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Safety and tolerability of cell culture-derived and egg-derived trivalent influenza vaccines in 3 to <18-year-old children and adolescents at risk of influenza-related complications

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SUMMARY

Background: This descriptive, non-comparative, phase III study evaluated the safety and tolerability of cell culture-derived (TIVc) and egg-derived (TIV) seasonal influenza vaccines in children at risk of influenza-related complications.

Methods: Four hundred and thirty subjects were randomized 2:1 to TIVc or TIV. Subjects aged 3 to <9 years received one dose (if previously vaccinated, $n = 89$) or two doses (if not previously vaccinated, $n = 124$) of the study vaccines; the 9 to <18-year-olds ($n = 213$) received one dose. Reactogenicity was assessed for 7 days after vaccination; safety was monitored for 6 months.

Results: After any vaccination, the most frequently reported solicited local adverse event (AE) was tenderness/pain (TIVc 44%, 66%, 53% and TIV 56%, 51%, 65% in the age groups 3 to <6 years, 6 to <9 years, and 9 to <18 years, respectively) and the systemic AE was irritability (22% TIVc, 24% TIV) in 3 to <6-year-olds and headache in 6 to <9-year-olds (20% TIVc, 13% TIV) and 9 to <18-year-olds (21% TIVc, 26% TIV). There were no cases of severe fever (≥40 °C). No vaccine-related serious AEs were noted. New onset of chronic disease was reported in ≤1% of subjects.

Conclusion: TIVc and TIV had acceptable tolerability and similar safety profiles in at-risk children (NCT01998477).

1. Introduction

Influenza vaccines have traditionally been produced in embryonated chicken eggs.[1] However, using the egg-based manufacturing platform has challenges, such as low production yields, risk of microbial contamination, and poor growth of some human influenza viruses in eggs, which can cause potential delays, long lead times, and a limited manufacturing capacity to meet the global need, especially in the event of a pandemic.[1,2] Cell culture-based technology offers an alternative manufacturing method that can be used as a supplement during times of influenza vaccine demand .[2] To date, three mammalian cell lines have been used for

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the production of influenza vaccines: the Madin–Darby canine kidney (MDCK) cell line, a monkey kidney cell line (Vero), and an adenovirus-transformed human retinal cell line (PER.C6).[1]

Optaflu (Novartis Vaccines and Diagnostics GmbH, Marburg, Germany) is a seasonal, trivalent, cell culture-derived, inactivated subunit vaccine (TIVc) prepared by propagation of influenza virus in MDCK continuous cell lines.[3] This vaccine has been approved in Europe since 2007 for use in individuals aged ≥18 years. It was subsequently licensed in the USA in 2012 for use in adults, under the trade name Flucelvax.[4,5] Clinical trials have shown the efficacy, immunogenicity, and safety profile of the TIVc vaccine to be similar to that of egg-derived influenza vaccines in adults and the elderly.[6–10] A review of data from the Vaccine Adverse Event Reporting System for people aged <18 years vaccinated with a seasonal, trivalent, cell culture-derived, inactivated subunit vaccine did not identify any adverse event (AE) patterns of concern or any new events compared with those reported in clinical trials. The most common category reported was 'general disorders and administration conditions' (n = 152, 49%), related mostly to injection site and systemic reactions, and there were 19 (6%) serious AEs.[11] TIVc is not yet approved for use in children.

In a recent report, the US Centers for Disease Control and Prevention estimated that influenza vaccination prevented approximately 7.2 million illnesses, 3.1 million medically attended illnesses, and 90 000 hospitalizations associated with influenza in the 2013–2014 influenza season in the USA alone.[12] Seasonal influenza vaccination is recommended by the World Health Organization (WHO) particularly for those at higher risk of serious influenza-related complications.[13] These include pregnant women, children aged 6 months to 5 years, the elderly (≥65 years of age), individuals with chronic medical conditions, and healthcare workers. The burden of influenza is highest in children, and studies have shown that the impact is higher in children with comorbidities.[14,15] In this descriptive, non-comparative study, the safety and tolerability profile of the cell culture-derived influenza vaccine (TIVc) and a licensed egg-based trivalent influenza vaccine (TIV) was assessed in children and adolescents aged 3 to <18 years with underlying medical conditions, who are at high risk of influenza-related complications.

2. Methods

This phase III, randomized, observer-blind, non-comparative multicentre descriptive study was conducted in Spain (12 centres) and Italy (four centres) from October 2013 to July 2014. The study was conducted in accordance with the Declaration of Helsinki and Good Clinical Practice and was approved by the ethics review committees concerned. Written informed consent was obtained from the parents or guardians of all participants, and where applicable, informed assent was also sought from the subjects. This study is registered at ClinicalTrials.gov (NCT01998477).

Based on previous influenza vaccination status (number of doses and type of influenza vaccines received), children were classified into two subpopulations: subjects aged <9 years who had not received at least two doses of seasonal influenza vaccine since the year 2010 were classified as 'not previously vaccinated' (vaccine-naive); all subjects >9 years by default and subjects <9 years who had received at least two doses of seasonal influenza vaccine since the year 2010 were considered as 'previously vaccinated' (vaccine-non-naive). Randomization was stratified by age (3 to <9 years; 9 to <18 years) and by previous influenza vaccination status (previously vaccinated; not previously vaccinated). The list of randomization assignments was produced by a validated web-based randomization (WBR) system used by the Biostatistics and Clinical Data Management (BCDM) department of Novartis Vaccines and Diagnostics. Enrolled subjects within each stratum were randomized in a blinded manner 2:1 to receive either TIVc or TIV by the investigator. In each vaccine group, subjects were further stratified by age (3 to <9 years cohort and 9 to <18 years cohort). Subjects in the 3 to <9 years cohort received either one dose (if previously vaccinated) on day 1 or two doses of the vaccine (if not previously vaccinated) administered 4 weeks apart (day 1 and day 29); all subjects in the 9 to <18 years of age cohort received one dose of the vaccine on day 1. There was no stratification by comorbidity. The subjects were followed for safety for 6 months after last vaccination.

The trial was designed as an observer-blind study. During the study, designated unblinded nurses and physicians were responsible for administering the study vaccines to the subjects and were instructed not to reveal the identity of the study vaccines to either the subject or the investigative site personnel involved in the monitoring of the conduct of the trial, except in an emergency.

2.1. Subjects

Male and female children aged 3 to <18 years who were at high risk of influenza-related complications with a confirmed medical history of any of the following diseases were eligible for enrolment: endocrine disorders, chronic cardiovascular diseases, chronic pulmonary diseases, chronic renal or hepatic diseases, neurological and neurodevelopmental conditions, blood disorders, metabolic disorders, weakened immune system due to disease (such as HIV, AIDS, or cancer) or medication (such as corticosteroids), morbid obesity (defined by local standards), or recipient of long-term aspirin therapy. Subjects were excluded if they had a history of Guillain–Barré syndrome or a history of anaphylaxis; hypersensitivity or previous adverse reactions to any vaccine components; were hospitalized or terminally ill (life-expectancy <12 months); had received any vaccination within 2 weeks (inactivated vaccines) or 4 weeks (live vaccines) preceding the trial; had received seasonal influenza vaccination within 6 months prior to study start or any other investigational agent 1 month prior to enrolment, or planned to receive such an agent before completion of the safety follow-up phase; had a body temperature ≥38 °C within 3 days of intended vaccination. Females who were pregnant or of child-bearing potential and who were unwilling to use acceptable birth control measures within 2 months prior to enrolment and through the course of the study were also excluded.

2.2. Vaccines

Each 0.5-ml dose of the investigational MDCK cell culture-derived trivalent influenza vaccine, Optaflu (Novartis Vaccines and Diagnostics), contained purified viral haemagglutinin (HA) and neuraminidase (NA) antigens, approximately 15 μg of HA for each of the WHO reference strains recommended for the 2013–2014 influenza season for the Northern Hemisphere: A/Brisbane/10/2010 (H1N1), A/Texas/50/2012 (H3N2), and B/Massachusetts/2/2012.

Each 0.5-ml dose of the egg-derived trivalent influenza vaccine, Agrippal (Novartis Vaccines and Diagnostics, Italy), contained approximately15 μg of HA for each strain A/California/07/2009 (H1N1), A/Texas/50/2012 (H3N2), and B/Massachusetts/2/2012, as recommended for the 2013–2014 influenza season.

The vaccines were administered by unblinded personnel in the deltoid muscle, preferably of the non-dominant arm. To avoid any potential bias, the unblinded personnel had no other involvement in subject evaluation or other aspects of the study.

2.3. Safety

AEs were gathered separately after both the first and second vaccinations. Subjects were observed for 30 min after both the first

and second vaccinations to monitor for AEs. Subjects or their parents/guardians were provided with diary cards to record the solicited AEs occurring within 1 week of each vaccination (i.e., days 1–7 for all subjects and additionally between days 29 and 36 for not previously vaccinated subjects) as well as the unsolicited AEs occurring after vaccination between days 1 and 29 (previously vaccinated subjects who received one dose) and days 1 and 57 (not previously vaccinated subjects who received two doses). Age-appropriate solicited AEs were collected. For subjects aged <6 years, the solicited local AEs were injection site erythema, injection site ecchymosis, and injection site tenderness, and the systemic AEs were change in eating habits, shivering, sleepiness, irritability, vomiting, diarrhoea, and fever (body temperature ≥38 °C). For subjects aged ≥6 years, the solicited local AEs were injection site erythema, injection site ecchymosis, and injection site pain, and the systemic AEs were loss of appetite, nausea, fatigue, generalized myalgia, generalized arthralgia, headache, shivering, vomiting, diarrhoea, and fever. In addition, other indicators of reactogenicity were also collected, including the use of analgesic/antipyretic medication for prophylaxis and treatment. The AEs were classified as 'mild' if the event was transient with no limitation in normal daily activity, 'moderate' if the event led to some limitation in normal daily activity, or 'severe' if the event meant the subject was unable to perform normal daily activities.

The new onset of chronic disease (NOCD), serious AEs (SAEs), medically attended AEs, and AEs leading to withdrawal were captured throughout the study period, i.e., days 1–181 for previously vaccinated subjects and days 1–209 for not previously vaccinated subjects. All AEs were to be monitored until resolution, or if an AE became chronic, a cause was to be determined. For any ongoing AEs at study conclusion, the need for follow-up was based on the investigator's assessment. Unsolicited AEs were judged as probably related, possibly related, or not related to vaccination by the investigator.

2.4. Statistical analysis

Assuming a 10% dropout rate, a total of 504 subjects were planned to be enrolled in the study so as to have 450 evaluable subjects (300 in the TIVc group and 150 in the TIV group). With a sample size of 300 subjects in the TIVc group, the probability of observing at least one AE was 95% when the rate of the event is 0.01 (1 in 100), while among 450 subjects exposed to any of the two study vaccines, the probability would be 99%. Safety data were evaluated descriptively and for each vaccination are expressed as the frequencies and percentages of subjects reporting AEs. Safety analyses are presented by overall vaccine group (3 to <18 years) and individual age cohorts (3 to <9 years and 9 to <18 years) and by vaccination status (previously vaccinated and not previously vaccinated). All subjects who received at least one study vaccination and provided post-vaccination reactogenicity data were included in the safety set solicited AEs; all subjects who provided post-vaccination unsolicited data were included in the safety set unsolicited AEs. The overall safety set consisted of all participants who provided either post-vaccination reactogenicity data or unsolicited data.

3. Results

The first subject was recruited on October 25, 2013 and the last completed the study on July 31, 2014. Follow-up was approximately 6 months for each patient. The study was stopped at the

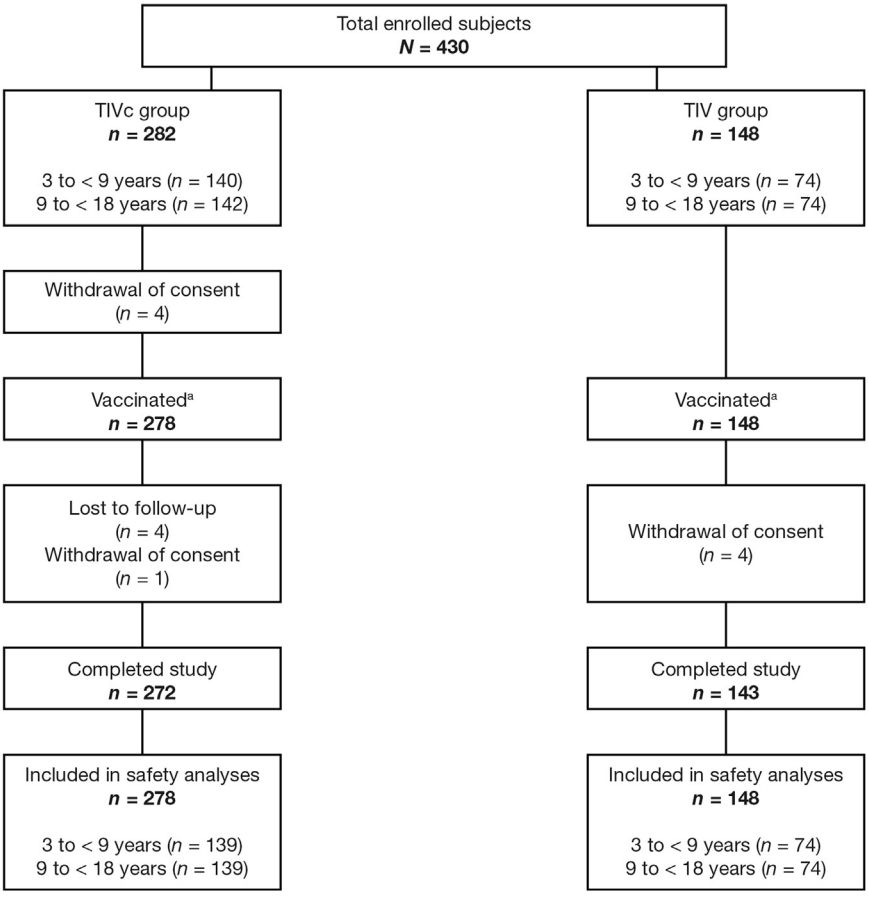

Figure 1. Participant Flow.

end of the vaccination season before reaching the target enrolment of 504 participants. Of the total 430 subjects enrolled in the study, 96% of subjects in the TIVc group and 97% in the TIV group completed the study protocol (Figure 1). A total of 426 vaccinated subjects provided safety data for at least one time point and were included in the overall safety set. The baseline demographics of the enrolled study population are presented in Table 1. Both vaccine groups were balanced with respect to mean age, sex, and ethnicity. A summary of the underlying medical conditions of the subjects enrolled is given in Table 2; these were similar in the two vaccine groups .

3.1. Solicited adverse events

Overall, 73% of subjects in the TIVc and TIV groups reported at least one solicited AE. Rates of any solicited AEs were lower after the second vaccine dose than following the first dose, and this difference was more pronounced in the TIV group than in the TIVc group (Table 3). The majority of AEs were mild or moderate in severity. The most commonly reported solicited local and systemic AEs, respectively, were injection site tenderness (44% in TIVc and 56% in TIV) and irritability (22% in TIVc and 24% in TIV) in subjects aged <6 years, and injection site pain (57% in TIVc and 60% in TIV) and headache (21% TIVc and 22% in TIV) in subjects ≥6 years of age. Mild to moderate fever (≥38 °C to ≤40 °C) was reported by 7% of subjects in the TIVc group and 4% in the TIV group. No cases of severe fever (≥40.0 °C) were reported in the study. A similar percentage of subjects in the two vaccine groups reported analgesic and antipyretic use for treatment (12% in TIVc and 9% in TIV) and prophylaxis (8% in TIVc and 7% in TIV).

In the 3 to <6 years previously vaccinated group, injection site tenderness was reported in 36% (n = 10/28) of subjects in the TIVc group and 58% (n = 11/19) in the TIV group (Table 4). One case each of severe erythema (>50 mm), severe induration (>50 mm), severe chills, and severe irritability were reported in the TIV group. Fever was reported by four subjects in the TIVc group and by three subjects in the TIV group (Table 4). In the 3 to <6 years not previously vaccinated TIVc group, one subject reported severe chills and 18% of subjects reported fever (n = 9/50; eight subjects after the first vaccination and two subjects after the second vaccination). No fever was reported in the TIV group (Table 4).

In the 6 to <9 years previously vaccinated group, solicited local and systemic AEs were reported by 62% and 35% of subjects in the TIVc group and by 50% and 31% of subjects in the TIV group, respectively (Table 5). There were no reports of severe AEs in the TIVc group; one case each of severe erythema (>100 mm) and severe induration (>100 mm) were reported in the TIV group. Fever was reported by one subject (4%) in the TIVc group (Table 5). In the 6 to <9 years not previously vaccinated group, injection site pain was reported by 73% (n = 24/33) of subjects in the TIVc group and 52% (n = 12/23) in the TIV group (Table 5). One case each of severe pain and severe myalgia were reported in the TIVc group (after second vaccination). Fever was reported by one subject in the TIVc group (after first vaccination); there were no reports of fever in the TIV group.

In the 9 to <18 years group, solicited local and systemic AEs were reported by 59% and 42%, respectively, of subjects in the TIVc group and by 68% and 43%, respectively, of subjects in the TIV group. Severe pain was reported by one subject in each vaccine group. Two subjects in the TIVc group had severe erythema (>100 mm); four subjects in the TIV group had severe induration (>100 mm). Severe systemic AEs were reported by 1% of subjects in the TIVc group and by 1–3% of subjects in the TIV group. Fever was reported by 3% (n = 4/137) of subjects in the TIVc group and 4% (n = 3/72) in the TIV group (Table 6).

3.2. Unsolicited adverse events

During the entire study period (day 1 to day 181/day 209), a similar percentage of subjects reported unsolicited AEs in the TIVc and the TIV groups (Table 7). A higher percentage of subjects in the 3 to <9 years group, both in the previously vaccinated (85% TIVc and 83% TIV) and not previously vaccinated (91% TIVc and 82% TIV) groups, reported unsolicited AEs than in the 9 to <18 years group (65% in TIVc and 68% in TIV). Across age groups, between 5% and 7% of subjects in the TIVc group and 5% and 13% in the TIV group

Table 1
Study population demographics

	3 to <9 years		9 to <18 years	
	TIVc n = 140	TIV n = 74	TIVc n = 142	TIV n = 74
Mean age, years ± SD	5.3 ± 1.8	5.7 ± 1.7	12.1 ± 2.3	12.3 ± 2.5
Not previously vaccinated, n	86	39	-	-
Previously vaccinated, n	54	35	142	74
Male, %	58	58	56	57
Race, %				
White	86	85	91	92
Black/African American	1	1	<1	4
Asian	0	3	1	0
Other	12	11	7	4
Mean weight, kg ± SD	22.4 ± 7.5	24.2 ± 8.6	48.7 ± 16.6	47.5 ± 13.8
Mean height, cm ± SD	114.2 ± 13.7	115.7 ± 13.2	152.0 ± 14.3	151.8 ± 14.8

TIVc, cell culture-derived trivalent influenza vaccine; TIV, egg-derived trivalent influenza vaccine; SD, standard deviation.

Table 2
Underlying medical history by system organ class (SOC) of enrolled subjects in each vaccine group

Medical history summary	TIVc n = 282, n (%)	TIV n = 148, n (%)
Blood and lymphatic system disorders	9 (3)	6 (4)
Cardiac disorders	7 (2)	3 (2)
Congenital, familial, and genetic disorders	60 (21)	30 (20)
Ear and labyrinth disorders	2 (1)	1 (1)
Endocrine disorders	10 (4)	5 (3)
Eye disorders	13 (5)	7 (5)
Gastrointestinal disorders	18 (6)	14 (9)
General disorders and administration site conditions	2 (1)	1 (1)
Hepatobiliary disorders	2 (1)	3 (2)
Immune system disorders	15 (5)	13 (9)
Infections and infestations	63 (22)	36 (24)
Investigations	5 (2)	1 (1)
Metabolism and nutrition disorders	40 (14)	15 (10)
Musculoskeletal and connective tissue disorders	14 (5)	12 (8)
Neoplasms benign, malignant, and unspecified	13 (5)	8 (5)
Nervous system disorders	29 (10)	11 (7)
Psychiatric disorders	21 (7)	9 (6)
Renal and urinary disorders	14 (5)	6 (4)
Reproductive system and breast disorders	1 (<1)	0
Respiratory, thoracic, and mediastinal disorders	138 (49)	75 (51)
Skin and subcutaneous tissue disorders	23 (8)	7 (5)
Surgical and medical procedures	21 (7)	9 (6)
Vascular disorders	1 (<1)	2 (1)

TIVc, cell culture-derived trivalent influenza vaccine; TIV, egg-derived trivalent influenza vaccine.

Table 3
Subjects aged 3 to <18 years with solicited AEs from day 1 to day 7 after each/any vaccination, by vaccine group (safety set solicited AEs)

	TIVc, *n* (%)			TIV, *n* (%)		
	Dose 1 *n* = 277	Dose 2[a] *n* = 83	Any *n* = 277	Dose 1 *n* = 145	Dose 2[a] *n* = 40	Any *n* = 146
Any solicited AE	193 (70)	49 (59)	202 (73)	105 (72)	21 (53)	107 (73)
Local	149 (54)	41 (49)	163 (59)	89 (61)	18 (45)	91 (62)
Systemic	117 (42)	29 (35)	128 (46)	55 (38)	8 (20)	58 (40)

AE, adverse event; TIVc, cell culture-derived trivalent influenza vaccine; TIV, egg-derived trivalent influenza vaccine.
[a] Second vaccine dose was given only to vaccine-naive children (not previously vaccinated) in the 3 to <9 years cohort.

Table 4
Solicited adverse events in subjects aged 3 to <6 years[a]

Event		Previously vaccinated, *n* (%)		Not previously vaccinated, *n* (%)	
		TIVc *n* = 28	TIV *n* = 19	TIVc *n* = 50	TIV *n* = 15
Local adverse events					
Tenderness	Mild	7 (25)	9 (47)	17 (34)	4 (27)
	Moderate	3 (11)	2 (11)	7 (14)	4 (27)
	Severe	0	0	0	0
Erythema	Mild	5 (19)	1 (6)	1 (2)	3 (20)
	Moderate	2 (7)	2 (11)	3 (6)	1 (7)
	Severe	0	1 (6)	0	0
Induration	Mild	2 (7)	0	3 (6)	3 (20)
	Moderate	0	1 (5)	5 (10)	1 (7)
	Severe	0	1 (5)	0	0
Ecchymosis	Mild	1 (4)	1 (5)	4 (8)	2 (13)
	Moderate	1 (4)	1 (5)	1 (2)	2 (13)
	Severe	0	0	0	0
Systemic adverse events					
Change in eating habits	Mild	2 (7)	1 (5)	9 (18)	4 (27)
	Moderate	3 (11)	2 (11)	3 (6)	0
	Severe	0	0	0	0
Chills	Mild	1 (4)	1 (5)	6 (12)	0
	Moderate	1 (4)	0	0	0
	Severe	0	1 (5)	1 (2)	0
Diarrhoea	Mild	3 (11)	1 (5)	2 (4)	1 (7)
	Moderate	0	0	0	1 (7)
	Severe	0	0	0	0
Irritability	Mild	4 (14)	3 (16)	9 (18)	2 (13)
	Moderate	2 (7)	1 (5)	2 (4)	1 (7)
	Severe	0	1 (5)	0	0
Sleepiness	Mild	4 (14)	3 (16)	7 (14)	2 (13)
	Moderate	0	0	3 (6)	0
	Severe	0	0	0	0
Vomiting	Mild	3 (11)	1 (5)	4 (8)	1 (7)
	Moderate	0	0	1 (2)	0
	Severe	0	0	0	0
Fever	Mild	3 (11)	1 (5)	7 (14)	0
	Moderate	0	2 (11)	2 (4)	0
	Severe	1 (4)	0	0	0

TIVc, cell culture-derived trivalent influenza vaccine; TIV, egg-derived trivalent influenza vaccine.
[a] Grading for erythema, ecchymosis, and induration: mild (1–9 mm), moderate (10–50 mm), severe (>50 mm); mild fever (≥38 °C to <39 °C), moderate fever (≥39 °C to <40 °C), severe fever (≥40 °C).

reported AEs that were considered as possibly or probably related to study vaccination. The majority of the possibly related AEs were solicited local and systemic AEs that persisted beyond day 7, in both vaccine groups.

The most frequently reported unsolicited AEs in both vaccine groups, as per the Medical Dictionary for Regulatory Activities (MedDRA) system organ classes (SOC) were classified as 'infections and infestations' (51% in TIVc and 49% in TIV), 'respiratory, thoracic, and mediastinal disorder' (26% in TIVc and 23% in TIV), and 'general disorders and administrative conditions' (12% in TIVc and 16% in TIV).

3.3. Serious adverse events

A total of 22 SAEs were reported by 16 subjects; 17 SAEs in 12 subjects (4%) in the TIVc group and five SAEs in four subjects (3%) in the TIV group. No SAE was considered to be vaccine-related (Table 7). The SAEs were reported by a similar percentage of subjects in the TIVc (4%) and the TIV (6%) groups in the 3 to <9 years previously vaccinated group. Among the 3 to <9 years not previously vaccinated group, 5% of subjects in the TIVc group reported SAEs, while there were no reports of SAEs in the TIV group. In the 9 to <18 years age cohort, 4% of subjects in the TIVc group and 3% in the TIV group reported SAEs.

3.4. New onset of chronic disease

NOCD was reported in four subjects (*n* = 3 in TIVc and *n* = 1 in TIV). The following NOCDs by MedDRA preferred term were reported in the study: refraction disorder, drug resistance and hypertensive crisis, toxic encephalopathy, and vomiting and decreased appetite.

Table 5
Solicited adverse events in subjects aged 6 to <9 years[a]

Event		Previously vaccinated, *n* (%)		Not previously vaccinated, *n* (%)	
		TIVc *n* = 26	TIV *n* = 16	TIVc *n* = 32–35	TIV *n* = 22–24
Local adverse events					
Pain	Mild	13 (50)	5 (31)	16 (48)	12 (52)
	Moderate	2 (8)	3 (19)	7 (21)	0
	Severe	0	0	1 (3)	0
Erythema	Mild	1 (4)	2 (13)	5 (15)	2 (8)
	Moderate	0	1 (6)	2 (6)	3 (12)
	Severe	0	1 (6)	0	0
Induration	Mild	1 (4)	0	4 (12)	1 (4)
	Moderate	0	2 (13)	2 (6)	3 (13)
	Severe	0	1 (6)	0	0
Ecchymosis	Mild	2 (8)	0	5 (15)	4 (17)
	Moderate	0	0	0	1 (4)
	Severe	0	0	0	0
Systemic adverse events					
Chills	Mild	3 (12)	0	5 (15)	0
	Moderate	0	1 (6)	0	0
	Severe	0	0	0	0
Diarrhoea	Mild	1 (4)	0	4 (12)	1 (4)
	Moderate	0	0	0	0
	Severe	0	0	0	0
Vomiting	Mild	0	0	4 (12)	0
	Moderate	0	1 (6)	0	0
	Severe	0	0	0	0
Arthralgia	Mild	2 (8)	1 (6)	4 (12)	1 (5)
	Moderate	0	0	0	0
	Severe	0	0	0	0
Fatigue	Mild	2 (8)	2 (13)	3 (9)	0
	Moderate	0	0	2 (6)	0
	Severe	0	0	0	0
Headache	Mild	0	2 (13)	10 (30)	1 (5)
	Moderate	1 (4)	1 (6)	1 (3)	1 (5)
	Severe	0	0	0	0
Loss of appetite	Mild	2 (8)	1 (6)	7 (21)	1 (4)
	Moderate	0	1 (6)	3 (9)	2 (9)
	Severe	0	0	0	0
Myalgia	Mild	4 (15)	1 (6)	5 (15)	1 (5)
	Moderate	0	1 (6)	1 (3)	0
	Severe	0	0	1 (3)	0
Nausea	Mild	1 (4)	1 (6)	2 (6)	0
	Moderate	0	1 (6)	1 (3)	0
	Severe	0	0	0	0
Fever	Mild	1 (4)	0	1 (3)	0
	Moderate	0	0	0	0
	Severe	0	0	0	0

TIVc, cell culture-derived trivalent influenza vaccine; TIV, egg-derived trivalent influenza vaccine.
[a] Grading for erythema, ecchymosis, and induration: mild (1–9 mm), moderate (10–50 mm), severe (>50 mm); mild fever (≥38 °C to <39 °C), moderate fever (≥39 °C to <40 °C), severe fever (≥40 °C).

3.5. Medically attended AEs

A similar percentage of subjects reported medically attended AEs in the two vaccine groups (Table 7). When analyzed by age cohort, the frequency of medically attended AEs was 84% in the TIVc group and 72% in the TIV group in the 3 to <9 years not previously vaccinated group. In the 3 to <9 years previously vaccinated group, the reported rates were 70% in TIVc and 69% in TIV. Overall, the rate of medically attended AEs in the 3 to <9-year-old subjects was higher in those who had received two doses than in those who had received one dose of TIVc, while rates were similar in the TIV groups. In the 9 to <18 years group, the rates of medically attended AEs were lower (56% in TIVc and 59% in TIV) than those reported in the younger age group in either vaccine group. The most frequently reported medically attended MedDRA AEs were 'infections and infestations', followed by 'respiratory, thoracic, and mediastinal disorders' and 'gastrointestinal disorders'.

Table 6
Solicited adverse events in subjects 9 to <18 years of age[a]

Event		TIVc *n* = 138 *n* (%)	TIV *n* = 72 *n* (%)
Local adverse events			
Pain	Mild	57 (41)	33 (46)
	Moderate	15 (11)	13 (18)
	Severe	1 (1)	1 (1)
Erythema	Mild	0	0
	Moderate	3 (2)	2 (3)
	Severe	2 (2)	0
Induration	Mild	1 (1)	1 (2)
	Moderate	4 (3)	2 (3)
	Severe	1 (1)	4 (7)
Ecchymosis	Mild	0	0
	Moderate	0	0
	Severe	0	0
Systemic adverse events			
Chills	Mild	14 (10)	4 (6)
	Moderate	4 (3)	2 (3)
	Severe	1 (1)	0
Diarrhoea	Mild	7 (5)	3 (4)
	Moderate	4 (3)	1 (1)
	Severe	0	0
Vomiting	Mild	6 (4)	1 (1)
	Moderate	0	2 (3)
	Severe	2 (1)	0
Arthralgia	Mild	14 (10)	6 (8)
	Moderate	3 (2)	1 (1)
	Severe	1 (1)	0
Fatigue	Mild	11 (8)	9 (13)
	Moderate	10 (7)	4 (6)
	Severe	2 (1)	1 (1)
Headache	Mild	18 (13)	12 (17)
	Moderate	10 (7)	6 (8)
	Severe	1 (1)	1 (1)
Loss of appetite	Mild	14 (10)	9 (13)
	Moderate	5 (4)	3 (4)
	Severe	1 (1)	0
Myalgia	Mild	24 (17)	9 (13)
	Moderate	4 (3)	2 (3)
	Severe	1 (1)	0
Nausea	Mild	10 (7)	6 (8)
	Moderate	5 (4)	2 (3)
	Severe	2 (1)	2 (3)
Fever	Mild	4 (3)	3 (4)
	Moderate	0	0
	Severe	0	0

TIVc, cell culture-derived trivalent influenza vaccine; TIV, egg-derived trivalent influenza vaccine.
[a] Grading for erythema, ecchymosis, and induration: mild (1–9 mm), moderate (10–50 mm), severe (>50 mm); mild fever (≥38 °C to <39 °C), moderate fever (≥39 °C to <40 °C), severe fever (≥40 °C).

3.6. AEs leading to withdrawal and deaths

There were no AEs reported that led to withdrawal of the subject in either vaccine group. No deaths were reported during the study period.

4. Discussion

TIVc is at present the only cell culture-derived seasonal influenza vaccine that is licensed for use in adults and is especially indicated for individuals at risk of influenza infection.[4] This descriptive, non-comparative study is the first to evaluate the safety of TIVc in 3 to <18-year-old children and adolescents who are at risk of influenza-related complications. The two vaccine groups were balanced with respect to mean age, sex, and ethnicity, and the comorbidities recorded were similar in the two groups. These results show TIVc has an acceptable tolerability profile in this age group, with most solicited AEs resolving within 7 days of vaccination. The reported rates of AEs were similar in the TIVc and TIV groups.

Table 7
Subjects aged 3 to <18 years reporting unsolicited adverse events after any vaccination, by vaccine group

Vaccine groups	TIVc *N* = 278 *n* (%)	TIV *N* = 148 *n* (%)
Any AE	213 (77)	111 (75)
Any possibly related AE	17 (6)	12 (8)
Any SAE	12 (4)	4 (3)
Any possibly related SAE	0	0
Any NOCD	3 (1)	1(1)
Medically attended AEs	187 (67)	94 (64)
AEs leading to withdrawal	0	0
Death	0	0

TIVc, cell culture-derived trivalent influenza vaccine; TIV, egg-derived trivalent influenza vaccine; AE, adverse event; SAE, serious adverse event; NOCD, new onset of chronic disease.

The types of AEs observed in at-risk children following TIVc vaccination in this study are similar to those reported in healthy children.[16–18] As in healthy subjects, the most common solicited local AE with TIVc in at-risk children was tenderness/pain at the injection site; the most common systemic AE was irritability in younger children (<6 years) and headache and myalgia in older children. Some studies in adults have reported a higher frequency of injection site pain associated with TIVc compared to TIV.[10] However, this was not observed in the present paediatric study. Furthermore, the rates of AEs observed in this study are higher than those reported from a phase III study of the same cell culture-derived vaccine in healthy children and adolescents.[17] This may be because children who are at risk of influenza complications, such as those enrolled in this study, are likely to have comorbidities that could increase their susceptibility to reactions.

The findings from clinical trials involving >5000 healthy children and adolescents and from a large pooled safety analysis from two studies in 4 to <17-year-old healthy children and adolescents have demonstrated that the safety profile of TIVc is similar to that of licensed egg-derived influenza vaccines.[16–18] The results of the present study are consistent with these findings, as they demonstrated that the relative risk estimates for experiencing solicited AEs with TIVc were similar to those with egg-derived vaccine.[18] Cell culture-based manufacturing of influenza vaccines provides a valuable alternative to egg-based systems and can yield a virus that, for some strains, is a closer match to the circulating strains than those produced in eggs;[1,2] therefore it is important to assess the safety of cell culture-derived vaccines in all populations. To date, safety assessments have shown that vaccines produced using MDCK cell lines are non-oncogenic and non-tumorigenic,[19,20] and post-marketing surveillance has shown that the influenza A/H1N1 monovalent adjuvanted cell culture-derived influenza vaccine is not associated with any AEs of special interest.[21]

This study had some limitations. The target sample size was not achieved, primarily because the study started almost a month after influenza vaccinations had begun for the season. As high-risk subjects receive influenza vaccination as a priority, the delayed start affected enrolment. The sample size was also not sufficient to capture rare AEs and no formal statistical comparisons were performed between the two vaccine groups. Nevertheless, these results do provide a substantial insight into the safety of cell culture-derived influenza vaccine in at-risk children.

In conclusion, TIVc had an acceptable tolerability profile in children and adolescents with comorbidities in this study, with no potential safety signals identified with vaccines during the 6-month follow-up. The pattern of AEs observed after TIVc vaccination was similar to that observed with the licensed egg-based TIV in this population. These study results also expand the safety database of TIVc vaccine in the paediatric population.

Author contributions

Richa Chandra and Richard de Rooij participated in the conception and design of the trial. Javier Diez-Domingo, Maurizio De Martino, Jose Garcia-Sicilia Lopez, Gian Vincenzo Zuccotti, Giancarlo Icardi, Alberto Villani, David Moreno-Perez, María Méndez Hernández, and Javier Álvarez Aldean managed study sites and enrolled participants. Richa Chandra, Richard De Rooij, and Ahmed Abdul Mateen performed study management for the study sponsor. Igwebuike Enweonye performed all statistical operations. All authors were involved in the interpretation of analyzed data and the decision to submit for publication, and contributed to the development of the manuscript from the initial draft.

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Ethical approval: The study was conducted in accordance with the Declaration of Helsinki and Good Clinical Practice and was approved by the ethics review committees concerned. Written informed consent was obtained from parents or guardians of all participants, and where applicable informed assent was also sought from the subjects.

Conflict of interest: Ahmed Abdul Mateen is an employee of Novartis Pharmaceuticals Canada Inc. Igwebuike Enweonye and Richard de Rooij were employees of Novartis Pharma BV at the time of study execution, and Richa Chandra was an employee at Novartis Vaccines and Diagnostics, Inc. at the time of study execution. All other authors declare no further potential conflict of interest in this trial.

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