% (12/14)	0% (0/10)	43.8% (14/32)

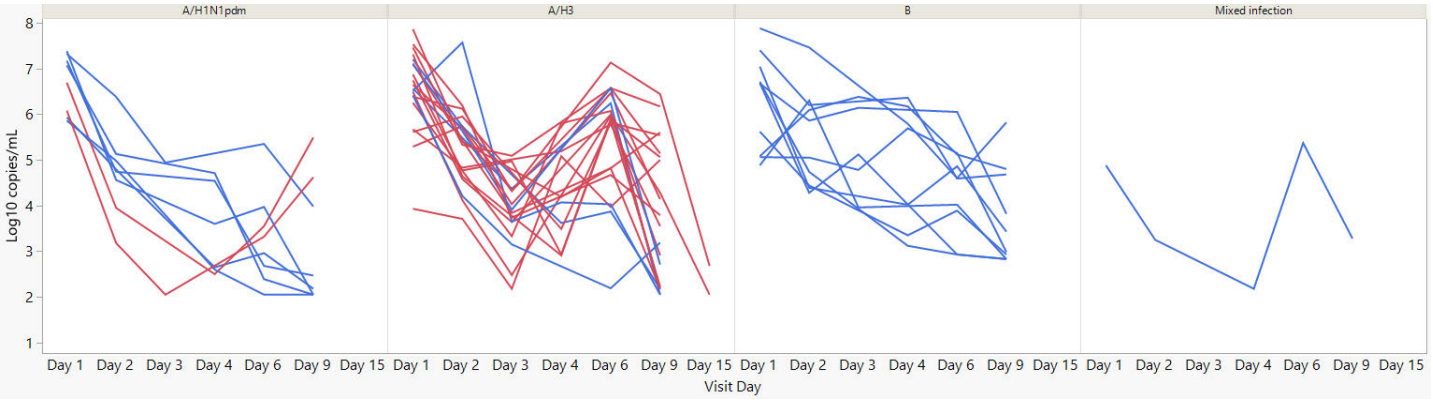
Source: Adapted from Applicant Report 1113741 Appendix 2, Table 3.
a. One subject with both A/H1N1 and A/H3N2 virus in the 2 mg/kg dose group was evaluated for TE RAS in A/H1N1 virus and no RAS were detected.

Figure 2: Virus (A) and viral RNA (B) shedding by virus type/subtype in evaluated subjects with (red curves) and without (blue curves) TE RAS. Source: FDA analysis of ALDB1, ADSL datasets. One subject had a mixed infection of A/H1N1 + A/H3N2 at baseline.

A



B



Analyses of Resistance in Pooled Data from Pediatric and Adult/Adolescent Baloxavir Treatment Trials

Analyses of the frequency, association with baseline variables, and clinical and virologic impacts of TE RAS were carried out on data pooled from all pediatric and adult/adolescent trials submitted to the FDA to date (Table 7). Subjects in the pooled resistance analyses included those in the ITTI populations (confirmed influenza virus-infected by RT-PCR and received a baloxavir marboxil dose) in each study with baseline and post-baseline PA sequence data (unless otherwise noted). TE RAS included all substitutions previously identified in FDA resistance analyses of these trials carried out to date (E23G/K/R, A37T, I38F/M/N/S/T, E199G). A caveat of this analysis approach is the exclusion of subjects without post-baseline sequence data, which appeared to bias the dataset toward subjects who shed virus for longer and who had longer durations of symptoms in most subsets (Figure 3).

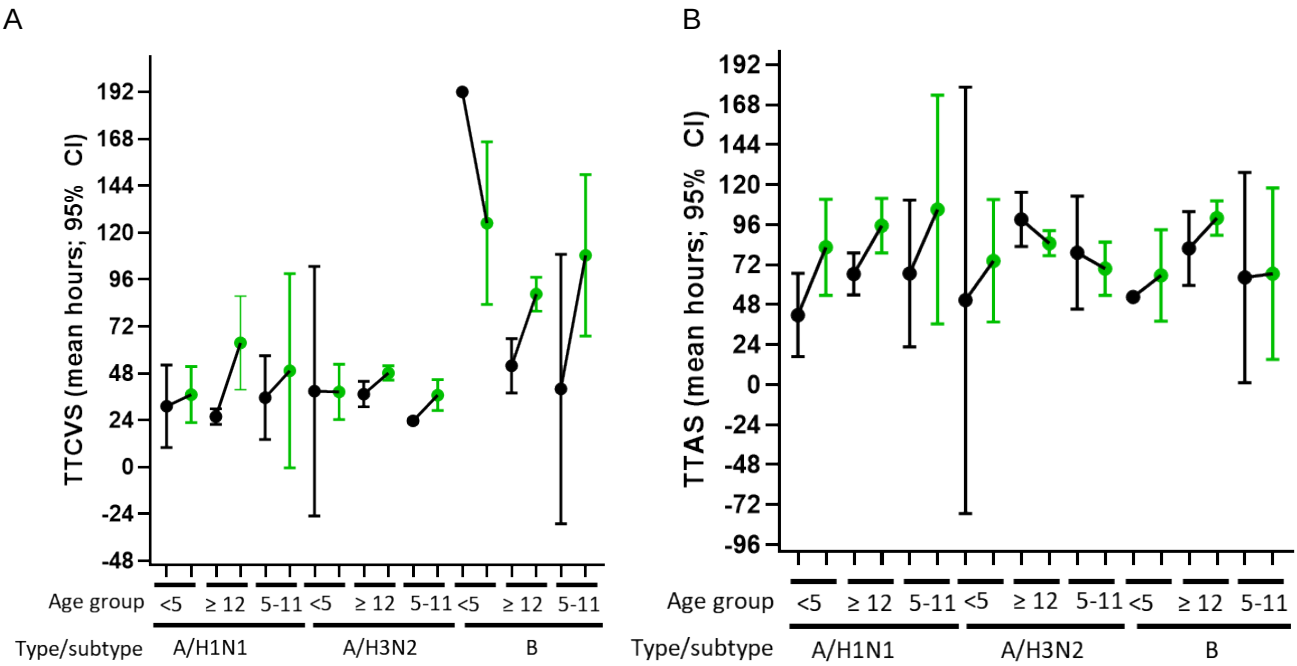
Table 7: Age and Dose Ranges in Trials Included in the Pooled Analyses

Study	Age range (per inclusion criteria)	Actual number (%) of pediatric patients per Age Range treated with baloxavir		Adult/Adolescents ITTI, treated with baloxavir	Dose [a]
		< 5 years	5 to <12 years	≥12 years	

T0831 (CAPSTONE-1) (Global)	12-64 years	-	-	456	• 40 mg: <80 kg • 80 mg: ≥80 kg
T0832 (CAPSTONE-2) (Global) [b]	≥12 years (≥40 kg)	-	-	388	• 40 mg: <80 kg • 80 mg: ≥80 kg
T0821 (Japan)	12-64 years	-	-	300	• 10, 20 or 40 mg (randomized)
T0822 (Japan)	6 months to <12 years	14/104 (13.5%)	90/104 (86.5%)	-	• 5 mg (5 to <10 kg) • 10 mg (10 to <20 kg) • 20 mg (20 to <40 kg) • 40 mg (≥40 kg)
T0833 (Japan)	<12 years (<20 kg)	28/33 (84.8%)	5/33 (15.2%)	-	• 1 mg/kg: <10 kg • 10 mg: 10 to <20 kg
T0835 (Japan)	<6 years (<20 kg)	35/43 (81.4%)	8/43 (18.6%)	-	• 1 mg/kg: <10 kg and <3 months • 2 mg/kg: <10 kg and ≥3 months • 20 mg: 10 to <20 kg
CP40563 (MINISTONE-2) (global)	1 to <12 years	30/124 (24.2%)	94/124 (75.8%)	-	• 2 mg/kg: <20 kg • 40 mg: ≥20 kg
CP40559 (MINISTONE-1) (global, study ongoing)	<1 years	13/13 (100%)	-	-	• 1 mg/kg: <3 months • 2 mg/kg: ≥3 months

Source: Applicant Report 1113741 Table 2.
a. Baloxavir marboxil administered as a single dose according to body weight as specified (except Study T0821).
b. Study included patients with high risk of developing complications.

Figure 3: Time to fist virus-negative sample (TTCVS) (A) and time to alleviation of symptoms (TTAS) (B) in subjects with (green) and without (black) paired baseline and post-baseline sequence data. Subjects with TE RAS were excluded. Source: FDA analysis of pooled ADTTE dataset. Pooled dataset of subjects in the ITTI population treated with baloxavir in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.



Resistance Frequencies Across Clinical Trials
The frequencies of treatment-emergent resistance were variable across adult/adolescent and pediatric studies but are primarily reflective of the age range of subjects enrolled in each study and the representations of influenza virus type and subtype in each study (Table 8); each of these factors is statistically significantly associated with the frequency of treatment-emergent resistance, as discussed below.

Table 8: Frequency of Treatment-Emergent Resistance in Trials Included in the Pooled Analysis

	Virus Type/Subtype Detected in Subject with TE RAS							
Trial ID	**A/H1N1**	**A/H3N2**	**B**	**A/H3N2 + A/H1N1**	**A/H1N1 + B**	**A/H3N2 + B**	**A/UNK+B**	**Total**
Adult/Adolescent Trials								
T0821	4.5% (5/112)	0% (0/14)	0% (0/55)	0% (0/0)	0% (0/1)[a]	0% (0/0)	0% (0/0)	2.7% (5/182)
T0831	0% (0/4)	11.6% (38/327)	0% (0/32)	0% (0/0)	0% (0/0)	25% (1/4)[b]	0% (0/3)[a]	10.5% (39/370)
T0832	5.9% (1/17)	10% (14/140)	0.8% (1/129)	0% (0/0)	0% (0/1)[c]	0% (0/1)[d]	0% (0/2)[a]	5.5% (16/290)
Total (adult/ adolescent)	**4.5% (6/133)**	**10.8% (52/481)**	**0.5% (1/216)**	**0% (0/0)**	**0% (0/2)**	**20% (0/5)**	**0% (0/5)**	7.1% (59/830)
Pediatric Trials								
T0822	0% (0/1)	29.9% (20/67)	0% (0/6)	0% (0/1) [d]	0% (0/0)	0% (0/2)[d]	0% (0/1)[a]	25.6% (20/78)
T0833	33.3% (2/6)	44.4% (4/9)	0% (0/11)	0% (0/0)	0% (0/0)	0% (0/0)	0% (0/0)	23.1% (6/26)
CP40563	16.7% (2/12)	23.3% (10/43)	0% (0/2)	0% (0/0)	0% (0/0)	0% (0/0)	0% (0/0)	21.1% (12/57)
1813T0835	25% (2/8)	75% (15/20)	0% (0/10)	0% (0/1) [c]	0% (0/0)	0% (0/0)	0% (0/0)	43.6% (17/39)
CP40559	20% (1/5)	0% (0/3)	0% (0/2)	0% (0/0)	0% (0/0)	0% (0/0)	0% (0/0)	10% (1/10)
Total (pediatric)	**21.9% (7/32)**	**34.5% (49/142)**	**0% (0/31)**	**0% (0/2)**	**0% (0/0)**	**0% (0/2)**	**0% (0/1)**	27.1% (56/207)
All Trials								
Grand Total	**7.9% (13/165)**	**16.2% (101/623)**	**0.4% (1/247)**	**0% (0/2)**	**0% (0/2)**	**14.3% (1/7)**	**0% (0/6)**	

Source: FDA analysis [pooled ADSL; individual study CSRs]. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.
a. Only type B virus sequenced.
b. Both type A/H3N2 and type B for 1 subject were sequenced in whom both virus types/subtypes harbored an I38T substitution; only A/H3N2 sequenced for 3 subjects other subjects.
c. Only A/H1N1 sequenced.
d. Only A/H3N2 sequenced.

Association of Baseline Characteristics with Treatment-Emergent Resistance
The Applicant evaluated the association of baseline characteristics with TE RAS in the pooled dataset, including data from all trials listed in Table 1, unless otherwise indicated. In univariate analyses carried out separately in subjects ≥12 years of age and in subjects <12 years of age, continuous variables that were analyzed for their association with TE RAS included age, weight, baseline virus titer, and baseline influenza virus hemagglutination inhibition (HI) antibody titer. The test of association with antibody titer was performed separately in subjects infected with type A/H1N1, A/H3N2 and B virus. For influenza virus type B, antibody titers corresponding to B/Victoria and B/Yamagata were analyzed individually. Categorical variables included race (Asian vs non-Asian), influenza virus type (type A vs B), influenza virus type A subtype (A/H1N1 vs A/H3N2), and whether subjects had been vaccinated within 6 months of the start of treatment. In the univariate analysis, the Applicant found that TE RAS was statistically significantly associated with A/H3N2 infection, younger age (for subjects <12 years of age only), lower weight, and lower baseline influenza antibody titer to

the infecting virus (only for A/H3N2, data for other types/subtypes were more limited, but an association cannot be ruled out) (Table 9). Of note, age, weight, and antibody titer were themselves correlated in the dataset; younger aged subjects had lower weight (Figure 4) and lower baseline geometric mean antibody titers against A/H3N2 virus (17.3, 42.5, and 59.8 HI units for <5 year, 5-11 year, and ≥12 year age groups, respectively; FDA reviewer analysis of all subjects in pooled ADSUB dataset); however, even with the <5 year age group, the geometric mean antibody titer against A/H3N2 was lower in subjects with TE RAS (13.4 HI units) compared to those without TE RAS (22.0 HI units) (FDA reviewer analysis of all subjects with baseline and post-baseline sequence data in the pooled ADSUB dataset).

Table 9: Univariate Analyses of the Association of Baseline Variables with TE RAS

	All Adolescent + Adult (≥ 12 yrs)		All Pediatric (<12 yrs)	
Summary statistic / Category	RAS	No RAS	RAS	No RAS
Continuous Variables				
Age (years)				
N	60	782	56	154
Median (95% CI)	43 (32, 53)	40 (39, 42)	3 (3, 4)	6 (5, 7)
p-value[a]	0.4481		0.0057*	
Weight (kg)				
N	60	782	56	154
Median 95% CI	57.65 (54, 66.8)	66.45 (65, 68)	15.5 (14.4, 17.7)	19.9 (18.7, 23)
p-value[a]	0.0004*		0.0045*	
Baseline virus titer (all subtypes) [b]				
N	59	776	56	154
Median 95% CI	6.5 (6, 7.2)	6.2 (6, 6.5)	5.7 (5.2, 6)	5.25 (4.75, 5.7)
p-value[a]	0.1236		0.4249	
Baseline antibody titer (A/H1N1) [b]				
N	6	129	4	13
Median 95% CI	10 (10, 1280)	10 (10, 10)	10 (10, 10)	10 (10, 10)
p-value[a]	0.8119		0.6774	
Baseline antibody titer (A/H3N2) [b]				
N	53	429	39	61
Median 95% CI	40 (20, 80)	57 (40, 80)	10 (10, 20)	40 (20, 40)
p-value[a]	0.03*		<0.0001*	
Baseline antibody titer (B/Yamagata) [b]				
N	2	225	0	30
Median 95% CI	10 (10, 10)	10 (10, 20)	NE	10 (10, 10)
p-value[a]	0.1963		NE	
Baseline antibody titer (B/Victoria) [b]				
N	2	225	0	30
Median 95% CI	10 (10, 10)	10 (10, 20)	NE	10 (10, 10)
p-value[a]	0.2262		NE	
Categorical Variables				
Race, % (n)				
Asian	8.8% (54/616)	91.2% (562/616)	30.6% (44/144)	69.4% (100/144)
Non-Asian	2.7% (6/226)	97.3% (220/226)	18.5% (12/65)	81.5% (53/65)
p-value[c]	0.0014*		0.0910	
Influenza virus type (A or B), % (n)				
Type A	9.4% (58/614)	90.6% (556/614)	31.8% (56/176)	68.2% (120/176)
Type B	0.5% (1/216)	99.5% (215/216)	0	100.0% (31/31)
p-value[c]	<0.0001*		<0.0001*	
Influenza virus type A subtype (A/H1N1 and A/H3N2), % (n) [d]				
A/H1N1	4.5% (6/133)	95.5% (127/133)	21.9% (7/32)	78.1% (25/32)
A/H3N2	10.8% (52/481)	89.2% (429/481)	34.5%(49/142)	65.5% (93/142)
p-value[c]	0.0288*		0.2106	
Vaccination Status, % (n)				
Vaccinated	7.2% (16/223)	92.8% (207/223)	33.3% (24/72)	66.7% (48/72)
Not vaccinated	7.1% (44/619)	92.9% (575/619)	23.2% (32/138)	76.8% (106/138)
p-value[c]	1.0		0.1392	

Source: Applicant Report 1113741 Table 4. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.
a. TE RAS vs no TE RAS, Mann-Whitney U test.

b. Baseline antibody titer data is not collected in studies CP40559 and CP40563.
c. TE RAS vs no TE RAS, Fisher exact test.
d. Denominator for Virus subtype is the number of patients infected with Influenza A virus
*p<0.05 (highlighted)
NE: not evaluable.

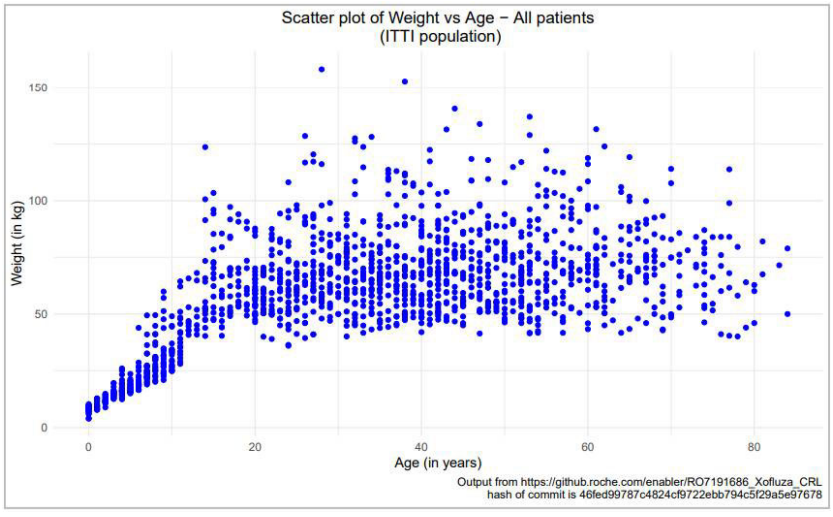
The Applicant also performed multivariate logistic regression modelling of the association of age (years), weight (kg), race (Asian vs non-Asian), dose (mg), vaccination status, baseline virus titer and virus type/subtype with the risk of developing TE RAS. The results of the multivariate analysis were generally consistent with the results of the univariate analyses, with infection with type A/H3N2 virus being the dominant risk factor for TE RAS in the overall pooled population (Table 10). Other factors associated with an increased risk of TE RAS in the overall pooled population included weight and Asian race; however, the Applicant notes that the association with race may be due to lower average weight in Asians compared to non-Asians. Younger age was not associated with increased TE RAS risk in the overall pooled population in this analysis; however, the Applicant concluded this was due to the correlation between age and weight in subjects <12 years of age, which confounded the analysis (Figure 4).

Table 10: Logistic Regression Analysis of Pooled Data from All Trials

Parameter	Estimate	Standard Error	p-value	Odds Ratio		
				Point Estimate	95% LCL	95% UCL
Age	0.0117	0.0075	0.1194	1.012	0.997	1.027
Weight	-0.0553	0.0096	<0.0001*	0.946	0.929	0.964
Race (Non-Asian vs Asian)	-0.6708	0.3276	0.0406*	0.511	0.269	0.972
Vaccination Status (Yes vs No)	0.1729	0.2273	0.4470	1.189	0.761	1.856
Virus subtype (A/H1N1 vs A/H3)	-0.8425	0.3425	0.0139*	0.431	0.220	0.843
Virus subtype (B vs A/H3)	-3.8196	1.0149	0.0002*	0.022	0.003	0.160
Virus subtype (Other vs A/H3)	-1.6034	1.0712	0.1344	0.201	0.025	1.642
Dose (mg)	0.0182	0.0117	0.1204	1.018	0.995	1.042
Baseline Virus Titer	0.0959	0.0676	0.1558	1.101	0.964	1.257

Source: Applicant Report 1113741 Table 5. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.

Figure 4: Plots of weight vs age in pooled dataset. Source: Applicant Report 1113741 Figure 2. Pooled dataset of subjects in the ITTI in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.



To account for observed differences in TE RAS frequencies in pediatric and adults and the correlation between age and weight, subjects <12 and ≥12 years of age, and parameters of age and weight (for subjects <12 years of age) were evaluated in separate models. In subjects <12 years of age, type A/H3N2 infection, younger age, and lower weight were each found to be significantly associated with an increased risk of TE RAS in this model (Tables 11 and 12). In subjects ≥12 years of age, type A/H3N2 infection was also the dominant factor for increased risk of TE RAS, but in contrast to subjects <12 years of age, older age was associated with a slightly increased risk of TE RAS (Table 13).

Interestingly, being vaccinated within 6 months was associated with an increased risk of TE RAS in subjects <12 years of age, which appears to be inconsistent with the association of TE RAS with lower baseline antibody titer but may reflect the consistently low vaccine effectiveness against type A/H3N2 (Belongia et al., 2016; Belongia and Mclean, 2019; Chung et al., 2022), the subtype in which TE RAS are significantly more frequent; consistent with this observation, in the <5-years-of-age group, which exhibited the highest frequency of TE RAS, rates of vaccination were proportionally higher among subjects infected with type A/H3N2 virus compared to subjects infected with type A/H1N1 or type B virus (37% for A/H3N2 vs 25% for A/H1N1 and B combined, respectively; FDA reviewer analysis of subjects with sequence data; ADSL dataset). Geometric mean anti-A/H3N2 antibody titers for unvaccinated and vaccinated subjects were lowest in the <5 year age group (14.8 vs 22.6, respectively), compared to subjects in the 5-11 year (37.9 vs 59.7, respectively) and ≥12 year (44.12 vs 143.26, respectively) age groups (FDA reviewer analysis of all subjects in pooled ADSUB dataset).

Table 11: Logistic Regression Analysis with Age as a Covariate in Subjects <12 Years of Age

Parameter	Estimate	Standard Error	p-value	Odds Ratio		
				Point Estimate	95% LCL	95% UCL
Age	-0.2109	0.0554	0.0001*	0.810	0.726	0.903
Race (Non-Asian vs Asian)	-0.7963	0.4017	0.0475*	0.451	0.205	0.991
Vaccination Status (Yes vs No)	0.8098	0.3643	0.0262*	2.247	1.100	4.590
Virus subtype (Other vs A/H3)	-2.1011	0.4760	<0.0001*	0.122	0.048	0.311

Source: Applicant Report 1113741 Table 6. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.
LCL: lower confidence limit; UCL: upper confidence limit.
*p<0.05.

Table 12: Logistic Regression Analysis with Weight as a Covariate in Subjects <12 Years of Age

Parameter	Estimate	Standard Error	p-value	Odds Ratio		
				Point Estimate	95% LCL	95% UCL
Weight	-0.0735	0.0202	0.0003*	0.929	0.893	0.967
Vaccination Status (Yes vs No)	0.7808	0.3580	0.0292*	2.183	1.082	4.404
Virus subtype (Other vs A/H3)	-2.0532	0.4685	<0.0001*	0.128	0.051	0.321

Source: Applicant Report 1113741 Table 7. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.
LCL: lower confidence limit; UCL: upper confidence limit.
*p<0.05.

Table 13: Logistic Regression Analysis of Subjects ≥12 Years of Age

Parameter	Estimate	Standard Error	p-value	Odds Ratio		
				Point Estimate	95% LCL	95% UCL
Age	0.0177	0.00805	0.0277*	1.018	1.002	1.034
Weight	-0.0330	0.0103	0.0013*	0.968	0.948	0.987
Virus subtype (A/H1N1 vs A/H3)	-1.0657	0.4477	0.01738	0.344	0.143	0.828

Virus subtype (B vs A/H3)	-3.2832	1.0178	0.0013*	0.038	0.005	0.276
Virus subtype (Other vs A/H3)	-0.3361	1.0750	0.7545	0.715	0.087	5.876

Source: Applicant Report 1113741 Table 10. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.
LCL: lower confidence limit; UCL: upper confidence limit.
*p<0.05.

Based on the results of the univariate and multivariate analyses, the frequency of the TE RAS was evaluated across weight and age strata to identify a potential cutoff for risk of TE RAS within influenza virus type/subtype subsets.

TE RAS Frequency by Weight
The frequency of TE RAS was highest in lower weight ranges, with the frequencies peaking in 10 kg to <12.5 kg weight bands across virus type/subtype subsets (Table 14).

Table 14: Proportion of Subjects with TE RAS by Weight and Virus Type/Subtype

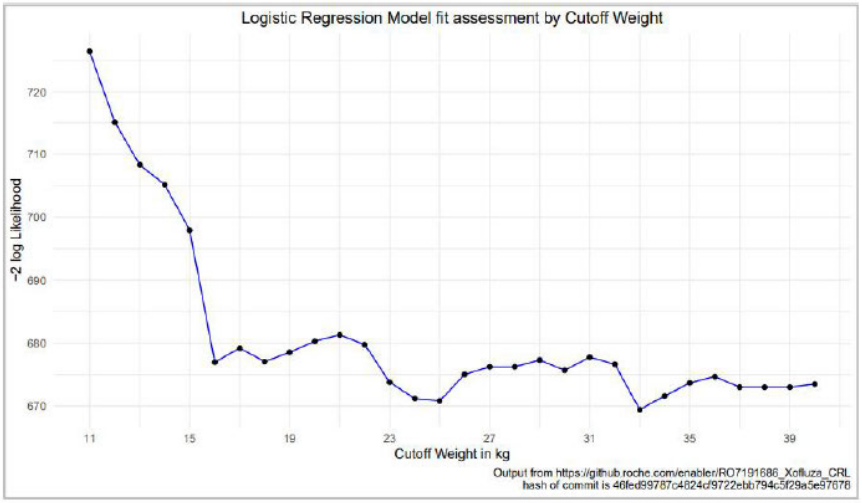
Weight	Total % (n/N) (95% CI)	A/H1N1 % (n/N) (95% CI)	A/H3N2% (n/N) (95% CI)	B % (n/N) (95% CI)
2.5 to <5 kg	50.00% (1/2) (1.26, 98.74)	100% (1/1) (2.50, 100)	0% (0/0) (NE)	0% (0/1) (0.00, 97.50)
5 to <7.5 kg	0% (0/5) (0.00, 52.18)	0% (0/2) (0.00, 84.19)	0% (0/2) (0.00, 84.19)	0% (0/1) (0.00, 97.5)
7.5 to <10 kg	23.81% (5/21) (8.22, 47.17)	20% (1/5) (0.51, 71.64)	36.36% (4/11) (10.93, 69.21)	0% (0/6) (0.00, 45.93)
10 to <12.5 kg	64.29% (9/14) (35.14, 87.24)	50% (1/2) (1.26, 98.74)	72.73% (8/11) (39.03, 93.98)	0% (0/1) (0.00, 97.50)
12.5 to <15 kg	42.11% (8/19) (20.25, 66.50)	33.33% (1/3) (0.84, 90.57)	63.64% (7/11) (30.79, 89.07)	0% (0/5) (0.00, 52.18)
15 to <17.5 kg	35.48% (11/31) (19.23, 54.63)	16.67% (1/6) (0.42, 64.12)	62.5% (10/16) (35.43, 84.80)	0% (0/10) (0.00, 30.85)
17.5 to <20 kg	20.83% (5/24) (7.13, 42.15)	0% (0/6) (0.00, 45.93)	35.71% (5/14) (12.76, 64.86)	0% (0/4) (0.00, 60.24)
20 to <22.5 kg	26.67% (4/15) (7.79, 55.10)	0% (0/2) (0.00, 84.19)	28.57% (4/14) (8.39, 58.10)	0% (0/0) NE
22.5 to <25 kg	28.57% (4/14) (8.39, 58.10)	50% (1/2) (1.26, 98.74)	30% (3/10) (6.67, 65.25)	0% (0/2) (0.00, 84.19)
25 to <30 kg	11.54% (3/26) (2.45, 30.15)	33.33% (1/3) (0.84, 90.57)	9.09% (2/22) (1.12, 29.16)	0% (0/2) (0.00, 84.19)
30 to <35 kg	19.05% (4/21) (5.45, 41.91)	0% (0/0) NE	21.05% (4/19) (6.05, 45.57)	0% (0/2) (0.00,84.19)
35 to <40 kg	16.67% (1/6) (0.42, 64.12)	33.33% (1/3) (0.84, 90.57)	0% (0/2) (0.00, 84.19)	0% (0/1) (0.00, 97.50)
40 to <45 kg	13.16% (5/38) (4.41, 28.09)	0% (0/5) (0.00, 52.18)	17.86% (5/28) (6.06, 36.89)	0% (0/5) (0.00, 52.18)
45 to <50 kg	12% (9/75) (5.64, 21.56)	0% (0/15) (0.00, 21.80)	17.65% (9/51) (8.40, 30.87)	9.09% (1/11) (0.23, 41.28)
50 to <55 kg	10.68% (11/103) (5.45, 18.31)	10.53% (2/19) (1.30, 33.14)	14.29% (9/63) (6.75, 25.39)	0%(0/21) (0.00, 16.11)
55 to <60 kg	9% (9/100) (4.20, 16.40)	0% (0/17) (0.00, 19.51)	14.06% (9/64) (6.64, 25.02)	0% (0/21) (0.00, 16.11)
60 to <65 kg	3.19% (3/94) (0.66, 9.04)	5.88% (1/17) (0.15, 28.69)	3.92% (2/51) (0.48, 13.46)	0% (0/26) (0.00, 13.23)
≥65 kg	5.41% (24/444) (3.49, 7.94)	3.28% (2/61) (0.40, 11.35)	8.64% (21/243) (5.43, 12.91)	0.7% (1/143) (0.02, 3.83)

Source: Applicant Report 1113741 Table 13. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.

Highlighted cells indicated weight bands with the highest frequency of treatment-emergent resistance.
NE: not evaluable.
Note: Subjects with mixed infection were counted once in total number of patients and once by each virus type/subtype category. Patients with unknown subtypes are only summarized under the Total column.

The Applicant evaluated the probability of developing RAS variants based on weight using a logistic regression model. Multiple models were fitted to the dataset dichotomized by weight to obtain the cutoff that provides the best model fit. The Applicant notes that because of the variability in body weights in any particular age group, weight does not give a distinct cutoff; however, log likelihood estimations appear to decline sharply at the ≥16 kg division and remain low for subjects with weights ≥16 kg, based on the Applicant's analysis (Figure 5).

Figure 5: Deviance for different weight dichotomizations. Source: Applicant Report 1113741 Figure 3. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.



TE RAS Frequency by Age

To identify an age cutoff in subjects <12 years of age, the Applicant summarized the frequency of TE RAS in pediatric subjects at each year of age within virus type/subtype subsets (Table 15 and Figure 6) and performed a logistic regression analysis on the pooled dataset dichotomized by age (Figure 6 and 7). Among subjects <12 years of age, the highest frequency of TE RAS was observed in the 2-year-old age band (62.5% including all infections and 81% for A/H3N2 infections) (Table 15); for subjects in 1-year age bands from 5 to 11 years of age, frequencies ranged from 0-29% overall, and 0-33% among A/H3N2 infections (Table 15). In subjects ≥12 years of age, TE RAS frequencies were generally lower than in the age bands between 5 and 11 years but ranged up to 29% in the 32-year age band (Figure 6); however, as noted by the Applicant, sample sizes at each age band vary considerably, which limited the interpretation of the proportions of TE RAS in each age band.

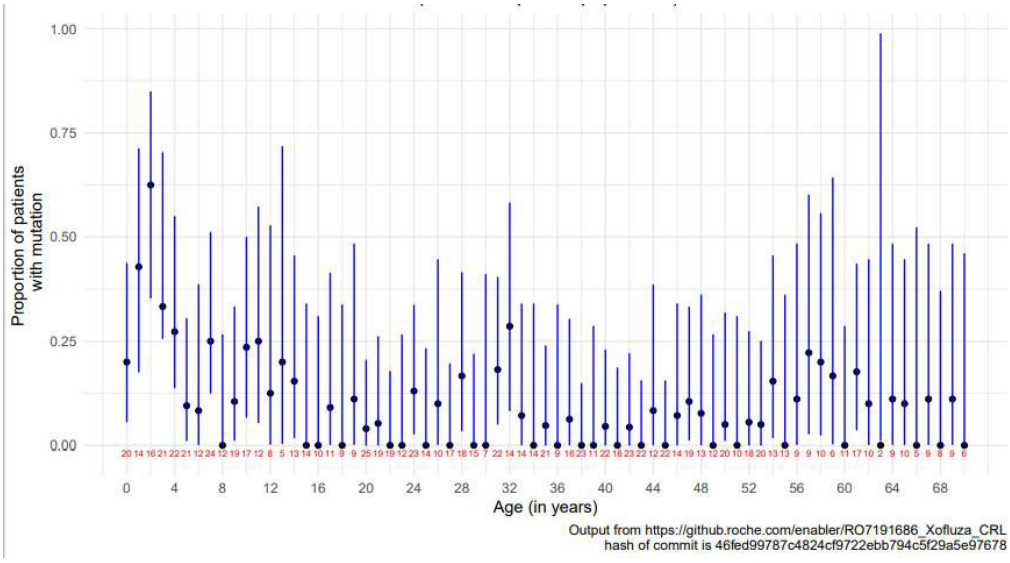
Table 15: Proportion of Subjects with TE RAS by 1-year Age Band and Virus Type/Subtype

Age	Total % (n/N) (95% CI %)	A/H1N1 % (n/N) (95% CI %)	A/H3N2 % (n/N) (95% CI %)	B % (n/N) (95% CI %)
<1 year old	20.00% (4/20) (5.73, 43.66)	25.00% (2/8) (3.19, 65.09)	20.00% (2/10) (2.52, 55.61)	0.00% (0/3) (0.00, 70.76)
1 year old	42.86% (6/14) (17.66, 71.14)	50.00% (1/2) (1.26, 98.74)	62.50% (5/8) (24.49, 91.48)	0% (0/4) (0.00, 60.24)
2 years old	62.50% (10/16) (35.43, 84.80)	50.00% (1/2) (1.26, 98.74)	81.82% (9/11) (48.22, 97.72)	0% (0/3) (0.00, 70.76)

3 years old	47.62% (10/21) (25.71, 70.22)	0% (0/4) (0.00, 60.24)	76.92% (10/13) (46.19, 94.96)	0% (0/4) (0.00, 60.24)
4 years old	31.82% (7/22) (13.86, 54.87)	16.70% (1/6) (0.42, 64.12)	54.55% (6/11) (23.38, 83.25)	0% (0/5) (0.00, 52.18)
5 years old	9.52% (2/21) (1.17, 30.38)	0% (0/3) (0.00, 70.76)	18.18% (2/11) (2.28, 51.78)	0% (0/8) (0.00, 36.94)
6 years old	8.33% (1/12) (0.21, 38.48)	0% (0/2) (0.00, 84.19)	11.11% (1/9) (0.28, 48.25)	0% (0/1) (0.00, 97.50)
7 years old	29.17% (7/24) (12.62, 51.09)	33.30% (1/3) (0.84, 90.57)	33.33% (6/18) (13.34, 59.01)	0% (0/3) (0.00, 70.76)
8 years old	0% (0/12) (0.00, 26.47)	0% (0/1) (0.00, 97.50)	0% (0/11) (0.00, 28.49)	0% (0/1) (0.00, 97.50)
9 years old	10.53% (2/19) (1.30, 33.14)	0% (0/1) (0.00, 97.50)	11.76% (2/17) (1.46, 36.44)	0% (0/1) (0.00, 97.50)
10 years old	23.53% (4/17) (6.81, 49.90)	100% (1/1) (2.50, 100.00)	18.75% (3/16) (4.05, 45.65)	0% (0/1) (0.00, 97.50)
11 years old	25.00% (3/12) (5.49, 57.19)	0% (0/1) (0.00, 97.50)	27.27% (3/11) (6.02, 60.97)	0% (0/0) (NE, NE)

Source: Applicant Report 1113741 Table 14. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.
Note: Subjects with mixed infection were counted once in total number of subjects and once by each virus type/subtype category. Subjects with unknown subtypes are only summarized under the Total column.

Figure 6: The proportion of subjects with TE RAS by age. Numbers in red denote the total number of subjects in each individual age group. Bars indicate 95% CI. Source: Applicant Report 1113741 Figure 5. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.



Applying a linear regression model to pooled data for all evaluated pediatric and adult/adolescent subjects indicated that a cutoff of 5 years of age provided the best model fit (lowest deviance) and could best describe the probability of developing TE RAS viruses among all fitted models, according to the Applicant (Figure 7). The Applicant notes, however, that when the logistic regression model was applied to subjects <12 years of age only (Figure 8), the deviance at age 8 (673.8) approaches that at age 5 (670.8), and a cutoff at age 8 would not significantly reduce frequency of TE RAS viruses compared to a cutoff at age 5. The overall frequency of TE RAS viruses was 15% among subjects 8 to <12 years of age compared to 16.24% among subjects 5 to <12 years age.

Figure 7: Plot of Deviance for Different Age Dichotomizations (all pooled data). Source: Applicant Report 1113741 Figure 6. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.

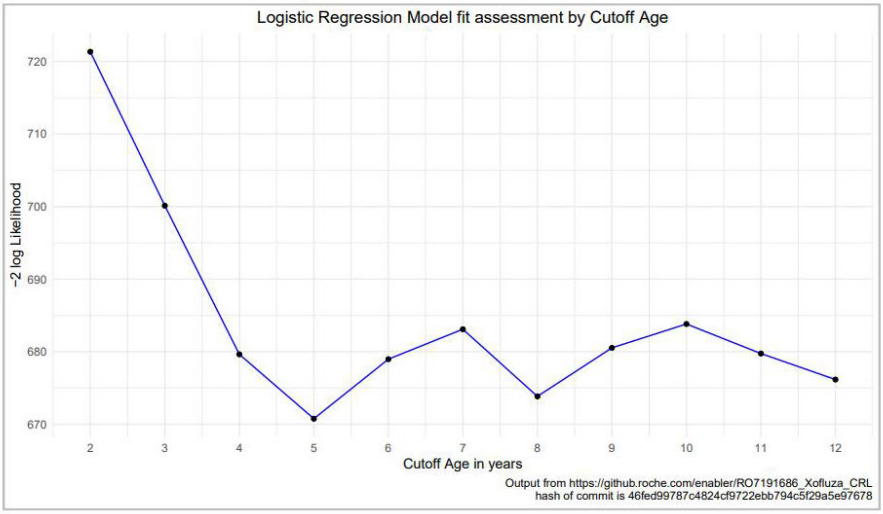
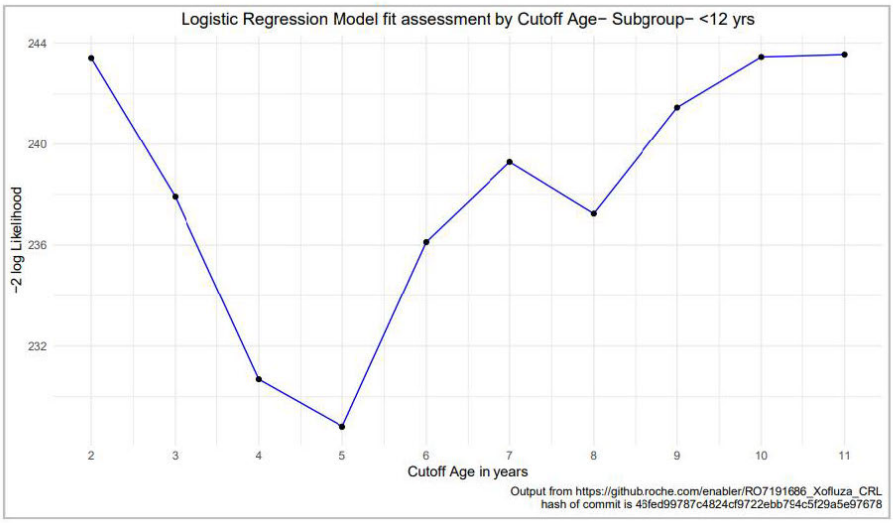


Figure 8: Plot of Deviance for Different Age Dichotomizations (Patients <12 years). Source: Applicant Report 1113741 Figure 7. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.



The Applicant concludes that the age cutoff of 5 years is in line with the logistic regression analysis to identify a weight cutoff, which found the best model fit for subjects with weights ≥16 kg, which approximately matches the weight of 5-years-olds (median 5-year-old weight ~ 18 kg, 3-97 percentile range 14-24 kg: US Charts, see CDC weight for age charts for boys and girls; international charts, see weight for age charts published by the WHO; Japanese growth charts, see Suwa and Tachibana 1993). The Applicant proposes to select age instead of weight as the basis of a cutoff for the risk of TE RAS, as it provides a clearer cutoff point, and because age, more so than weight, is associated with immune system maturation (Bodewes et al. 2012, Simon et al. 2015), which is hypothesized to be a determinant of TE RAS risk for influenza antivirals in general (Roosenhoff et al., 2011).

Among subjects <12 years of age, the frequency of TE RAS is statistically significantly different (non-overlapping 95% CI; P=0.0002, Fishers exact test) between subjects <5 years of age (39.8%) and subjects 5 - 11 years of age (16.24%) (Table 16). However, the difference in frequency between subjects 5 - 11 years of age (16.24%) and subjects ≥12 years of age (7.13%) is also statistically significant (non-overlapping 95% CI; P=0.0019, Fishers exact test). These results indicate that while the magnitude of the difference in TE RAS frequency is greater between the <5 and 5-11 years of age strata than between the 5-11 and ≥12 strata, the difference in frequency between pediatric and adults/adolescent subjects remains significant even when subjects <5 years of age are excluded. Consistent with this observation, the odds ratio associated with the development of TE RAS is lower for subjects ≥12 years compared to subjects 5-11 years of age and subjects <5 years of age (Table 17).

Table 16: Proportion of Subjects with TE RAS by Age Group and Virus Type/Subtype

	Total	A/H1N1	A/H3N2	B
Key Age Categories – TE RAS % (subjects with TE RAS/total evaluated)				
<5 years	39.78% (37/93)	22.73% (5/22)	60.38% (32/53)	0% (0/19)
95% CI	(29.78%, 50.46%)	(7.82%, 45.37%)	(46%, 73.55%)	(0%, 17.65%)
5 - 11 years	16.24% (19/117)	16.67% (2/12)	18.28% (17/93)	0% (0/15)
95% CI	(10.07%, 24.19%)	(2.09%, 48.41%)	(11.02%, 27.65%)	(0%, 21.80%)
≥12 years	7.13% (60/842)	4.44% (6/135)	10.91% (53/486)	0.88% (2/228)
95% CI	(5.48%, 9.08%)	(1.65%, 9.42%)	(8.28%, 14.02%)	(0.11%, 3.13%)
Other Age Categories for Comparison - TE RAS % (subjects with TE RAS/total evaluated)				
1 - <5 years	45.2% (33/73)	21.43% (3/14)	69.77% (30/43)	0% (0/16)
95% CI	(33.52%,57.30%)	(4.66%,50.80%)	(53.87%,82.82%)	(0%,20.59%)
1 - 11 years	27.37% (52/190)	19.23% (5/26)	34.56% (47/136)	0% (0/31)
95% CI	(21.16%,34.29%)	(6.55%,39.35%)	(26.62%,43.19%)	(0%,11.22%)
12- 17 years	9.84% (6/61)	0% (0/1)	11.11% (6/54)	14.29% (1/7)
95% CI	(3.70, 20.19)	(0.00, 97.50)	(4.19, 22.63)	(0.36, 57.87)
≥18 years	6.91% (54/781)	4.48% (6/134)	10.88% (47/432)	0.45% (1/221)
95% CI	(5.24, 8.93)	(1.66, 9.49)	(8.10, 14.20)	(0.01, 2.50)

Source: Applicant Report 1113741 Table 15. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.
Clopper Pearson 95% CI are presented in parentheses.
Note: Subjects with mixed infection were counted once in total number of subjects and once by each virus type/subtype category. Subjects with unknown subtypes are only summarized under the Total column. See FDA analysis in Appendix A.

Table 17: Odds Ratios Associated with TE RAS in <5 and 5-11 Year Age Groups Relative to Subjects ≥12 Years of Age

Age group	Total	A/H1N1	A/H3	B
<5 years	8.61 (5.27, 14.07)	6.32 (1.74, 22.97)	12.45 (6.7,23.14)	0
5 - 11 years	2.53 (1.45, 4.41)	4.3 (0.77, 24.13)	1. 83 (1.00,3.32)	0

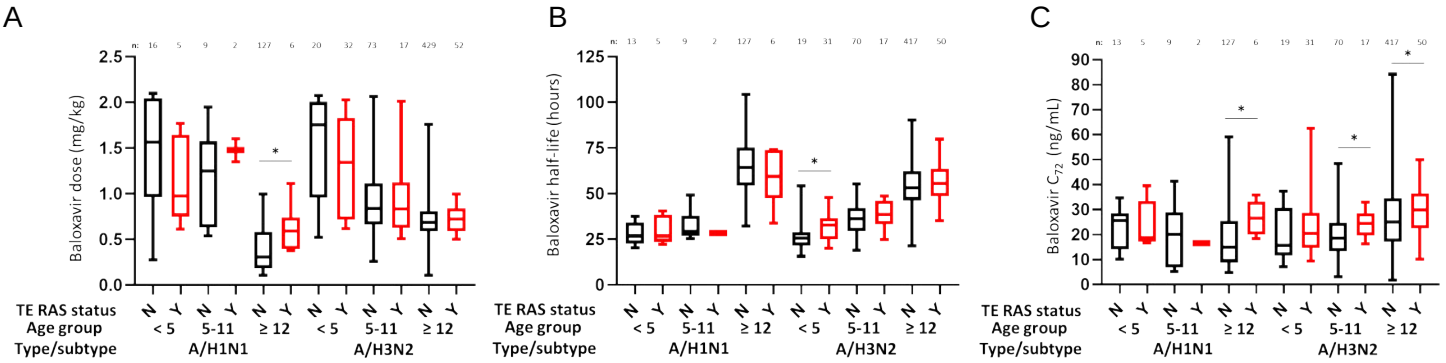
Source: Applicant Report 1113741 Table 16. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.
Reference group is ≥12 years. 95% CI are presented in parentheses.
Note: Subjects with mixed infection were counted once in total number of subjects and once by each virus type/subtype category. Subjects with unknown subtypes are only summarized under the Total column.

Association of Dose and Pharmacokinetic Parameters with Treatment-Emergent Resistance

The Applicant evaluated the association of dose and pharmacokinetic parameters on the frequency of TE RAS in subjects <12 years of age, who received variable weight-based dose levels of baloxavir marboxil. There did not appear to be a consistent dose-dependent association with TE RAS frequency in subjects <12 years of age based on the Applicant's analysis (Applicant Report 1113741 Appendix 4). In an FDA analysis of the data provided by the Applicant for type A virus infections, there was no consistent pattern of association between dose and TE RAS; there was a trend toward lower doses in subjects with TE RAS in the <5 years of age subset regardless of type A virus subtype (Figure 9A), but in contrast, among subjects 5-11 years of age

infected with A/H1N1, there was an association detected between higher dose levels and TE RAS (Figure 9A). In some subsets, longer baloxavir plasma half-life and higher plasma C_{72} values were associated with TE RAS, potentially indicating a relationship between TE RAS and increased selective pressure, rather than reduced drug exposure (Figure 9B and C). The identified associations did not reach statistical significance considering multiple comparisons.

Figure 9: Baloxavir dose level (A), plasma half-life (B), and plasma C_{72} in subjects with and without TE RAS across age groups and type A subtype. Paired sequencing population. "N": subjects without TE RAS; "Y": subjects with TE RAS. Subjects with mixed infections were excluded. * P < 0.05, Mann-Whitney. Source: FDA analysis. Source: FDA analysis of pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.



Conclusions: Key factors associated with increased frequency of TE RAS

Factors associated with increased risk or frequency of TE RAS in baloxavir-treated subjects include:

1. Virus type and subtype: TE RAS occurred significantly more frequently in influenza type A virus infections compared to type B virus infections, and within type A virus, in A/H3N2 infections more frequently than A/H1N1 infections. Baloxavir has reduced activity against type B virus, and therefore exerts a reduced selective pressure on type B virus that may account, in part, for the lower frequency of treatment-emergent resistance in type B.
2. Age: TE RAS occurred at the highest frequency in subjects <5 years of age (39.78%), followed by subjects 5-11 years of age (16.24%). The frequency of TE RAS in subjects 5-11 is over twice the frequency in subjects ≥12 years of age.
3. Baseline influenza antibody titer: Lower baseline influenza antibody titer was associated with increased frequency of TE RAS.
4. Weight: Lower weight was associated with an increased frequency of TE RAS; however, the association was confounded by the correlation between age and weight in subjects <12 years of age.
5. Baseline virus titer: Median baseline virus titer was numerically higher in subjects with TE RAS compared to those without TE RAS; however, the difference was not statistically significant.

Impact of TE RAS on Virus Shedding and Time to Alleviation of Symptoms

Viral Rebound:

In an analyses of pooled viral shedding data, virus rebound was defined using three different definitions:

- FDA rebound definition: Any rise in titer from the previous visit.
- Applicant rebound definition 1: Virus titer increases by at least 0.68 $\log_{10}$ $TCID_{50}$/mL for influenza A virus and 0.86 $\log_{10}$ $TCID_{50}$/mL for influenza B virus, compared to any previous post-baseline visit.

- Applicant rebound definition 2: Virus titer increases by at least 0.68 log_{10} $TCID_{50}$/mL for influenza A and 0.86 log_{10} $TCID_{50}$/mL for influenza B virus, compared with the virus titer at any previous post-baseline visit, up to the current visit following a decrease in log_{10} $TCID_{50}$/mL of at least 0.68 or 0.86 log_{10} $TCID_{50}$/mL for influenza A or B viruses, respectively.

Rebound percentages were similar across rebound definitions, particularly for Type A virus infections; however, the simplified FDA rebound definition was the most sensitive and was used in subsequent analyses unless otherwise noted (Table 18).

Table 18: Rebound Frequency in Baloxavir-Treated Subjects by Rebound Definition (overall population)

	FDA rebound definition		Applicant definition 1		Applicant definition 2	
Age group	**Type A**	**Type B**	**Type A**	**Type B**	**Type A**	**Type B**
<5 years	60% (45/75)	76% (13/17)	59% (44/75)	71% (12/17)	59% (44/75)	71% (12/17)
5-11 years	17% (22/133)	56% (10/18)	13% (17/133)	22% (4/18)	12% (16/133)	22% (4/18)
≥12 years	14% (117/817)	28% (73/259)	9% (73/817)	15% (40/259)	8% (67/817)	9% (24/259)

Source: FDA analysis of pooled dataset of subjects in the ITTI population, with a positive baseline virus titer, treated with baloxavir, and in trials CP40563, T0822, T0831, T0832, T0821, T0833, T0835 (CP40559 excluded) and pooled virus shedding data from individual trials.

Among subjects with baseline and post-baseline sequence data with type A virus infections, virus rebound was significantly more frequent in subjects with TE RAS across age groups, with no significant difference in frequency between age groups among those with TE RAS (Table 19). Virus rebound among subjects without TE RAS was overall more frequent in subjects infected with type B compared to type A virus and exhibited a similar pattern of increased frequency in the younger age cohorts; virus rebound was more frequent in subjects <5 years compared to subjects in the 5-11-year and ≥12-year cohorts. Peak virus rebound generally occurred later in subjects with TE RAS compared to subjects without TE RAS, and peak virus rebound titers were generally higher in subjects with TE RAS compared to subjects with rebound in the absence of TE RAS (Table 20).

While rebound titers tended to be higher in subjects with TE RAS compared to those without TE RAS, rebound titers in individual subjects were less than their baseline virus titers in most cases (Table 20, Figure 10), and rebound above baseline was not otherwise significantly associated with TE RAS in the 5-11-year age group and the ≥12-year age groups (Table 20, Figure 10). There was a trend toward rebound titers greater than baseline titers being associated with TE RAS in subjects <5 years of age, but the difference was not statistically significant (Table 20).

Table 19: Virus Rebound Frequency in Subjects with and without TE RAS

		Influenza Type A		Influenza Type B	
Parameter	**Age group**	No TE RAS	TE RAS	No TE RAS	TE RAS
Percent virus rebound (FDA definition)	<5 years [a]	43% (13/30) [b, c]	84% (31/36) [e, f]	75% (12/16) [b]	0% (0/0)
	5-11 years [a]	8% (7/83) [d]	74% (14/19) [g]	69% (9/13)	0% (0/0)
	≥12 years [a]	11% (61/550)	82% (46/56)	31% (65/210)	100% (1/1)

Source: FDA analysis of Pooled dataset of subjects in the ITTI population, with a positive baseline virus titer, treated with baloxavir and with baseline and post-baseline sequence data in trials CP40563, T0822, T0831, T0832, T0821, T0833, T0835 (CP40559 excluded) and pooled virus shedding data from individual trials.

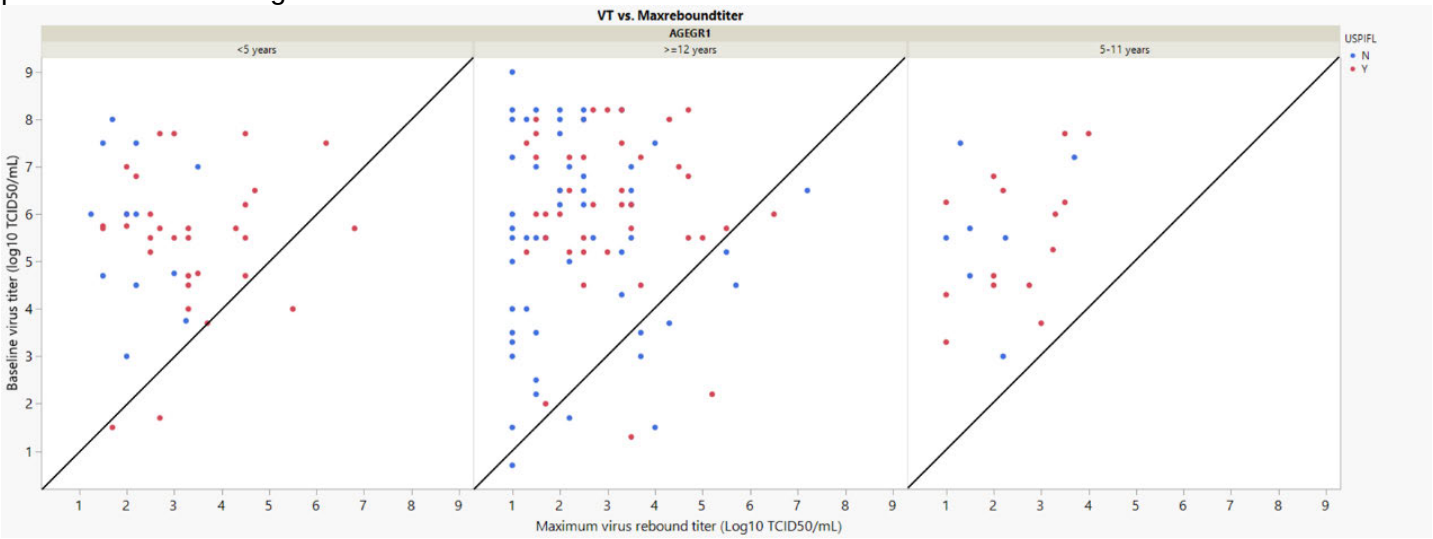
a. P < 0.0001 for frequency of rebound in subjects with TE RAS vs those without.
b. P < 0.0006 for frequency of rebound in subjects <5 years vs ≥12 years.
c. P < 0.0001 for frequency of rebound in subjects <5 years vs 5-11 years.
d. P = 0.5714 for frequency of rebound in subjects 5-11 years vs ≥12 years.
e. P = 0.7748 for frequency of rebound in subjects <5 years vs ≥12 years.
f. P = 0.2884 for frequency of rebound in subjects <5 years vs 5-11 years.
g. P = 0.5096 for frequency of rebound in subjects 5-11 years vs ≥12 years.

Table 20: Characteristics of Virus Rebound Associated with TE RAS and Not Associated with TE RAS

	Influenza Type A		Influenza Type B	
Age group	**No TE RAS**	**TE RAS**	**No TE RAS**	**TE RAS**
Day of peak rebound – median day (95% CI; n)				
<5 years	6 (5, 7; n=13)	5 (5, 6; n=31)	4.5 (4, 5; n=12)	NA
5-11 years	4 (4, 8; n=7)	5 (4, 6; n=14)	4 (3,6; n=9)	NA
≥12 years	3 (3, 4; n=61)	5 (5, 6; n=46)	3 (3, 3; n=65)	4 (NE, n=1)
Peak virus rebound titer – median log_{10} $TCID_{50}$/mL (95% CI; n)				
<5 years	2.2 (1.5, 3; n=13)	3.3 (2.5, 3.7; n=31)	4.15 (3, 5.5; n=12)	NA
5-11 years	1.5 (1, 3.7; n=7)	2.48 (1, 3.5; n=14)	4.5 (1.5, 5.5; n=9)	NA
≥12 years	1.7 (1.3, 2.2; n=61)	2.85 (2.5, 3.3; n=46)	4.5 (3.7, 5.2; n=65)	2.2 (NE)
Change from baseline of maximum rebound titer – median log_{10} $TCID_{50}$/mL (95% CI; n)				
<5 years	-3.5 (-5.3, -1.75; n=13)	-2.2 (-3.2, -1.25; n=31)	-1.65 (-2.8, -0.9; n=12)	
5-11 years	-3.5 (-6.2, -0.8; n=7)	-2.725 (-4.3, -2; n=14)	-3.2 (-4.8, 0.5; n=9)	
≥12 years	-4 (-4.7, -2.8; n=61)	-3.6 (-4.5, -2.7; n=46)	0% (-1.1, 0.2; n=65)	-5.3 (NE; n=1)
Proportion of rebound subjects with max rebound titers > than baseline titer				
<5 years	0% (0/13)	16% (5/31) [a]	17% (2/12)	0% (0/0)
5-11 years	15% (9/61)	7% (3/46)	51% (33/65)	0% (0/1)
≥12 years	0% (0/7)	0% (0/14)	22% (2/9)	0% (0/0)

Source: FDA analysis of pooled dataset of subjects in the ITTI population with positive baseline virus titer, treated with baloxavir, and with baseline and post-baseline sequence data in trials CP40563, T0822, T0831, T0832, T0821, T0833, T0835 (CP40559 excluded) and pooled virus shedding data from individual trials.
a. P = 0.3064, Fisher's exact test for no TE RAS vs TE RAS.

Figure 10: Baseline virus titer vs rebound virus titer for influenza type A-infected, baloxavir-treated subjects with (red dots) and without (blue dots) TE RAS. Note: datapoints are overlapping. Source: FDA analysis of pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials (CP40559 excluded), CP40563, T0822, T0831, T0832, T0821, T0833, T0835 and pooled virus shedding data from individual trials.



Time to Sustained Virus Negativity

Consistent with the association with virus rebound, TE RAS was also significantly associated with increased time to sustained virus negativity (TTSVN), the last negative time point after which no positive time point was recorded (Table 21). The duration of virus shedding in subjects with TE RAS did not appear to be significantly different between age cohorts; however, among subjects with TE RAS, the median TTSVN was shorter in the 5-11 years group (192 hours) compared to the ≥12 years group (216 hours) (Table 21).

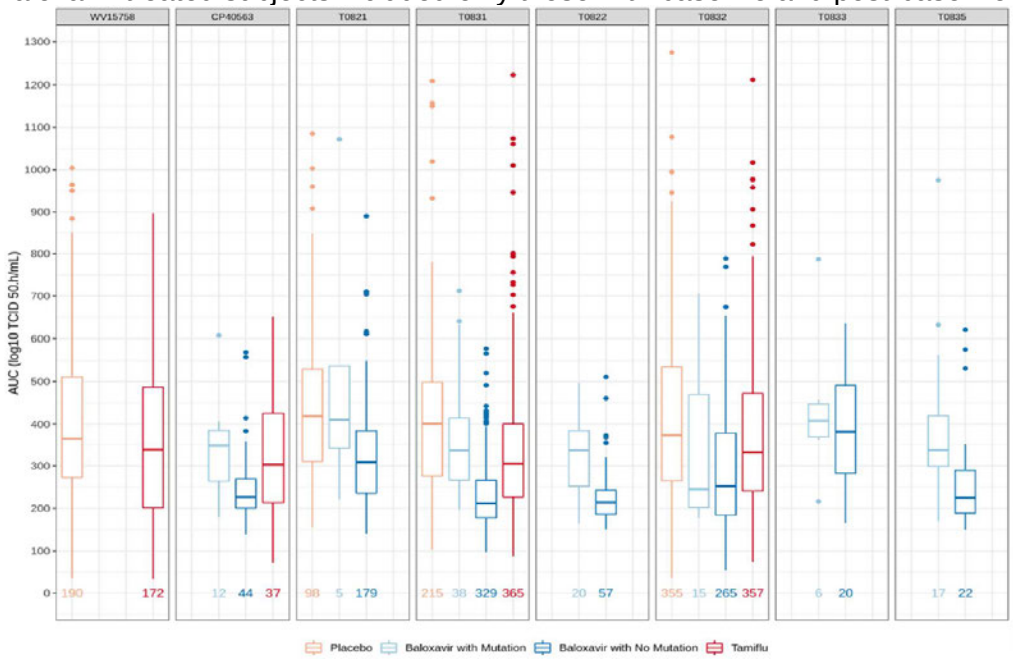
Table 21: Time to Sustained Virus Negativity (TTSVN) by Age Group and TE RAS Status

Age Group	<5 years		5 - 11 years		<12 years		≥12 years	
Substitution Status	No TE RAS	TE RAS	No TE RAS	TE RAS	No TE RAS	TE RAS	No TE RAS	TE RAS
n	47	36	97	19	144	55	776	60
Median (hours)	192.0	240.0	48.0	192.0	72.0	216.0	72.0	216.0
95% CI	96.0-192.0	214.8-264.0	48.0-72.0	117.1-216.0	48.0-96.0	192.0-240.0	72.0-96.0	192.0-216.0
25-75 percentile	48.0-216.0	192.0-264.0	48.0-122.3	72.0-216.0	48.0-168.0	165.8-240.0	48.0-144.0	192.0-216.0
Range	23-264 [b]	27-408	18-264	24-264	18-264 [b]	24-408	24 [b] -528 [b]	48-408
P value [a] (No RAS vs RAS)	0.0003*		0.0004*		<0.0001*		<0.0001*	

Source: Applicant Report 1113741 Table 24.
TTSVN: The time (hours) between the start of study treatment to the first time when the virus titer was below the limit of detection (0.7 log_{10} $TCID_{50}$/mL) and remained below the limit of detection at subsequent time points.
Source: Applicant Report 1113741 Table 24. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.
a. Wilcoxon test.
b. Censored.
* P<0.05 (TE RAS vs no TE RAS).

Median viral AUC in baloxavir-treated subjects with TE RAS were generally higher than viral AUC in baloxavir-treated subjects without TE RAS across trials (Figure 11). While virus shedding was observed to be longer in baloxavir-treated subjects with TE RAS compared to placebo in placebo-controlled adult/adolescent trials (N210854.000; N210854.S001.077), the median viral AUC of baloxavir-treated subjects with TE RAS remained below the viral AUC in placebo subjects in these trials (Figure 11). The median viral titer AUC of baloxavir-treated subjects with TE RAS was higher than that of subjects in the oseltamivir arm in pediatric trial CP40563 and adult/adolescent trial T0831, but was lower in the high risk adult/adolescent trial T0832 (Figure 11).

Figure 11: Viral titer AUC from time of dosing to last viral titer per treatment and per study. “Baloxavir with Mutation”: Baloxavir-treated subjects with TE RAS; “Baloxavir with No Mutation”: Baloxavir-treated subjects without TE RAS; colored numbers are the number of subjects. Source: Applicant Report 1113741 Figure 12. Baloxavir-treated subjects included only those with baseline and post-baseline sequence data.



Time to Alleviation of Symptoms

Time to alleviation of symptoms (TTAS) was generally longer in baloxavir-treated subjects with TE RAS compared to those without, except in the <5-year age group, where TTAS was shorter in subjects with TE RAS (Table 22), despite more frequent and higher magnitude virus rebound associated TE RAS in this age group compared to other age groups (see Tables 19 and 20). Of note, the longer duration of TTAS associated with TE RAS was statistically significant in subjects 5-11 years of age, but not in other age groups. Similar trends were observed for TTAS among the subset of subjects infected with influenza type A virus (Table 23).

Table 22: Time to Alleviation of Influenza Symptoms (TTAS) by Age Group and TE RAS Status

Age Group	**<5 years**		**5 - 11 years**		**<12 years**		**≥12 years**	
Substitution Status	**No TE RAS**	**TE RAS**	**No TE RAS**	**TE RAS**	**No TE RAS**	**TE RAS**	**No TE RAS**	**TE RAS**
n	47	36	97	18	144	54	782	60
Median (hours)	58.9	45.3	42.8	86.3	45.3	68.1	61.2	63.3
95% CI	37.8-74.7	37.4-103.7	29.6-55.2	44.0-122.2	38.2-61.0	42.4-94.9	55.0-65.8	55.0-77.3
25-75 percentile	28.5-112.8	26.1-189.2	22.8-92.4	42.4-125.4	24.0-103.8	35.7-144.4	38.1-115.3	43.1-101.4
Range	10-290	1-321[b]	5-321[b]	23-172	5-321[b]	1-321[b]	0-324[b]	12-321[b]
P value [a] (No RAS vs RAS)	0.6557		0.0477*		0.0683		0.7351	

Source: Applicant Report 1113741 Table 18. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.
a. Wilcoxon test.
b. Censored.
*P<0.05.

Table 23: Time to Alleviation of Influenza Symptoms (TTAS) by Age Group and TE RAS Status – Type A Virus

Age Group	**<5 years**		**5 - 11 years**		**<12 years**		**≥ 12 years**	
Substitution Status	**No RAS**	**RAS**	**No RAS**	**RAS**	**No RAS**	**RAS**	**No RAS**	**RAS**
n	31	36	84	18	115	54	567	59
Median (hours)	64.8	45.3	43.6	86.3	46.7	68.1	53.4	63.2
95% CI	38.9-77.2	35.7-103.7	29.9-61.0	44.0-122.2	39.0-65.7	42.4-94.9	49.8-56.4	55.0-76.3
25-75 percentile	29.1-112.8	26.1-189.2	23.0-109.0	42.4-125.4	24.1- 112.8	35.7-144.4	32.4-104.9	40.9-101.8
Range	12-290	1-321[b]	5-321[b]	23-172	5-321[b]	1-321[b]	0-323[b]	12-321[b]
P value [a] (No RAS vs RAS)	0.9101		0.0641		0.1127		0.2800	

Source: Applicant Report 1113741 Table 19. Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.
a. Wilcoxon test.
b. Censored.

Within oseltamivir-controlled pediatric trial CP40563, the median TTAS in subjects treated with baloxavir and with TE RAS was longer than the median TTAS for subjects treated with oseltamivir, except for subjects <5 years of age (Table 24). Note that treatment-emergent resistance to oseltamivir was not detected among the 36 subjects evaluated by sequencing in trial CP40564 (Integrated Review 214410.000/S-004/S-010); however, oseltamivir resistance, while also increased in pediatric subjects relative to adults, occurs less frequently in A/H3N2 virus, which was dominant in this trial, compared to in A/H1N1 virus (Integrated Review 214410.000/S-004/S-010).

Table 24: Time to Alleviation of Influenza Signs and Symptoms in Trial CP40564 – Comparison to Oseltamivir

Summary statistic	**<5 years**			**≥5 years**			**<12 years**		
	Baloxavir marboxil		**Oseltamivir**	**Baloxavir marboxil**		**Oseltamivir**	**Baloxavir marboxil**		**Oseltamivir**
	No RAS	**RAS**		**No RAS**	**RAS**		**No RAS**	**RAS**	
n	11	5	10	33	7	33	44	12	43
Median (hours)	77.7	67.1	112.4	44.2	122.2	71.1	65.9	108.6	94.3
95% CI	64.8 - 114.2	26.3 – NE	50.7 – 163.5	29.1 – 74.2	23.9 – 171.9	55.4 – 118.2	42.8 – 77.7	45.7 - 147.4	56 – 118.4
Range	23 - 169	26 - 249	46 - 248	17 - 269	23 – 172	15 - 248	17 - 269	23 – 249	15 - 248

Source: Applicant Report 1113741 Table 19. Trial CP40563. Baloxavir marboxil: ITTI population with paired baseline and postbaseline sequence data. Oseltamivir: ITTI. Time to alleviation of influenza signs and symptoms excludes time to return to normal activity.

Evaluations of the Potential for Transmission of RAS Variants

Analysis of Trial T0834 (postexposure prophylaxis)
No clinical studies to date have formally evaluated the incidence of transmission of influenza virus carrying RAS from a baloxavir-treated index patient (IP); however, trial T0834 provided an opportunity to evaluate the potential for RAS variant transmission in a *post hoc* analysis (see Integrated Review 214410.000/S-004/S-010, Section 7.7.1.). In this trial, RAS variants were not detected at baseline in any IP or subject [household contacts (HHCs) of infected IPs] prior to baloxavir prophylaxis or rescue treatment (in placebo-prophylaxed HHCs). Of the 100 subjects (HHCs) in the placebo-prophylaxis arm evaluated at baseline and/or post baseline for RAS variants, 47 of whom were evaluated after their associated IP initiated baloxavir therapy, none harbored a RAS variant virus prior to baloxavir rescue therapy, thus there were no confirmed cases of transmission of RAS variants in this trial; however, with respect to the analysis of the potential for RAS variant transmission, trial T0834 has several key limitations, including:

1. The number of IPs who were incidentally treated with baloxavir and thus had the potential to develop and transmit a RAS variant, and who were matched with a placebo-treated subject (HHC) in whom a RAS variant could have been definitively identified as transmitted, was small (n=172 overall, including IPs in households with no reported transmission events).
2. TE RAS was not accounted for in IPs, and thus the number and identity of IPs who could potentially transmit a RAS variant was unknown.

Exploratory Analysis of the Potential for Transmission of RAS Variants in Trial T0834
In an attempt to leverage the data in trial T0834 to estimate the likelihood of transmission of resistant virus, the Applicant calculated expected frequencies of RAS variants in baloxavir-treated IP based on historical age and virus type/subtype data and then used these frequencies to predict the number of expected RAS variant infections in placebo-prophylaxed HHCs assuming, for the purposes of this analysis, an equal likelihood of transmission for RAS variants and for wild-type viruses. The applicant used estimations of the frequency of TE RAS in IPs and observed and estimated overall transmission rates of influenza virus in the PEP trial to determine the probability of observing no transmission events of RAS variants among the subset of placebo-prophylaxed HHCs associated with baloxavir-treated IPs in trial T0834. In the scenario based on the observed overall transmission rate of 24.3% (37 household transmission events from 152 IPs) in trial T0834, and an assumed frequency of RAS variants in IPs of 10.7%, the Applicant estimated a probability of observing no RAS variants among transmitted virus of 0.0179, and thus concluded that transmission of RAS variants is less likely than transmission of wild type virus (Table 25); however, should the actual frequency of TE RAS have been 5%, and assuming equal transmission probability, the likelihood of observing no RAS among transmission events was estimated to be 0.155 (Table 25).

Table 25: Probability of Detecting Zero RAS Viruses in Households Under Alternative Assumptions.

Assumed RAS variant Frequency in IPs	HH Transmission rate	Predicted Number of IPs with RAS Viruses	Expected Number of HHs with RAS Virus	Probability of Detecting 0 RAS Viruses in HHs
20%	24.3% [a]	30.4	7.40	0.0005
20%	20% [b]	30.4	6.08	0.0020
20%	15% [b]	30.4	4.56	0.0098
10.7%	20% [b]	16.3 [c]	3.26	0.0371
10.7%	**24.3% [a]**	**16.3 [c]**	**3.97**	**0.0179**
10.7%	15% [b]	16.3 [c]	2.44	0.0850
5%	24.3% [a]	7.6	1.85	0.1555
5%	20% [b]	7.6	1.52	0.2170
5%	15% [b]	7.6	1.14	0.3184

Source: Applicant Report 1113741 Table 34.
HH: household; IP: index patient; RAS: resistance-associated substitution

Calculations assume independent HHs with a probability of being infected with RAS viruses defined as the product of the RAS virus rate in IPs and the HH transmission rate.
a. Observed rate of household transmission events (24.3%).
b. Alternative rates of household transmission events (15% and 20%) examined for illustrative purposes.
c. Predicted number of IPs with RAS viruses calculated by the Applicant (Applicant Report 1113741 Table 33), giving an assumed RAS virus frequency in IPs of 10.7%.

This result is consistent with the hypothesis that wild type virus is more likely to be transmitted in households regardless of the frequency of development of resistance to baloxavir in IPs because the period of greatest contagiousness is around the time of peak virus shedding, which occurs soon after symptoms onset for influenza virus infections and before baloxavir treatment is likely to be initiated (and selection of RAS) in an IP. In addition, RAS variants are selected after a period of shedding of wild-type virus, and virus shedding titers at the time that RAS variants emerge in treated patients are generally reduced relative to baseline titers (see Table 20) likely due to immune (innate and adaptive) pressure and potentially reduced fitness of RAS variants.

The null hypothesis the Applicant establishes for their statistical test sets a high bar, by assuming equal opportunity and fitness for resistant virus. The statistical test carried out by the Applicant asks only of the probability of observing zero events when at least 3.97 (16.3 IPs predicted to have a RAS variant x 37 transmission events / 152 IPs) are expected; however, the probability of observing zero events of transmission of RAS variant increases as the number of expected transmission events of RAS variants decreases. For example, if treatment-emergent resistant virus is expected to be transmitted 25% of the time relative to wild type, then the probability of observing zero transmitted RAS variants in T0834, based on the assumed TE RAS frequency of 10.7% and using the Applicant's approach, would be 0.37 (3.97*0.25/152=0.0065; 1-0.0065 = 0.993; 0.993^152 = 0.37).

Trial T0834 was not designed to investigate the probability of transmitted resistance because it did not account for the actual frequency of treatment-emergent resistance in IPs and was too small to confidently estimate a minimum or maximum frequency.

Transmission Modeling
Assuming the probability of transmitting influenza virus from an IP to a HHC is directly proportional to the virus titer level of the IP, the Applicant attempted to model the impact of different antiviral treatments on the likelihood of influenza virus transmission, as well as the risk of TE RAS variant transmission from baloxavir-treated IPs in different scenarios for different age categories (see Applicant Report 1113741 p. 89 for detailed methods). Briefly, a logarithmic relationship between viral shedding titer in an IP and their contagiousness (or "infectiousness"), was assumed based on previously published analyses (Handel et al., 2013 and Handel et al., 2014). Virus shedding and resistance data from all pediatric and adult/adolescent subjects in all trials were used to assign virus shedding titers and the "coefficient of infectiousness" from Days 1-5 for each simulated IP. The number of simulated HHCs associated with each simulated IP was based on a mean of 1.8 HHCs (SD=0.5; normal distribution) to mimic the number of HHCs in trial T0834. In the simulation, infections occurred between Day 1 and Day 5 were assessed. Observed data from trial T0834 was used to validate the analysis (Integrated Review 214410.000/S-004/S-010).

Under different scenarios, including different proportions of influenza type A and B virus infections (75% to 100%), ranges of coefficients of infectiousness (±30%), the overall simulated transmission rates ranged from 24.9-40.5% for IPs treated with placebo, and 15.9-30.4% for IPs treated with baloxavir (Applicant Report 1113741 Table 36). The simulated transmission rates of RAS variants (limited to I38X substitutions for the purpose of the analysis) ranged from 6.1% to 13.4% when any transmission event from an IP with a TE RAS was considered to be the transmission of the RAS variant if occurring at the time the TE RAS was detected,

and 2.9% 4.9% when the transmission of the RAS variant was only assumed if the transmission event occurred after the day of rebound in the IP (Applicant Report 1113741 Table 36).

Based on the Applicant's approach as described, it appears that a major limitation of the modeling of transmission of TE RAS variants specifically, is the exclusion of transmission events detected after Day 5; the median Day of TE RAS detection was Day 7, Day 6, and Day 5 in subjects in the < 5 years, 5-11 years, and ≥12 years age groups, respectively (FDA analysis of pooled ADSL dataset), and the median day of peak virus rebound in these subjects was Day 5 (see Table 20), thus the model may have been unlikely to capture the impact of TE RAS-associated virus rebound on the risk of transmission if only considering virus shedding titers and transmission events occurring between Days 1-5. In addition, these simulations are based on assumptions that may be impacted by differences in virus shedding patterns and transmission fitness of influenza type/subtype variants that can change from season to season, including genetic changes that may improve the fitness of influenza viruses that acquire resistance to baloxavir. The significance of the analyses with respect to the risk of transmission of RAS variants therefore remains unclear.

Global Surveillance of Baloxavir Resistance

Annual Resistance Update Reports are currently produced by the Applicant in accordance with PMC 3503-10 (NDA 210854). To date, 3 reports have been produced, corresponding to the reporting periods December 2018 to November 2019 (N210854.210), December 2019 to November 2020 (N210854.363), and December 2020 to November 2021 (N210854.510). With their current submission, the Applicant provided a cumulative summary of baloxavir resistance surveillance data included in previously submitted reports. The sources for baloxavir resistance surveillance data include public information from the World Health Organization (WHO), which includes contributions from the WHO Collaborating Centers [which includes the US Centers for Disease Control and Prevention (CDC) and National Institute of Infectious Diseases Japan (NIID)], data from the GISAID ("Global initiative on sharing all influenza data") EpiFlu Influenza Virus Database, and published surveillance studies.

Surveillance

Japan (NIID)

In Japan, where baloxavir marboxil was first approved and is more frequently prescribed, the National Institute of Infectious Disease (NIID) tests viruses by a combination of phenotypic and genotypic assays. Clinical specimens and the corresponding patient records are collected at 500 sentinel sites for laboratory-based surveillance. Public Health Institutes isolate influenza viruses from the specimens. Approximately 10-15% of total isolates are randomly selected and sent to the National Institute of Infectious Diseases (NIID). NIID analyzes all viruses sent by the Public Health Institutes by a combination of phenotypic and genotypic assays. As of 7/27/2021, only 6 viruses were tested and no baloxavir RAS were detected. Only one virus (type A/H3N2) has been evaluated for the 2021/2022 season and no RAS were reported.

US CDC

As of 2/7/2022, 520 influenza viruses (3 A/H1N1, 497 A/H3N2, 20 B [Victoria], and 0 B [Yamagata]) collected since 3/10/2021 had been evaluated for susceptibility to baloxavir and no variants with reduced susceptibility were identified (CDC weekly surveillance report).

GISAID

Viral sequences from the GISAID EpiFlu Influenza Virus Database were downloaded through the GISAID platform (accessed 12/2/2021). The GISAID database includes sequences from the INSDC databases (NCBI, EBI & DDBJ) which constitute at least 35%, 34% and 30% of the total type A/H1N1, A/H3N2 and B influenza virus isolates (based on the annotation of Genbank accession numbers), and additional sequences that might

have been directly submitted to GISAID. Viral PA sequences of virus isolated from humans collected since 2009 were evaluated for the presence of RAS. Among these sequences, baloxavir RAS were collectively observed at frequencies of 0.13%, 0.1% and 0.008% among type A/H3N2, A/H1N1, and B viruses, respectively (Tables 26, 27, and 28).

Table 26: Baloxavir RAS and Their Prevalence in Type A/H3N2 Influenza Virus (GISAID EpiFlu database).

PA								
resistance mutation	Position	Reference Residue	Observed Residue	Observed Seq Count	Seq Percent	Observed Mutation	Observed Isolate Count	Isolate Percent
E23K/R/G	23	E	E	38911	99.82%	0.18%	38790	99.81%
			-	68	0.17%		68	0.17%
			X	3	0.01%		3	0.01%
			M	1	0.00%		1	0.00%
A37T	37	A	A	38909	99.81%	0.19%	38790	99.81%
			-	59	0.15%		59	0.15%
			X	6	0.02%		6	0.02%
			T	3	0.01%		3	0.01%
			N	2	0.01%		2	0.01%
			I	2	0.01%		2	0.01%
			F	1	0.00%		1	0.00%
			S	1	0.00%		1	0.00%
I38F/T/M/N/S	38	I	I	38839	99.63%	0.37%	38718	99.63%
			-	55	0.14%		55	0.14%
			X	35	0.09%		35	0.09%
			T	33	0.08%		33	0.08%
			M	11	0.03%		11	0.03%
			V	6	0.02%		6	0.02%
			L	2	0.01%		2	0.01%
			A	1	0.00%		1	0.00%
			N	1	0.00%		1	0.00%
E199G	199	E	E	38928	99.86%	0.14%	38807	99.86%
			X	23	0.06%		23	0.06%
			-	16	0.04%		15	0.04%
			G	6	0.02%		6	0.02%
			D	6	0.02%		6	0.02%
			K	3	0.01%		3	0.01%
			L	1	0.00%		1	0.00%

Source: Applicant Report 1113741 Table 40.

Table 27: Baloxavir RAS and Their Prevalence in Type A/H1N1 Influenza Virus (GISAID EpiFlu database).

PA								
resistance mutation	Position	Reference Residue	Observed Residue	Observed Seq Count	Seq Percent	Observed Mutation	Observed Isolate Count	Isolate Percent
E23K/R/G	23	E	E	30925	99.65%	0.35%	30878	99.66%
			-	86	0.28%		83	0.27%
			G	15	0.05%		15	0.05%
			K	5	0.02%		5	0.02%
			X	2	0.01%		2	0.01%
A37T	37	A	A	30948	99.73%	0.27%	30901	99.73%
			-	71	0.23%		69	0.22%
			X	7	0.02%		7	0.02%
			S	4	0.01%		4	0.01%
			T	1	0.00%		1	0.00%
			N	1	0.00%		1	0.00%
			E	1	0.00%		1	0.00%
I38F/T/M/N/S	38	I	I	30909	99.60%	0.4%	30862	99.61%
			-	71	0.23%		69	0.22%
			V	22	0.07%		22	0.07%
			X	14	0.05%		14	0.05%
			L	8	0.03%		8	0.03%
			T	4	0.01%		4	0.01%
			S	3	0.01%		3	0.01%
			M	1	0.00%		1	0.00%
			A	1	0.00%		1	0.00%
E199G	199	E	E	30958	99.76%	0.24%	30909	99.76%
			-	43	0.14%		43	0.14%
			X	16	0.05%		16	0.05%
			D	10	0.03%		10	0.03%
			K	3	0.01%		3	0.01%
			G	3	0.01%		3	0.01%

Source: Applicant Report 1113741 Table 41.

Table 28: Baloxavir RAS and Their Prevalence in Type B Influenza Virus (GISAID EpiFlu database).

PA								
resistance mutation	Position	Reference Residue	Observed Residue	Observed Seq Count	Seq Percent	Observed Mutation	Observed Isolate Count	Isolate Percent
E23K/R/G	23	E	E - N	24365 29 1	99.86% 0.12% 0.00%	0.12%	24280 29 1	99.88% 0.12% 0.00%
A37T	37	N	N - A	24368 26 1	99.89% 0.11% 0.00%	0.11%	24283 26 1	99.89% 0.11% 0.00%
I38F/T/M/N/S	38	I	I - V M T	24360 26 7 1 1	99.86% 0.11% 0.03% 0.00% 0.00%	0.14%	24275 26 7 1 1	99.86% 0.11% 0.03% 0.00% 0.00%
E199G	199	G	G - R X E D	24353 19 13 7 2 1	99.83% 0.08% 0.05% 0.03% 0.01% 0.00%	0.17%	24268 18 13 7 2 1	99.83% 0.07% 0.05% 0.03% 0.01% 0.00%

Source: Applicant Report 1113741 Table 42.

Cases of Potential Transmission of Baloxavir RAS

The Applicant provided a summary of the surveillance data from Japanese NIID, US CDC, and WHO agencies documenting cases of baloxavir resistance, including cases of potential transmission of baloxavir-resistant virus, along with estimations of total baloxavir marboxil usage in each agency jurisdiction (Table 29). The Applicant reported a cumulative total of 42 viruses containing baloxavir RAS (8 in type A/H1N1, 34 in type A/H3N2, and 0 in type B virus) out of 7,592 influenza virus infections (0.33% of 2,444 type A/H1N1 viruses; 2.3% of 1,482 type A/H3N2 viruses, and 0% of 3,666 type B viruses). Of the 42 RAS viruses, 6 isolates were from untreated individuals, representing the possibility of transmission of treatment-emergent RAS virus from a treated individual. It is unclear if any of these cases represent secondary or tertiary transmission events that may lead to sustained transmission of baloxavir-resistant virus.

Table 29: Summary of Baloxavir Resistance Observed in Surveillance Data and Overall Baloxavir Usage

Region	Season/Reporting Period	Estimated Baloxavir Marboxil Use in Post-Marketing Setting	Incidence of Isolates with Reduced Susceptibility by Subtype			Notes
			A/H1N1	A/H3	B	
Japan (NIID)	2017/2018	(b) (4)	0/254	0/241	0/316	Approval Feb 2018
	2018/2019		6/335 (1.8%)	34/356 (9.6%)	0/42	5/34 no antiviral treatment
	2019/2020		1/831 (0.12%)	0/80	0/130	1/1 no antiviral treatment
	2020/2021		0/2	0/4	0/0	
US (CDC)	2019/2020		0/57	0/101	0/2015	
	2020/2021		No data provided			
WHO	April-Aug 2020		0/821	0/428	0/505	
	Sept 2020-Aug 2021		1/144	0/272	0/658	

Source: Applicant Report 1113741 Table 39.

(b) (4)

The surveillance data available to date do not indicate frequent transmission or sustained circulation of virus with baloxavir RAS, including in Japan where baloxavir marboxil has been distributed most widely; however, given the significant reduction in influenza virus infections worldwide during the 2020-2021 and 2021-2022

influenza virus seasons, surveillance data remain limited as a tool for assessing the risk of sustained transmission of baloxavir-resistant virus.

Applicant's Plan for Enhanced Surveillance
To address the concern that the elevated frequency of TE RAS in pediatric subjects could increase the risk of widespread resistance to baloxavir, the Applicant proposed a post-marketing surveillance strategy that includes comprehensive analyses of all major, public sequence databases, and more frequent reporting. Key features of the enhanced surveillance plan include:

- Review and reporting of more granular data from WHO GISRS collaborating centers than are typically included in the WHO semiannual (every six months) reports on influenza virus surveillance, including the age and treatments received for patients with identified baloxavir-resistant virus, when available.
- Global Xofluza® use by region and age group.
- Semiannual (every six months) reporting of baloxavir resistance surveillance analyses, to align with the WHO surveillance reporting cycle.

The Applicant proposes to submit these semiannual (twice yearly) reports for the first 3 years after a potential approval for baloxavir in subjects down to 5 years of age. The Applicant's plan is generally acceptable.

PROPOSED LABELING
Applicant edits are in red font
FDA edits are in green font

1 INDICATIONS AND USAGE

1.1 Treatment of Influenza

(b) (4)

1.2 ~~Post Exposure~~Postexposure Prophylaxis of Influenza

XOFLUZA is indicated for ~~post exposure~~postexposure prophylaxis of influenza in persons(b) (4)5 years of age and older following contact with an individual who has influenza *[see Clinical Studies (14.3)]*.

1.3 Limitations of Use

Influenza viruses change over time, and factors such as the virus type or subtype, emergence of resistance, or changes in viral virulence could diminish the clinical benefit of antiviral drugs. Consider available information on drug susceptibility patterns for circulating influenza virus strains when deciding whether to use XOFLUZA *[see Microbiology (12.4)* and *Clinical Studies (14)]*.

(b) (4)

12.4 Microbiology

Mechanism of Action

Baloxavir marboxil is a prodrug that is converted by hydrolysis to baloxavir, the active form that exerts anti-influenza virus activity. Baloxavir inhibits the endonuclease activity of the polymerase acidic (PA) protein, an influenza virus-specific enzyme in the viral RNA polymerase complex required for viral gene transcription, resulting in inhibition of influenza virus replication. The 50% inhibitory concentration (IC_{50}) values of baloxavir ranged from 1.4 to 3.1 nM (n=4) for influenza A viruses and 4.5 to 8.9 nM (n=3) for influenza B viruses in a PA endonuclease assay. Viruses with reduced susceptibility to baloxavir have amino acid substitutions in the PA protein.

Antiviral Activity

The antiviral activity of baloxavir against laboratory strains and clinical isolates of influenza A and B viruses was determined in an MDCK cell-based plaque reduction assay. The median 50% effective concentration (EC_{50}) values of baloxavir were 0.73 nM (n=31; range: 0.20–1.85 nM) for subtype A/H1N1 strains, 0.83 nM (n=33; range: 0.35–2.63 nM) for subtype A/H3N2 strains, and 5.97 nM (n=30; range: 2.67–14.23 nM) for type B strains. In an MDCK cell-based virus titer reduction assay, the 90% effective concentration (EC_{90}) values of baloxavir against avian subtypes A/H5N1 and A/H7N9 were in the range of 0.80 to 3.16 nM. The relationship between antiviral activity in cell culture and clinical response to treatment in humans has not been established.

Resistance

~~Cell culture~~ *Cell Culture:*

Influenza A virus isolates with reduced susceptibility to baloxavir were selected by serial passage of virus in cell culture in the presence of increasing concentrations of baloxavir. Reduced susceptibility of influenza A virus to baloxavir was conferred by amino acid substitutions I38T (A/H1N1 and A/H3N2) and E199G (A/H3N2) in the PA protein of the viral RNA polymerase complex.

Clinical Studies

Clinical studies in adults and adolescents subjects (≥ 12 years of age): Influenza A and B viruses with treatment-emergent amino acid substitutions at positions associated with reduced susceptibility to baloxavir in cell culture were observed in clinical studies (Table (b) (4)7). In adult and adolescent subjects who had a confirmed influenza virus infection, the overall frequencies of treatment-emergent amino acid substitutions associated with reduced susceptibility to baloxavir were (b) (4)5% (6/134), (b) (4)11% (53/485), and (b) (4)% (2/224) in influenza A/H1N1, A/H3N2, and B virus infections, respectively, in pooled data from Trials (b) (4) *[see Clinical Studies (14)]*. In Trial (b) (4)303 subjects, (b) (4)≥ 12 years of age, who received

XOFLUZA ~~post-exposure~~postexposure prophylaxis, (b)(4)32 were viral RNA-positive post-baseline, including (b)(4)17 subjects who were evaluated for resistance. Of these (b)(4)17 subjects, influenza virus with substitutions associated with reduced susceptibility to baloxavir was identified in (b)(4)4/4 subjects who developed clinical influenza (as described for the primary endpoint) and (b)(4)6/13 other subjects evaluated who did not meet the primary endpoint definition for clinical influenza *[see Clinical Studies (14)]*.

Clinical studies in pediatrics subjects (5 to < 12 years of age): Selection of influenza viruses with treatment-emergent amino acid substitutions associated with reduced susceptibility to baloxavir has occurred at higher frequencies in pediatric subjects (b)(4)such viruses were detected with overall frequencies of (b)(4)17 (b)(4)% (2/12), (b)(4)18.(b)(4)% (17/93), and 0% (0/(b)(4)13) in influenza A/H1N1, A/H3N2, and B virus infections, respectively, in pooled data from (b)(4)4 pediatric treatment trials in subjects 5 to < 12 years of age.

> **[NOTE TO APPLICANT**: Please confirm the change (b)(4) for type B. These frequencies should only include the virus type/subtypes that were successfully sequenced in subjects with mixed infections. Two subjects in T0822 that had mixed A/H3 +B infection but were only successfully evaluated for A/H3 (8EA003 and 8EA004)].

In Trial (b)(4) of a subgroup of 57 subjects 5 to < 12 years of age who received XOFLUZA ~~post-exposure~~postexposure prophylaxis, 12 were viral-RNA positive post-baseline, including 10 subjects who were evaluated for resistance. Of these 10 subjects, influenza virus with substitutions associated with reduced susceptibility to baloxavir was identified in 2/2 subjects who developed clinical influenza (as described for the primary endpoint) and 1/8 other subject who did not meet the primary endpoint definition for clinical influenza *[see Clinical Studies (14)]*.

Clinical studies in pediatrics subjects < 5 years of age: (b)(4) frequencies of treatment-emergent resistance have been observed in pediatric patients < 5 years of age (b)(4), treatment-emergent amino acid substitutions associated with reduced susceptibility to baloxavir occurred in 24% (4/17), 65% (32/49), and 0% (0/17) of influenza A/H1N1, A/H3N2, and B virus infections, respectively, in pooled data from 4 pediatric treatment trials.

> **[NOTE TO APPLICANT:** RAS frequencies in subjects <5 years of age were reported by virus type/subtype for consistency. Subjects from trial CP40559 were excluded]

Table (b)(4)7 Treatment-Emergent Amino Acid Substitutions in PA Associated with Reduced Susceptibility to Baloxavir

Influenza Type/Subtype	**A/H1N1**	**A/H3N2**	**B**
Amino Acid Substitution	E23K/R, I38F/N/S/T	E23G/K, A37T, I38M/T, E199G	I38T

None of the treatment-emergent substitutions associated with reduced susceptibility to baloxavir were identified in virus from pretreatment respiratory specimens in the clinical studies. (b)(4)

Prescribers should consider available information from the U.S. CDC and/or a local health department on current (b) (4) influenza virus drug susceptibility patterns and treatment effects when deciding whether to use XOFLUZA.

Cross-Resistance

Cross-resistance between baloxavir and neuraminidase (NA) inhibitors, or between baloxavir and M2 proton pump inhibitors (adamantanes), is not expected because these drugs target different viral proteins. The NA inhibitor oseltamivir is active against viruses with reduced susceptibility to baloxavir, including A/H1N1 virus with PA substitutions E23K or I38F/T, A/H3N2 virus with PA substitutions E23G/K, A37T, I38M/T, or E199G, and type B virus with the PA substitution I38T. Influenza virus may carry amino acid substitutions in PA that reduce susceptibility to baloxavir and at the same time carry resistance-associated substitutions for NA inhibitors and M2 proton pump inhibitors. Baloxavir is active against NA inhibitor-resistant strains, including A/H1N1 and A/H5N1 viruses with the NA substitution H275Y (A/H1N1 numbering), A/H3N2 virus with the NA substitutions E119V or R292K, A/H7N9 virus with the NA substitution R292K (A/H3N2 numbering), and type B virus with the NA substitutions R152K or D198E (A/H3N2 numbering). (b) (4) The clinical relevance of phenotypic cross-resistance evaluations has not been established.

Immune Response

Interaction studies with influenza vaccines and baloxavir marboxil have not been conducted.

POST-MARKETING COMMITMENTS/REQUIREMENTS

PMR Number	PMR Description	Timetable
1	Evaluate PA substitution F314S, alone and in combination with A231V, for its impact on baloxavir susceptibility in A/H1N1 virus.	Study Completion: July 2023 Final Report Submission: September 2023
	Rationale: F314S was identified as a treatment-emergent substitution in combination with polymorphism A231V in a subject who exhibited virus rebound (GSC004) in trial T0835.	

PMC Number	PMC Description	Timetable
1	Conduct a prospective, multicenter, observational study in baloxavir marboxil-treated patients over at least five influenza seasons that will capture the susceptibility and genotype of influenza viruses in baseline and on-treatment respiratory samples to determine the frequency of baseline and treatment-emergent baloxavir resistance and the impact on outcomes.	Final Protocol Submission: January 2023 Study Completion: April 2027 Final Report Submission: October 2027

	Rationale: Treatment-emergent resistance to baloxavir has been observed in approximately 7% of subjects in adult/adolescent trials, and in 27% of subjects in pediatric trials conducted to date; however, the frequency, genetic determinants, and impact of baloxavir resistance have been variable across influenza virus types/subtypes and across trials conducted in different influenza seasons. Experience with existing influenza antivirals indicates that monitoring of the susceptibility of virus isolates and frequency of viral genetic determinants of susceptibility in patients treated with baloxavir marboxil over multiple seasons will be required to capture the overall frequency, patterns, and clinical impact of baloxavir resistance, including the potential emergence and impact of widely circulating resistance to baloxavir.

PMC Number	PMC Description	Timetable
2	Provide a semi-annual (twice-yearly) update on global baloxavir usage and emergence of resistance to baloxavir as an integrated review of information from national and international influenza drug resistance databases and sequence databases, including but not limited to World Health Organization and US Centers for Disease Control and Prevention surveillance, data collected by the sponsor, and information in the published literature. Each update will include information on the methodologies (e.g. viral gene sequencing and phenotypic assay descriptions) used in studies during that reporting period. Substitutions of particular interest include all those listed as resistance-associated in the USPI, as well as substitutions currently identified or identified in the future that reduce susceptibility to baloxavir marboxil.	Initial Submission: May 2023 Final Submission: May 2026
	Rationale: Influenza evolves from season to season, and experience with existing influenza antivirals indicates that global monitoring of the frequency of known viral genetic determinants of susceptibility over multiple seasons is required to capture the potential emergence of widely circulating resistance.	

CONCLUSION

The supplements for treatment and postexposure prophylaxis for subjects 5 to 11 years of age are approvable from a Virology perspective. Overall, the increased risks of widespread resistance to baloxavir that may be associated with the increased frequency of treatment-emergent resistance in pediatric patients 5-11 years of age, relative to adult and adolescent patients, remains unknown, and relative to the same risks for other influenza antivirals, does not outweigh the potential benefit of baloxavir treatment in this population. The frequency of treatment-emergent resistance in subjects <5 years of age was significantly higher than the frequency observed in subjects 5-11 years of age or in subject ≥12 years of age; baloxavir marboxil is not approved for treatment of patients <5 years of age, as additional data are required to better assess the risks associated with the increased frequency of resistance in patients <5 years of age.

The data and analyses provided by the Applicant were consistent with independent FDA analyses and confirm that the primary risk factors for baloxavir TE RAS are infection with A/H3N2 virus and younger age. Subjects 5 to 11 years of age exhibited frequencies of treatment-emergent resistance that were approximately twice that of subjects ≥12 years of age, the population in which baloxavir is currently approved. TE RAS was significantly associated with virus rebound and prolonged virus shedding across all age groups, but the association did not vary significantly between subjects 5-11 years of age and subjects ≥12 years of age, and the impact of TE RAS on virus shedding was similar in subjects 5-11 years of age and subjects ≥12 years of age. While TE RAS has been observed to result in prolonged virus shedding compared to placebo in placebo-controlled adult/adolescent (subjects ≥12 years of age) trials, overall virus shedding, as measured by viral AUC, in subjects with TE RAS tended to be lower than in placebo arms across trials. TE RAS was significantly associated prolonged time to alleviation of symptoms in subjects 5-11 years of age, although a similar trend was also observed in subjects ≥12 years of age, and generally did not exceed times to alleviation of symptoms in placebo arms of placebo-controlled adult/adolescent trials.

Modeling analyses of the risk of transmission of treatment-emergent resistance and observed results of PEP trial T0834 did not indicate a significant risk of TE RAS variant transmission; however, the support that these analyses provide are limited by their assumptions and the lack of empirical data from trials evaluating transmission in the context of baloxavir treatment. While additional data from the Applicant's ongoing transmission trial (MV40618) will provide more robust data for assessing the risk of transmitted resistance, data from this trial will be limited to several influenza seasons, including seasons with very low circulation of influenza virus (2020-2021 and 2021-2022). Understanding the risk of sustained transmission of baloxavir-resistant virus will require data collected over multiple, active influenza virus seasons in which baloxavir usage is widespread. Therefore, the risk of sustained and widespread transmission of baloxavir-resistant virus may remain unknown until real-world data are collected over multiple seasons.

Finally, as previously detailed (Integrated Review 214410.000/S-004/S-010), it is not clear that the risk of widespread transmission of baloxavir-resistant virus due to increased use of baloxavir in subjects 5 to 11 years of age would significantly exceed that of oseltamivir, for which increased frequencies of treatment-emergent resistance are also observed in pediatric patient populations for which it is currently approved. In an analysis of the Influenza Resistance Information Study (IRIS), treatment-emergent resistance was evaluated using allele-specific RT-PCR targeting known oseltamivir resistance-associated mutations (IRIS) (NIH 2009; Roosenhoff et al. 2020). Among type A virus infections, the frequency of oseltamivir resistance among the 1622 subjects evaluated postbaseline was 5.8% and 1.9% in A/H1N1 and A/H3N2 virus infections, respectively. In type A subtype and age subsets, the highest frequencies of postbaseline oseltamivir resistance observed within age group bands were 36.4% (4/11) in subjects <1 year of age and 15% (31/206) in subjects up to 5 years of age infected with A/H1N1 virus. Overall, frequencies of postbaseline oseltamivir resistance were 8.5% (42/493) for subjects ≤5 years of age and 1.6% (7/438) for subjects 6 to 12 years of age (NIH 2009). No oseltamivir resistance was observed in the approximately 330 type B virus-infected subjects in this study (Roosenhoff et al. 2020). Lina et al. found that 11.8% of patients 1 to 5 years of age and 1.4% of patients >5 years of age in IRIS who were infected with influenza A virus and treated within 48 hours of symptoms onset had postbaseline resistance detected, and as with baloxavir resistance, oseltamivir resistance was associated with longer duration of viral RNA shedding in this study (10.9 versus 8.1 days for patients with and without treatment-emergent resistance to oseltamivir, respectively) (Lina et al. 2018). It should be noted that the techniques used to detect treatment-emergent resistance to oseltamivir in the studies described above were likely less sensitive than those that have been used to detect treatment-emergent resistance to baloxavir, and thus the frequencies of treatment-emergent oseltamivir resistance that have been reported in past studies are likely underestimated.

DIVISION OF ANTIVIRAL PRODUCTS (HFD-530)

VIROLOGY REVIEW

NDA: 214410 / 210854 SE-009 / 210854 SE-005 SDN (EDR): 92 (0091) / 518 (515) / 519 (516)

DATE REVIEWED: 5/12/2022

Virology Reviewer: William Ince, Ph.D.

William L. Ince, Ph.D.
Clinical Virology Reviewer

CONCURRENCES

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HFD-530/Clin Virol TL/J O'Rear

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HFD-530/NDA
HFD-530/Division File
HFD-530/RPM/Kim

APPENDIX A: Sensitivity analysis of treatment-emergent RAS frequency with and without subjects from trials T0822 and T0821. Source: FDA analysis of pooled data.

Table A1: Frequency analysis of pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0822, T0831, T0832, T0821, T0833, T0835.

	Total [a]	**A/H1N1** [a]	**A/H3N2** [a]	**B** [a]
Key Age Categories – TE RAS % (subjects with TE RAS/total evaluated)				
<5 years	39.8% (37/93)	22.7% (5/22)	61.5% (32/52)	0 (0/19)
5 - 11 years	16.2% (19/117)	16.7% (2/12)	18.3% (17/93)	0 (0/13)
≥12 years	7.1% (60/842)	4.5% (6/134)	10.9% (53/485)	0.9% (2/224)

a. For mixed infections, only successfully sequenced virus type/subtype were included in total and in the respective type/subtype subsets.

Table A2: Frequency analysis of pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40563, T0822, T0831, T0832, T0821, T0833, T0835. Trial CP40559 removed.

	Total [a]	**A/H1N1** [a]	**A/H3N2** [a]	**B** [a]
Key Age Categories – TE RAS % (subjects with TE RAS/total evaluated)				
<5 years	43.4% (36/83)	23.5% (4/17)	65.3% (32/49)	0% (0/17)
5 - 11 years	16.2% (19/117)	16.7% (2/12)	18.3% (17/93)	0% (0/13)
≥12 years	7.1% (60/842)	4.5% (6/134)	10.9% (53/485)	0.9% (2/224)

a. For mixed infections, successfully sequenced virus type/subtype were included in total and in the respective type/subtype subsets.

Table A3: Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0831, T0832, T0821, T0833, T0835: Trial T0822 removed.

	Total	**A/H1N1** [a]	**A/H3N2** [a]	**B** [a]
Key Age Categories – TE RAS % (subjects with TE RAS/total evaluated)				
<5 years	35.8% (29/81)	23.8% (5/21)	57.1% (24/42)	0% (0/17)
5 - 11 years	13.7% (7/51)	20% (2/10)	15.2% (5/33)	0% (0/8)
≥12 years	7.1% (60/842)	4.5% (6/133)	11.1% (52/470)	0.6% (1/180)

a. Mixed infections excluded.

Table A4: Pooled dataset of subjects in the ITTI population treated with baloxavir and with baseline and post-baseline sequence data in trials CP40559, CP40563, T0831, T0832, T0821, T0833, T0835: Subjects in trial T0821 who received 10 or 20 mg dose (vs 40 mg dose) removed.

	Total	**A/H1N1** [a]	**A/H3N2** [a]	**B** [a]
Key Age Categories – TE RAS % (subjects with TE RAS/total evaluated)				
<5 years	35.8% (29/81)	23.8% (5/21)	57.1% (24/42)	0% (0/17)
5 - 11 years	13.7% (7/51)	20% (2/10)	15.2% (5/33)	0% (0/8)
≥12 years	8.2% (58/710)	8.2% (4/49)	10.8% (52/481)	0.5% (1/216)

a. Mixed infections excluded.

APPENDIX B: Postexposure prophylaxis trial T0834 outcomes and resistance by age group. Source: FDA analysis of T0834 datasets (see detailed analysis of trial T0834 in Integrated Review 214410.000/SE-004/005/009/010).

Table B1: Post-baseline virus type/subtype infections in trial T0834 by age group and baseline RT-PCR status. Source: FDA analysis of T0834 datasets. Safety population; excludes post baseline analysis failures (1 subject in the baloxavir marboxil arm)

		Baloxavir Marboxil					Placebo				
Age group (years)	**Baseline RT-PCR status**	**A/H1N1**	**A/H3N2**	**A/Unk**	**Total % positive (n)**	**Negative**	**A/H1N1**	**A/H3N2**	**A/Unk**	**Total % positive (n)**	**Negative**
<5	Negative	3	0	0	25% (3)	9	2	0	0	13.3% (2)	13
	Positive	0	2	0	100% (2)	0	2	3	0	100% (5)	0
5 to 11	Negative	4	5	1	18.5% (10)	44	7	5	0	26.7% (12)	33
	Positive	1	1	0	100% (2)	0	3	2	0	83.3% (5)	1
≥12	Negative	4	8	12	8.5% (24)	257	20	36	17	26.2% (73)	206
	Positive	2	4	2	36.4% (8)	14	3	13	1	68% (17)	8

Unk: Unknown subtype.

Table B2: Resistance-associated substitution (RAS) status of virus detected at baseline or post-baseline and subjected to sequence analysis. Source: FDA analysis of T0834 datasets. Safety population.

		Baloxavir marboxil RAS / total evaluated (for subjects meeting the primary endpoint) [a]				Placebo RAS / total evaluated (for subjects meeting the primary endpoint) [a]				
Age group (years)	**Baseline RT-PCR status**	A/H1N1	A/H3N2	All positive	Negative	A/H1N1	A/H3N2	A/Unk	All positive	Negative
<5	Negative	0/2	0/0	0/2	0/0	0/2	0/0	0/0	0/2	0/0
	Positive	0/0	2/2 (1/1)	2/2 (1/1)	0/0	0/2	0/3	0/0	0/5	0/0
5 to 11	Negative	2/3 (1/1)	1/5 (1/1)	3/8 (2/2)	0/0	0/7	1/5 (1/2)	0/0	1/12 (1/2)	0/0
	Positive	0/1	0/1	0/2	0/0	0/3	0/2	0/0	0/5	0/0
≥12	Negative	4/4 (2/2)	4/7 (1/1)	8/11 (3/3)	0/0	0/20	1/35 (1/22)	0/0	1/55 (1/22)	0/0
	Positive	1/2	1/4 (1/1)	2/6 (1/1)	0/8	0/3	0/13	0/1	0/17	0/3

a. Ratios in parentheses are for subjects who met the primary endpoint, defined as RT-PCR-confirmed influenza virus infection with fever and at least one respiratory symptom, and for whom virus was evaluated for the presence of RAS.

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