CLINICAL STUDY REPORT

Study title	**Immunogenicity, molecular profiling of a marketed quadrivalent influenza vaccine (Vaxigrip Tetra®) administered by the intramuscular route in participants aged 6 to 8 months.**
Product	Vaxigrip Tetra®
Protocol Number	INCENTIVE-QIV-3-EU
EudraCT Number	2021-003760-27
Clinical Phase	IV
Clinical Indication	Influenza vaccine immunization
Study Description	Phase IV non-randomized vaccine trial of approximately 2-month duration for each recruited participant after vaccination. The intervention includes a dose of Vaxigrip Tetra® intramuscular injection into the thigh muscle on Day 0 and Day 30 with follow-up visits on Days 3 and 58.
Issue Date (Version)	27-OCT-2021 (Version 2)
Study initiation date	19-NOV-2021
Study completion date	26-JUN-2023
Principal investigator	Prof. Tessa Goetghebuer, MD, PhD
Institution	CHU Saint Pierre, Rue Haute, 322 1000 Brussels, Belgium
Sponsor	Université libre de Bruxelles
Sponsor Representative	Prof. Arnaud Marchant, MD, PhD Institut d'Immunologie Médicale Université libre de Bruxelles 900, Route de Lennik 1070 Anderlecht Belgium
GCP compliance	This study was performed in compliance with the International Council for Harmonization GCP, including the archiving of essential documents as well as the ethical principles of the Declaration of Helsinki.
Issue date (version)	20/12/2024 (1.0)

1. SYNOPSIS

Sponsor: Université libre de Bruxelles	
Product: Vaxigrip Tetra®	
Study Title: Immunogenicity, molecular profiling of a marketed quadrivalent influenza vaccine (Vaxigrip Tetra®) administered by the intramuscular route in participants aged 6 to 8 months.	
Principal investigator: Prof. Tessa Goetghebuer	
Study Centre: Centre Hospitalier Universitaire Saint Pierre, Rue Haute, 322 1000 Brussels, Belgium	
Publication (s) (Reference): None at the time of this report.	
Study period: 19 November 2021 (First subject first visit) – 26 Juni 2023 (Last subject last visit)	**Phase of development:** IV

Background and rationale: Influenza causes heavy health and economic toll as it cycles recurrently between the human population and an animal reservoir. Understanding human susceptibility to the influenza virus is essential for reducing and possibly eliminating disease burden. Seasonal flu vaccines must be given annually, with effectiveness varying between 10 and 60%, while failing to adequately protect the most vulnerable - infants, elderly, individuals with co-morbidities and the developing world populations. This is due in large part to the recent recognition that in addition to antigen discovery and vaccine platform optimization, the influenza vaccine problem is primarily a human immunology problem, rooted in our lack of understanding of how to generate broadly protective, long-lasting immunity, in everyone.

The INCENTIVE (Indo-European Consortium for Next Generation Influenza vaccine Innovation) project is funded by the European Union's Horizon 2020 research and innovation program and the Department of Biotechnology (DBT), Govt. of India. The highly integrated INCENTIVE consortium comprises 19 institutions representing a true partnership between Indian and European/United States of America (US) groups that addresses the global health and economic challenge posed by influenza infections, to reduce the worldwide burden resulting from outbreaks. INCENTIVE's strategic goals are to provide seminal knowledge on the underlying mechanisms of poor responsiveness to influenza vaccines in vulnerable individuals and advance the development of two next generation universal influenza vaccines. This study in infants (parallel study in India and Europe) is part of a first step in this INCENTIVE project with phase IV studies in infants, children, and elderly. The aim was to assess immunogenicity in 3 different age groups with the same quadrivalent commercially available influenza vaccine. Immunogenicity evaluation included the traditional immunogenicity as well as more detailed analysis of the immune profile, including in depth analysis of antibodies and their effector functions and cell-mediated immunity, with the goal to identify correlates of responsiveness across populations in EU and India, predicting responses versus non-responses and the quality of responses to influenza vaccines across populations and according to gender. The identification of common or unique determinants of vaccine responses could provide essential guidance to the development of universal influenza vaccines protecting diverse populations.

Objectives and endpoints:	
Objectives	**Endpoints**
Primary	
To measure the level of immune response [HAI - Haemagglutinin Antibody Inhibition titres] of two intramuscular doses of the quadrivalent inactivated influenza vaccine (Vaxigrip Tetra®) in healthy participants aged six to eight months.	• HAI antibody titres on D0 and D58 • Proportion of participants with HAI titres ≥ 40 (1/dilution) at D58 • HAI antibody titres fold increase between D0 and D58 • Proportion of participants with seroconversion (i.e. titre < 10 [1/dilution] at D0 and post-vaccination titre ≥ 40 [1/dilution] at D58, or titre ≥ 10 [1/dilution] at D0 and a ≥ 4-fold increase in titre [1/dilution] at D58 • Proportion of high and low responders (HAI titres <40 (1/dilution) at D58).
Exploratory	
To measure the levels, avidity, biophysical characteristics and functionality of influenza-specific antibodies induced by the vaccine.	**a.** Neutralizing Ab titres will be measured for each vaccine strain with the microneutralization (MN) assay. The analyses will be performed on blood samples obtained on D0 and D58. • Individual MN Ab titre on D0 and D58 • Detectable MN (MN Ab titre ≥ 10 [1/dilution]) at D0, D58 • Proportion of participants with MN Ab titres ≥ 20 (1/dilution), ≥ 40 (1/dilution), ≥ 80 (1/dilution) at D58 • Individual MN Ab titre fold-increase D58 post-vaccination relative to D0 • Fold-increase in MN Ab titre [D58/D0] ≥ 2 and ≥ 4 **b.** Anti-Haemagglutinin (HA) and Neuraminidase (NA) antibody titres to vaccine strain and antibody avidity. • Individual HA and NA Ab titres on D0 and D58 • Detectable HA and NA Ab titre ≥ 10 [1/dilution]) at D0 and D58 • Proportion of participants with HA and NA Ab titres ≥ 20 (1/dilution), ≥ 40 (1/dilution), ≥ 80 (1/dilution) at D58 • Individual HA and NA Ab titre ratio (D58/ D0) • Fold-increase in HA and NA Ab titre [post/pre] ≥ 2 and ≥ 4 at D58 • Avidity index of HA and NA Ab at D0 and D58 **c.** Level (mean fluorescence intensity) and avidity (avidity index) of influenza-specific antibody isotypes at D0 and D58.

	d. Level of influenza-specific antibody isotypes triggering Fc-dependent effector functions (proportion of activated cells, phagocytic score or mean fluorescence intensity) at D0 and D58. **e.** Proportion of influenza-specific IgG Fc expressing individual glycans on D58. **f.** Level of binding (mean fluorescence intensity) of Fc receptors and complement by influenza-specific antibodies at D0 and D58. **g.** Number (n per microliter) and proportions of immune cell subsets in peripheral blood at D0 and D58. **h.** Level of expression of peripheral blood cell mRNA, plasma metabolites and plasma proteins (arbitrary units) at D0 and D3.
Study design: Phase IV non-randomized vaccine trial of approximately 2-month duration for each recruited participant after vaccination. The intervention includes a dose of Vaxigrip Tetra® intramuscular injection into the thigh muscle on Day 0 and Day 30 with follow-up visits on Days 3 and 58.	
Number of Participants: • Screened: 33 • Vaccinated: 33 • Completed study per protocol: 30 • Early withdrawals: 3	
Main Criteria for Inclusion and Exclusion **Main Inclusion Criteria:** Eligible participants must meet all the below criteria at the time of enrolment: • Male or female infants born at ≥ 36 weeks of gestation and aged 6 to 8 months; • Provide written informed consent from parents; • The parents are willing to comply with study protocol requirements, including availability for all scheduled visits of the study; • Subjects are healthy or with well-controlled pre-existing medical conditions by the opinion of the investigator.	
Main Exclusion Criteria: Participants meeting any of the below criteria at the time of enrolment will be ineligible to participate in the trial: • Acute illness, at the time of study vaccine administration (once acute illness is resolved, if appropriate, as per investigator assessment, participant will be re-revaluated for eligibility); • Recorded fever (for eligibility purpose defined as a body temperature greater than 37.5°C) within 3 days prior to study vaccine administration (once fever/acute illness is resolved, if appropriate, as per investigator assessment, participant will be re-evaluated for eligibility;	

- Current or previous, laboratory confirmed case of influenza during the past 6 months, based on anamnesis or medical file (if available) at screening visit;
- Household contact with and/or intimate exposure to an individual with any laboratory confirmed influenza infection during the past 6 months prior to vaccination;
- History of severe allergic reactions after previous vaccinations or hypersensitivity to any study vaccine component;
- Previous history of Guillain Barre Syndrome;
- Any confirmed or suspected condition with impaired/altered function of immune system (e.g. immunodeficient or autoimmune conditions);
- Having tested positive for Human Immuno-deficiency Virus (HIV), Hepatitis B or Hepatitis C;
- Having any bleeding disorder which is considered as a contraindication to intramuscular injection or blood draw according to the opinion of the investigator;
- Chronic administration (defined as more than 14 days) of immunosuppressant or other immune-modifying drugs within three months prior to the study vaccination or planned use throughout the study period. (For corticosteroids, this means prednisone, or equivalent, ≥ 0.5 mg/kg per day. Inhaled, intranasal and topical steroids are allowed;
- Administration of blood, blood products and/or plasma derivatives or any parenteral immunoglobulin preparation in the past 3 months or planned use throughout the study period;
- Administration of any vaccine within 28 days prior to enrolment in the study or planned administration of any vaccine during study participation;
- Use of any investigational or non-registered drug or vaccine within 30 days prior to the administration of study vaccines or planned during the study;
- Having received systemic antibiotic treatment within 3 days prior to enrolment;
- Acute or chronic, clinically significant pulmonary, cardiovascular, metabolic, neurological, hepatic, or renal functional abnormality, as determined by medical history or physical examination if uncontrolled or without appropriate treatment.

Any other condition that in the opinion of the investigator would jeopardize the safety or rights of the volunteer participating in the study or make it unlikely that the participant could complete the protocol.

Test product, Dose and Mode of Administration, batch Number(s):

Vaxigrip Tetra® (Quadrivalent Influenza Vaccine, seasons 2021 – 2022 and 2022 - 2023) is an inactivated quadrivalent influenza vaccine indicated for the prevention of influenza disease caused by influenza types A and B viruses contained in the vaccine.

All subjects received two doses with one month interval by intramuscular injection into the thigh muscle on Day 0 and Day 30. Further details on vaccine contents, manufacture, administration and storage are as per package insert. (https://bijsluiters.fagg-afmps.be/?localeValue=en)

Duration of Treatment:

Participants were vaccinated twice, on Day 0 and Day 30. Study duration for each participant is approximately two months after first vaccination.

Statistical methods:

Safety analyses:

The safety analysis was conducted on the total vaccinated population (TVP). Medical history and SAEs were coded using the Medical Dictionary for Regulatory Activities (MedDRA) (version 21.0 or later). The frequency count and percentage of participants having an event were summarized by system organ class and preferred term.

The laboratory investigations, vital signs and physical examinations were categorized as normal or abnormal. For abnormal findings, the results were indicated as either clinically not significant (CNS) or clinically significant (CS) by one of the investigators.

Immunogenicity Analyses:

The analysis of the primary endpoint was conducted on the according-to-protocol (ATP) population and on the TVP. Geometric mean titres (GMTs), mean geometric increases (MGIs), seroprotection rates and seroconversion rates at D58 as compared to D0 were calculated with their respective 95% and 98.75% confidence interval for HAI titres against influenza strains included in the Vaxigrip Tetra® vaccine.

As the primary endpoint data for India has not yet been finalized, the analyses comparing Belgium and India have not yet been performed. The exploratory endpoint analyses have not yet been performed at the time of this CSR.

Summary and conclusions:

Disposition and Demography:

A total of 33 subjects were enrolled and screened. All 33 participants were subsequentially vaccinated in the study. Of these, 30 vaccinated subjects completed the study. The mean age of the total vaccinated population was 6.5 months and participants were mainly from sub-Saharan African (60.6%) and Caucasian (30.3%) origin.

Safety Results:

No serious adverse events were reported in this study.

Immunogenicity results:

The following results were reported for the primary endpoint analyses:

- The D58 GMTs ranged between 23.3 and 98.5 (for the A/Victoria strain and the B/Phuket strain, respectively).
- The MGIs ranged between 6.7 and 9.1 (for the A/Victoria strain and the B/Washington strain, respectively).
- The proportion of participants with HAI titres ≥ 40 at D58, considered to be a 'high responder' in the protocol, was between 36.0% and 92.0% (for the A/Victoria strain and the B/Phuket strain, respectively).
- The proportion of participants with seroconversion, defined as pre-vaccination HAI titres < 10 and post-vaccination titres ≥ 40 or as pre-vaccination HAI titres ≥ 10 and a ≥ 4-fold increase in HAI titre post-vaccination, ranged between 36.0% % and 76.0% (for the A/Victoria strain and the A/Tasmania and

B/Phuket strain, respectively). The Committee for Medicinal Products for Human use (CHMP) seroconversion criterium of 30% (EMA) was therefore met for all viral strains included in the vaccine.

Conclusion:

The vaccine was well tolerated in all study participants; no serious adverse events were reported. Significant increases in HAI antibodies were elicited between D0 and D58 after vaccination with Vaxigrip Tetra®.

The results of this study with the Vaxigrip Tetra® (2021 – 2022 and 2022 – 2023) vaccine are consistent with the CHMP licensure criteria for HAI-based immunogenicity analyses in infants for the A/Victoria strain, the B/Washington strain, and the B/Phuket strain. Although not included in the vaccines, the criteria were also met for the A/Tasmania/503/2020/H3N2 strain, indicating cross protection for this influenza strain.

Date and version of Report: Final CSR version 1.0, 20DEC2024

2. TABLE OF CONTENTS

3. ABBREVIATIONS AND DEFINITIONS

AE	Adverse event
ATC	Anatomical Therapeutic Chemical Classification
ATP	According-to-protocol
CRO	Contract research organization
CSR	Clinical study report
DBT	Department of biotechnology
DDD	Defined daily dose
ELISA	Enzyme linked immunosorbent assay
ELLA	Enzyme-linked lectin assay
GCP	Good clinical practice
GMT	Geometric mean titre
HA	Hemagglutinin
HAI	Hemagglutination inhibition
HIV	Human immunodeficiency virus
HR	Heart rate
ICF	Informed consent form
ICH	International Council for Harmonisation
IEC	Independent ethics committee
IV	Intravenous
LLOQ	Lower limit of quantification
LSLV	Last subject last visit
MGI	Mean geometric increase
MN	Microneutralization
NK	Natural killer
QIV	Quadrivalent influenza vaccine
SAE	Serious adverse event
SmPC	Summary of product characteristics
TVP	Total vaccinated population

4. ETHICS

4.1 Independent Ethics Committee (IEC)

The study protocol (version 1.1, dated 14 June 2021), the informed consent (version 1.1, dated 14 September 2021), and other information that required pre-approval were reviewed and approved by the Comité d'Ethique Hospitalo-Facultaire Erasme-ULB (808 Route de Lennik, 1070 Anderlecht, Belgium). Approval was obtained before any study-related activities were performed.

4.2 Ethical conduct of the study

The study was conducted in accordance with the protocol, the applicable International Conference on Harmonization (ICH) guidelines for Good Clinical Practice (GCP), the ethical principles that have their origins in the Declaration of Helsinki and applicable laws and regulations.

4.3 Subject information and consent

Study information (consent form) is provided to potential participants either:

- During a usual clinic visit, or
- At the screening visit, by the investigator or the study coordinator.

Study information is provided and explained by the investigator and/or the study coordinator after which participants are provided enough time to review the material. The (sub-)investigator is available at every screening visit to answer any medical or scientific questions. Either the study investigator or the study coordinator may address procedural questions. The written consent will be obtained by the investigator.

Consent was obtained prior to initiating any study procedures and after the participant has reviewed the material and is satisfied that they have had all their questions answered. A copy of the signed consent was given to participants. More specifically, the original consent remained in the participant file.

Sample ICF version 4.0 is provided in Annex I.

5. INVESTIGATORS AND STUDY ADMINISTRATIVE STRUCTURE

5.1 Administrative structure

This single-centre study was conducted at the Centre Hospitalier Universitaire Saint Pierre in Belgium with Prof Dr Tessa Goetghebuer as the principal investigator (PI).

Annex II contains the signature of the PI as well as the signature of the Sponsor's responsible personnel (Prof Dr Arnaud Marchant, sponsor and representative of the Université libre de Bruxelles), indicating that this clinical study report CSR) accurately describes the conduct and results of this study.

5.1.1 Sponsor

Université libre de Bruxelles

5.2 Sponsor representative

Principal investigator
Tessa Goetghebuer, MD, PhD
Centre Hospitalier Universitaire Saint Pierre
322, rue Haute, 1000 Bruxelles, Belgium

Co-investigator

Arnaud Marchant, MD, PhD
Institut d'immunologie medical
Université libre de Bruxelles
900 Route de Lennik, 1070 Anderlecht

5.3 CRO activities and pharmacovigilance

Harmony Clinical Research
Adequate Business Centre
Brusselsesteenweg 159
9090 Melle
Belgium

5.4 Biostatistics

Statistical analyses performed by Pieter Pannus, Université libre de Bruxelles.

5.5 Clinical laboratories

TABLE 1: CLINICAL LABORATORIES RESPONSIBLE FOR THE DIFFERENT ANALYSES CONDUCTED IN THE STUDY.

Type of Analysis	**Responsible Laboratories**
Conventional serology	Influenza Centre Department of Clinical Science University of Bergen (UiB) 5th floor Laboratory Building Jonas Lies vei 87 N-5021 Bergen, Norway
Systems serology	Barcelona Institute for Global Health Hospital Clinic (ISGlobal) Universitat de Barcelona Carrer Roselló 153 (CEK building) E-08036 Barcelona, Catalonia, Spain Institute for Medical Immunology Campus Erasme, Université libre de Bruxelles (ULB) 808 Route de Lennik 1070 Brussels, Belgium
Transcriptome, proteome and metabolome analysis	Bioaster Fondation de Cooperation Scientifique 40 Avenue Tony Garnier 69007 Lyon, France
T and B cell analyses	Helmholtz Zentrum für Infektionsforschung GmbH (HZI) Dept. of Vaccinology and Applied Microbiology Inhoffenstr. 7 38124 Braunschweig, Germany
Immune cell analyses	Institut Pasteur (IP) Translational Immunology Lab Rue du Docteur Roux 25-28 75724 Paris Cedex 15, France

6. INTRODUCTION

6.1 Background

Influenza causes a heavy health and economic toll as it cycles recurrently between the human population and the animal reservoir. Understanding human susceptibility to the influenza virus is essential for reducing and possibly eliminating disease burden.

While significant progress in understanding the influenza virus has led to the development of multiple vaccines over the past 50 years, there has been limited progress in improving the breadth and length of the protection conferred by influenza vaccines. Seasonal flu vaccines are given annually, with effectiveness varying between 10-60%, while failing to adequately protect the most vulnerable - infants, elderly, individuals with co-morbidities and the developing world populations. This is due in large part to the recent recognition that in addition to antigen discovery and vaccine platform optimization, the influenza vaccine problem is primarily a human immunology problem, rooted in our lack of understanding of how to generate broadly protective, long-lasting immunity, in everyone. Thus, solving this problem is an essential prerequisite for development of a safe and effective universal influenza vaccine. There has been significant investment in influenza vaccine development over the past decade. However, the majority of funding has gone towards broadening manufacturing capabilities for seasonal vaccines and developing vaccines aimed at making incremental improvements to seasonal vaccines. In contrast, a paucity of resources has been directed towards deciphering the correlates and mechanisms of protective immunity [Berlanda Scorza 2016][1]. The recently constituted Global Funders Consortium for Universal Influenza Vaccine Development, consisting of key global stakeholders reinforced this issue, concluding that it is critical for universal influenza vaccine development to define the correlates of protective immunity and understand how to address immunologic imprinting. The recent convergence of advances across the biomedical, engineering and computational sciences have now enabled the ability to understand the human immune system's response to influenza virus and vaccines in ways not thought possible even five years ago.

The INCENTIVE (Indo-European Consortium for Next Generation Influenza vaccine Innovation) project is funded by the European Union's Horizon 2020 research and innovation program and the Department of Biotechnology (DBT), Govt. of India. The highly integrated INCENTIVE consortium comprises 19 institutions representing a true partnership between Indian and European/United States of America (US) groups that addresses the global health and economic challenge posed by influenza infections, to reduce the worldwide burden resulting from outbreaks. INCENTIVE's strategic goals are to provide seminal knowledge on the underlying mechanisms of poor responsiveness to influenza vaccines in vulnerable individuals and advance the development of two next generation universal influenza vaccines.

[1] Scorza FB et all, Universal influenza vaccines: Shifting to better vaccines. Vaccine. 2016 Jun 3; 34(26): 2926-2933

6.2 Rationale for the study and study design

This study in infants (parallel study in India and Europe) is part of a first step in this INCENTIVE project with phase IV studies in infants, children, and elderly. Its aim was to assess immunogenicity in three different age groups with the same quadrivalent commercially available influenza vaccine, including an analysis of gender-disaggregated data to better understand differences in the responses to the vaccine in terms of gender. The parallel trial in India and Europe would allow comparison in immune response between different continents. Immune response will cover the traditional immunogenicity as well as more in-depth analysis of the immune profile, including in depth analysis of antibodies and their effector functions and cell-mediated immunity. Immunome profiling at baseline and post immunization with standard influenza vaccines will identify correlates of responsiveness across populations in EU and India, predicting responses versus non-responses and the quality of responses to influenza vaccines across populations and according to gender.

By applying multimodal influenza immunity assessments and systems biology analyses of baseline immune states, the aim of INCENTIVE was to identify novel determinants of responses to standard and next-generation influenza vaccines across age groups in populations living in different environments. The identification of common or unique determinants of vaccine responses will provide essential guidance to the development of universal influenza vaccines protecting diverse populations.

The present study was a Phase IV, open-label vaccine trial with the quadrivalent vaccine Vaxigrip Tetra®, marketed by Sanofi Pasteur. In this ambulatory study, 31 healthy infant participants aged 6 to 8 months were enrolled in Belgium. In India, a similar (separate) study with the same study design is ongoing. The vaccine was given on Day 0 and Day 30 with a postvaccination observation period of 30 minutes. Subjects came for follow up visits on Days 3 and 58. SAEs related to study procedures or vaccination have been collected until the end of study.

Titres of influenza-specific antibodies were measured, and their biophysical and functional profile will be analysed to assess the magnitude and quality of vaccine responses. In parallel, profiling of immune cells and plasma will be performed to identify predictors of vaccine responses.

In this CSR, results of the primary objectives are presented. Results of exploratory objectives will be presented in a CSR amendment as soon as results become available.

6.3 Risk/benefit analysis

6.3.1 Potential risks

For information regarding potential risks, please refer to the Summary of Product Characteristics (SmPc) in Annex III.

6.3.2 Potential benefits

Subjects may have the benefit of protection against seasonal influenza A and B.

7. STUDY OBJECTIVES

Objectives	Endpoints
Primary	

To measure the level of immune response [HAI - Haemagglutinin Antibody Inhibition titres] of two intramuscular doses of the quadrivalent inactivated influenza vaccine (Vaxigrip Tetra®) in healthy participants aged six to eight months.	• HAI antibody titres on D0 and D58 • Proportion of participants with HAI titres ≥ 40 (1/dilution) at D58 • HAI antibody titres fold increase between D0 and D58 • Proportion of participants with seroconversion (i.e. titre < 10 [1/dilution] at D0 and post-vaccination titre ≥ 40 [1/dilution] at D58, or titre ≥ 10 [1/dilution] at D0 and a ≥ 4-fold increase in titre [1/dilution] at D58 • Proportion of high and low responders (HAI titres <40 (1/dilution) at D58).
Exploratory	
To measure the levels, avidity, biophysical characteristics and functionality of influenza-specific antibodies induced by the vaccine.	**a.** Neutralizing Ab titres will be measured for each vaccine strain with the microneutralization (MN) assay. The analyses will be performed on blood samples obtained on D0 and D58. • Individual MN Ab titre on D0 and D58 • Detectable MN (MN Ab titre ≥ 10 [1/dilution]) at D0, D58 • Proportion of participants with MN Ab titres ≥ 20 (1/dilution), ≥ 40 (1/dilution), ≥ 80 (1/dilution) at D58 • Individual MN Ab titre fold-increase D58 post-vaccination relative to D0 • Fold-increase in MN Ab titre [D58/D0] ≥ 2 and ≥ 4 **b.** Anti-Haemagglutinin (HA) and Neuraminidase (NA) antibody titres to vaccine strain and antibody avidity. • Individual HA and NA Ab titres on D0 and D58 • Detectable HA and NA Ab titre ≥ 10 [1/dilution]) at D0 and D58 • Proportion of participants with HA and NA Ab titres ≥ 20 (1/dilution), ≥ 40 (1/dilution), ≥ 80 (1/dilution) at D58 • Individual HA and NA Ab titre ratio (D58/ D0) • Fold-increase in HA and NA Ab titre [post/pre] ≥ 2 and ≥ 4 at D58 • Avidity index of HA and NA Ab at D0 and D58 **c.** Level (mean fluorescence intensity) and avidity (avidity index) of influenza-specific antibody isotypes at D0 and D58. **d.** Level of influenza-specific antibody isotypes triggering Fc-dependent effector functions (proportion of activated cells, phagocytic score or mean fluorescence intensity) at D0 and D58.

	e. Proportion of influenza-specific IgG Fc expressing individual glycans on D58. **f.** Level of binding (mean fluorescence intensity) of Fc receptors and complement by influenza-specific antibodies at D0 and D30. **g.** Number (n per microliter) and proportions of immune cell subsets in peripheral blood at D0 and D58. **h.** Level of expression of peripheral blood cell mRNA, plasma metabolites and plasma proteins (arbitrary units) at D0 and D3.

8. INVESTIGATIONAL PLAN

8.1 Study Design

This was a Phase IV non-randomized vaccine trial of approximately 2-month duration after vaccination for each recruited participant. Thirty-one healthy infants aged 6 to 8 months were enrolled in Belgium. In India a similar study with the same study design will be conducted in 100 participants.

Vaxigrip Tetra® was administered as 2 doses with 1 month interval into the thigh muscle on Day 0 and Day 30 with follow-up visits on Days 3 and 58. Further details on the vaccine contents, manufacture, administration and storage will be as per the package insert (via the link https://bijsluiters.fagg-afmps.be/?localeValue=en). Six ml of blood was collected at each visit (D0, D3, D58) for immunological analyses. Study related SAEs were collected until the end of study.

8.2 Selection of study population

8.2.1 Inclusion Criteria

Eligible participants had to meet all the below criteria at the time of enrolment:

1. Male or female infants born at ≥ 36 weeks of gestation and aged 6 to 8 months.
2. Provide written informed consent from the parents.
3. The parents are willing to comply with study protocol requirements, including availability for all scheduled visits of the study.
4. Subjects are healthy or with well-controlled pre-existing medical conditions by the opinion of the investigator.

8.2.2 Exclusion Criteria

Participants meeting any of the below criteria at the time of enrolment were ineligible to participate in the trial:

1. Acute illness, at the time of study vaccine administration (once acute illness is resolved, if appropriate, as per investigator assessment, participant will be re-revaluated for eligibility).
2. Recorded fever (for eligibility purpose defined as a body temperature greater than 37.5°C) within 3 days prior to study vaccine administration (once fever/acute illness is resolved, if appropriate, as per investigator assessment, participant will be re-evaluated for eligibility.

3. Current or previous, laboratory confirmed case of influenza during the past 6 months, based on anamnesis or medical file (if available) at screening visit.
4. Household contact with and/or intimate exposure to an individual with any laboratory confirmed influenza infection during the past 6 months prior to vaccination.
5. History of severe allergic reactions after previous vaccinations or hypersensitivity to any study vaccine component.
6. Previous history of Guillain Barre Syndrome.
7. Any confirmed or suspected condition with impaired/altered function of immune system (e.g. immunodeficient or autoimmune conditions).
8. Having tested positive for Human Immuno-deficiency Virus (HIV), Hepatitis B or Hepatitis C
9. Having any bleeding disorder which is considered as a contraindication to intramuscular injection or blood draw according to the opinion of the investigator.
10. Chronic administration (defined as more than 14 days) of immunosuppressant or other immune-modifying drugs within three months prior to the study vaccination or planned use throughout the study period. (For corticosteroids, this means prednisone, or equivalent, ≥ 0.5 mg/kg per day. Inhaled, intranasal and topical steroids are allowed).
11. Administration of blood, blood products and/or plasma derivatives or any parenteral immunoglobulin preparation in the past 3 months or planned use throughout the study period.
12. Administration of any vaccine within 28 days prior to enrolment in the study or planned administration of any vaccine during study participation.
13. Use of any investigational or non-registered drug or vaccine within 30 days prior to the administration of study vaccines or planned during the study.
14. Have received systemic antibiotic treatment within 3 days prior to enrolment.
15. Acute or chronic, clinically significant pulmonary, cardiovascular, metabolic, neurological, hepatic, or renal functional abnormality, as determined by medical history or physical examination if uncontrolled or without appropriate treatment.
16. Any other condition that in the opinion of the investigator would jeopardize the safety or rights of the volunteer participating in the study or make it unlikely that the participant could complete the protocol.

8.2.3 Criteria for Elimination from the According-To-Protocol Population

Subjects meeting the following criteria were excluded from the ATP population (see section 8.7.2):

- any disease or therapy that could significantly affect the subject's immune status;
- administration of any vaccine other than the study vaccine within 28 days of receipt of study vaccine and during the entire study period;
- lab-confirmed influenza disease in the previous six months before study vaccination.

These subjects were included in the Total Vaccinated Population (see section 8.7.2) and might have continued in the study for safety follow-up.

8.2.4 Contraindications to Vaccination

The following AE constitutes a temporary contraindication to administration of the study vaccine:

- acute febrile illness in a period of three days before vaccination deemed by the Investigator to be a contraindication for vaccination.

8.3 Composition and administration of vaccine

8.3.1 Study vaccine

Vaxigrip Tetra® (Quadrivalent Influenza Vaccine) is an inactivated quadrivalent influenza vaccine indicated for the prevention of influenza disease caused by influenza types A and B viruses contained in the vaccine. The vaccine is provided as a suspension for injection in pre-filled syringes. After gently shaking the vaccine is a colourless opalescent liquid. All subjects received a dose (0.5 ml) on Day 0 by intramuscular injection into the thigh muscle followed by a second dose at Day 30 (if fever, postpone until three days without fever). In this study, Vaxigrip Tetra® vaccines from seasons 2021-2022 and 2022-2023 were used in the respective flu seasons. Product specifications of both vaccines can be found in Annex III and on the website of the FAMHP via the link https://bijsluiters.fagg-afmps.be/?localeValue=en.

8.3.2 Packaging and Labelling

For detailed information on the packaging, we refer to the SmPCs (see Annex III). The study vaccine was labelled for use in clinical trials by the pharmacist of the site before study start. These labels are part of the submission package for the regulatory authorities.

8.3.3 Storage and Vaccine Accountability

The Investigator (or his/her designee) is responsible for the safe storage of all study vaccines assigned to the clinical site, in a locked, secure storage facility with access limited to those individuals authorized to dispense the study vaccine and maintained within the appropriate ranges of temperature. All study vaccines must be stored as specified at delivery and in the original packaging. Vaxigrip Tetra® should be stored at the defined temperature (i.e. +2 to +8°C).

Regular temperature logging of the study vaccine storage room at the clinical site should be performed. In case a deviation in storage conditions should occur, the clinical site must not further dispense the affected study vaccine and notify the Sponsor.

The Investigator is responsible for ensuring that all study vaccines received at the clinical site are inventoried and accounted for throughout the study.

Study vaccines should be dispensed under the supervision of the Investigator, a qualified member of the clinical staff, or by a hospital/clinic pharmacist. The Investigator must maintain accurate records demonstrating date and amount of vaccine administered to whom and by who. Study vaccine will be supplied only to subjects participating in the study.

The Sponsor's designated site monitor will periodically check the supplies of study vaccine held by the Investigator or pharmacist to ensure accountability and appropriate storage conditions of all study vaccines used. Unused study vaccine must be available for verification by the site monitor during on-site monitoring visits.

After the last visit of the last subject in the study (LSLV), any used and unused study vaccine will be returned to the Sponsor or destroyed at the clinical site with the Sponsor's written permission (in this case a certificate of destruction will be provided and filed in the Trial Master File [TMF]).

8.4 Blinding

No blinding procedures were applicable as this was an open-label study. All subjects received two doses of vaccine.

8.5 Prior and Concomitant Therapy

8.5.1 Prior medications and vaccines

Any medications (including vaccines) that were administered to the participant within 28 days prior to the study vaccination were considered as prior medications for this study. These were recorded in the eCRF.

8.5.2 Concomitant medications and vaccines

At each study visit, the investigator/designee asked the participants about any prescription or over-the-counter medication(s) taken since the last visit. Only medication to treat study related SAEs or medication that can alter the immune status were recorded in the source docs and eCRF with trade and/or generic name, indication, dose, start and end dates.

Any treatments and/or medications specifically contraindicated, e.g., any investigational or non-registered product, any immunosuppressant and immune-modifying drug including systemic steroids, any immunoglobulin and blood product were checked at each study visit after the study vaccination. If any became applicable during the study, it did not require withdrawal of the participant from the study but may have determined a participant's evaluability in the per-protocol analysis.

Any vaccine not foreseen in the study protocol in the period starting at Visit 1 (Day 1) and ending at end of study visits was recorded in the eCRF.

8.5.3 Compliance

All study vaccine administrations were supervised by the Investigator or his/her designee.

8.6 Assessment of safety and immunogenicity variables

8.6.1 Safety and immunogenicity measurements

Blood samples for immunogenicity were taken at days 0, 3, and 58. Immunogenicity measurements have been performed at different laboratories of the INCENTIVE consortium as shown in Table 2 below. The processing, labelling and storage of blood samples for immunological analyses was done on site based on the laboratory manual kept in place before the recruitment of the first participant. Detailed descriptions of the collection, handling, transport and processing of the blood samples are included in the laboratory manual. Sample shipments outside of Belgium were as per the existing regulatory guidelines at the time of shipment.

TABLE 2: INCENTIVE-QIV-3-EU IMMUNOLOGICAL MEASUREMENTS

Type of Analysis	Responsible Laboratories	Time points
Conventional serology	UiB	Day 0, Day 58
Systems serology	ISGlobal, ULB	Day 0, Day 58
Immune cell analysis	HZI, BioAster	Day 0, Day 58

Transcriptome, proteome and metabolome analysis	BioAster	Day 0, Day 3

Primary immunogenicity variable

- **Hemagglutination Inhibition Assay (HAI):** to measure responses to relevant influenza strains. The HAI method exploits the property of the influenza virus to hemagglutinate red blood cells. HAI titres are determined as the highest serum dilution inhibiting hemagglutination. An HAI titre of ≥40 is considered protective in adults. The primary efficacy is evaluated based on HAI titre at Day 58.

Note: Participants were recruited across two influenza seasons (2021-2022 and 2022-2023) and, consequently, received different vaccine formulations. Annex IV provides a detailed summary of the viral strains included in each vaccine, the viral strains tested in the HAI assay, and an evaluation of their antigenic overlap. The interpretation of the immunogenicity data presented in this report should consider these partial overlaps.

Exploratory immunogenicity variables

- **ELISA**: to measure IgG titres to HA and NA. IgG avidity were measured using chaotropic salts.
- **Enzyme-Linked Lectin Assay (ELLA):** for the detection of NA-specific antibodies using reverse genetics viruses with irrelevant HA to ensure only NA antibodies are measured.
- **Microneutralization (MN) assay:** to determine cross-reactivity to related virus strains in serum samples with detectable HAI titres.
- **Quantitative suspension array technology (qSAT)** applying the xMAP™ technology (Luminex Corp., TX): to measure IgG subclasses and their avidity to multiple influenza HA and NA antigens; to assess binding of influenza-specific IgG to Fc receptors and complement.
- **Capillary electrophoresis**: to profile IgG Fc glycans following affinity purification of influenza HA and NA specific antibodies.
- **Advanced multi-parametric flow cytometry:** to assess the proportion and phenotype of peripheral blood immune cells; to assess the interactions between influenza-specific IgG and innate immune cells (macrophages, neutrophils, NK cells).
- **Illumina NextSeq 500 system**: Transcriptomic profiling by Next Generation sequencing (RNA-seq). Bioinformatics data processing provide relative quantification and lists of differentially expressed genes between time points and study subjects.
- **Metabolomics and Proteomics:** Metabolomics consist of a comprehensive untargeted analysis of both polar and non-polar metabolites in plasma, that are analysed using an Ascend 600MHz NMR spectrometer from Bruker and a Q-Exactive (Thermo Scientific) HR mass spectrometer, respectively.

- **CyTOF system from Fluidigm**: to perform mass cytometry and together with an established panel of markers allowing comprehensive analysis of immune cell populations.

Safety measurements

- **Adverse events**: As Vaxigrip Tetra® is a marketed vaccine, only study related serious adverse events (SAEs) were recorded in the study. At regular intervals during the study, subjects were asked non-leading questions to determine the occurrence of SAEs.
- **Vital signs**: Vital sign parameters were recorded after 5 minutes of rest at days 0, 3, 30, and 58. The signs include systolic and diastolic blood pressure, pulse rate, respiratory rate, and oral body temperature. Any abnormal values were recorded in the eCRF and source documents.

8.6.2 Criteria for delayed blood sampling

After enrolment, participants may encounter clinical circumstances that warrant a delay in blood collection for immunogenicity assessments in this study. This situation is listed below. In case a participant meets a criterion for delay of blood collection, blood collection may proceed once the window for delay has passed. This is applicable only for Day 58.

- Participant has received a dose of systemic antibiotics within three days before the intended blood collection: if this occurs for any timepoint of blood sampling, the sponsor should be informed. To reduce the risk of delay for the first three blood samples, recorded fever (for eligibility purpose defined as a body temperature greater than 37.5°C) within three days prior to study vaccine administration will be temporarily excluded. Once fever/acute illness is resolved, if appropriate, as per investigator assessment, the participant will be re-evaluated for eligibility.

8.6.3 Data Quality Assurance

Several quality assurance and quality control systems were implemented to ensure the integrity and reliability of the data. Standard terminology and data consistency were maintained through a comprehensive information session for investigators, detailed instructions in the laboratory manual, and periodic monitoring by sponsor personnel. Data accuracy was further ensured by using central laboratories for all tests and measurements. Investigator meetings were conducted to standardise performance and prepare all participating sites.

8.7 Statistical Methods Planned in the Protocol and Determination of Sample Size

8.7.1 Statistical Analysis Plan

All statistical analyses are performed using Graphpad Prism 10. Medical history and SAEs are coded using the Medical Dictionary for Regulatory Activities (MedDRA) (version 21.0 or later). The frequency count and percentage of participants is summarized by system organ class and preferred term. Lab results, vital signs and physical examination are listed descriptively.

8.7.2 Analysis Populations

Enrolled Population

All participants who provide written informed consent, regardless of the participant's screening, randomization and treatment status in the study. This population will be used to account fully for participant disposition, starting with the informed consent.

Total Vaccinated Population (TVP)

Participants who have received a study vaccination and have post-vaccination immunogenicity measurements. The analysis based on this population will serve as supportive results for the immunogenicity endpoints.

According-To-Protocol population (ATP)

All participants in the total vaccinated population who have no major protocol deviations that are determined to potentially interfere with the immunogenicity assessment of the vaccines. This population will serve as the primary analysis population for the primary immunogenicity endpoints. The criteria for exclusion of participants from the ATP will be established before database lock.

Safety Population

All participants in the enrolled population who received vaccine and have any safety data available. All safety analyses will be performed using this population.

8.7.3 Analysis Plan

Summary of baseline data of subjects

Descriptive summary statistics will be presented for demographic (race, age, gender, height, weight) and baseline characteristics (medical history, pre-existing conditions and prior medication). Continuous variables are summarized by means and standard deviations for symmetrically distributed variables and medians and inter-quartile ranges otherwise. Categorical variables are presented as frequencies and percentages.

Primary endpoint analysis

The primary endpoint analysis is done for the ATP and the TVP. No imputation is made for missing values in HAI titres, except that values below the lower limit of quantitation (LLOQ) are assigned a value of ½ the limit of quantification.

The **Geometric Mean Titre (GMT)** for a given subset of participants and specific influenza strain was defined as the anti-log of the arithmetic mean of logarithmic transformed (base 10) HAI titres.

The **Mean Geometric Increase (MGI)** for a given subset of participants and specific influenza strain was defined as the anti-log of the arithmetic mean of logarithmic transformed (base 10) within-subject ratios of the Day 58 HAI titre to the Day 0 HAI titre.

The **seroprotection rate** for a given subset of participants and specific influenza strain was defined as the percentage of participants with an HAI titre ≥ 40.

The **seroconversion rate** for a given subset of participants and specific influenza strain was defined as the percentage of participants with a Day 28 HAI titre ≥ 40 if they had a Day 0 HAI titre < 10, or with a ≥ 4-fold increase of the Day 28 HAI titre compared to the Day 0 HAI otherwise.

Exploratory endpoint analysis

Exploratory endpoints are not in the scope of this CSR and will therefore not be reported on here.

Safety analyses

The safety analyses were performed on the Safety Population (see 8.7.2). The occurrence of serious adverse events was described.

8.7.4 Handling of Dropouts and Missing Data

Generally, no imputation is made for missing values in safety and immunogenicity analyses, except that immunogenicity values below the lower limit of quantitation (LLOQ) that are reported as "< LLOQ" are assigned a value of ½ the limit of quantification.

8.7.5 Determination of Sample Size

There was no formal sample size calculation done as it was a phase IV non-randomized pre- & post- study. The plan was to enrol 50 participants. This study in infants is part of a larger INCENTIVE phase IV study that will evaluate similar quadrivalent commercially available influenza vaccines in not only infants but also in elderly and children. In India, Fluquadri manufactured by Sanofi India will be used which is produced according to the same manufacturing procedure as Vaxigrip Tetra® and composition of influenza strains are the same for both vaccines.

9. STUDY PATIENTS

9.1 Study dates and participant enrolment

Study participants were enrolled between 19 November 2021 and 31 March 2023. The last participant completed the last study visit on 26 May 2023. A first group of 12 participants was enrolled in the 2021-2022 season, and a second group of 21 participants was enrolled in the 2022-2023 season.

9.2 Participant disposition

9.2.1 Number of participants

A total of 33 participants were screened, enrolled, and vaccinated in the study. Therefore, the enrolled population equals the TVP and safety population. Of these 33, 31 returned to the second study visit. The other 2 participants were terminated early by the sponsor due to an error in the administered vaccine (n=1) and the failure of collecting blood from the participant at the first visit (n=1). All 31 remaining participants returned to the third visit, but 1 participant was lost to follow-up before the fourth and final visit. Therefore, the ATP population includes 30 participants.

9.2.2 Protocol deviations

There have been no protocol deviations that were deemed to affect the participant's data related to the primary endpoints. Therefore, they did not result in exclusion from the analysis for the primary endpoints.

9.3 Demographic characteristics

In the total vaccinated population, 54.5 % of the participants were male and 45.5 % of the participants were female. The median age of the participants was 6 months. In terms of origin, 60.6% was sub-Saharan African, 30.3% was Caucasian, the remaining 9.1% were of different origins.

In the per protocol population, 53.3 % of the participants was of the male gender and 46.7 % of the participants was of the female gender. The median age was 6.5 months. In terms of origin, 63.3% was sub-Saharan African, 30.0% was Caucasian, the remaining 6.7% were of different origins.

9.4 Clinical characteristics

9.4.1 Medical history

In the TVP, a total of 46 events were reported in the medical history. The most frequently reported event was maternal pertussis vaccination (n=25, 75.8%). All other events are medical conditions commonly seen in early childhood. The most frequently reported respiratory issues (n=8, 24.2%) included bronchiolitis, spastic bronchiolitis, rhinitis, mucopurulent rhinitis, and more severe infections such as bronchopneumonia and pneumonia. Additionally, viral infections with fever and conjunctivitis were also reported (n=3, 6.1%).

Dermatological issues (n=3, 9.1%) included atopic dermatitis, hives (urticaria), and impetigo (two lesions). Structural or congenital conditions (n=4, 12.1%) included bilateral dilatation of the urinary tract, pyelocaliceal dilatation, an umbilical hernia, and a right preauricular infantile hemangioma.

Other reported conditions (n=3, 9.1%) included hypotonia (reduced muscle tone), spherocytosis (a hereditary blood disorder), and symptoms such as nasal congestion. This medical history reflects a broad range of common infectious, congenital, and inflammatory conditions in infants.

9.4.2 Concomitant medication

In the TVP, the use of a total of 57 drugs was reported during the first visit. The most frequently reported classes of medication, as per the first level of the ATC/DDD classification, were systemic hormonal preparations (n=23, 40.3%), analgesics (n=7, 12.3%), and antibiotics for systemic use (n=5, 8.8%).

9.4.3 Vital signs and physical examination

Vital signs were evaluated during each study visit. None of the participants had any clinically significant abnormality in terms of vital signs throughout the entire study.

A physical examination was also performed during each study visit. At Visit 1 and 2, no clinically significant abnormalities were registered. At Visit 3, three abnormalities (bilateral red tympanum and rhinitis, bilateral red tympanum, and otitis) were deemed clinically significant. At Visit 4, two abnormalities (bilateral conjunctivitis, and hypotonia) were deemed clinically significant. None of the reported clinically significant abnormalities became serious during the study.

10. IMMUNOGENICITY RESULTS

10.1 Data sets analysed

The statistical analyses were performed on both the ATP and the TVP.

10.2 According-To-Protocol population

10.2.1 Descriptive statistics

Descriptive statistics for HAI antibodies against influenza strains included in the Vaxigrip Tetra® vaccine are presented in tables 3 and 4.

TABLE 3: DESCRIPTIVE STATISTICS FOR THE HAI TITRES AGAINST INFLUENZA STRAINS INCLUDED IN THE VAXIGRIP TETRA® VACCINE IN THE ACCORDING-TO-PROTOCOL (ATP) POPULATION

Strain	Timing	N	Avg	SD	Min	q25	q50	q75	Max	Pos
A/Victoria/2570/2019/ H1N1	Day 0	29	8.0	13.7	5	5	5	5	80	4 (13.8%)
	Day 58	25	54.0	123.7	5	10	20	40	640	22 (88.0%)
A/Tasmania/503/2020/ H3N2	Day 0	29	16.7	33.1	5	5	5	5	160	6 (20.7%)
	Day 58	25	151.2	285.5	5	40	40	80	1280	24 (96.0%)
B/Washington/02/2019	Day 0	29	9.4	15.0	5	5	5	5	80	5 (17.2%)
	Day 58	24	72.3	75.8	5	37	40	80	320	23 (95.8%)
B/Phuket/3073/2013	Day 0	29	20.7	31.1	5	5	10	20	160	21 (72.4%)
	Day 58	25	146.4	144.0	10	80	80	160	640	25 (100.0%)

TABLE 4: DESCRIPTIVE STATISTICS FOR THE LOG10-TRANSFORMED HAI TITRES AGAINST INFLUENZA STRAINS INCLUDED IN THE VAXIGRIP TETRA® VACCINE IN THE ACCORDING-TO-PROTOCOL (ATP) POPULATION

Strain	Timing	N	Avg	SD	Min	q25	q50	q75	Max	Pos
A/Victoria/2570/2019/ H1H1	Day 0	29	1.8	0.54	1.6	1.6	1.6	1.6	4.4	4 (13.8%)
	Day 58	25	3.2	1.1	1.6	2.3	3.0	3.7	6.5	22 (88.0%)
A/Tasmania/503/2020/ H3N2	Day 0	29	2.0	0.97	1.6	1.6	1.6	1.6	5.1	6 (20.7%)
	Day 58	25	4.0	1.3	1.6	3.7	3.7	4.4	7.2	24 (96.0%)
B/Washington/02/2019	Day 0	29	1.9	0.67	1.6	1.6	1.6	1.6	4.4	5 (17.2%)
	Day 58	24	3.8	0.98	1.6	3.6	3.7	4.4	5.8	23 (95.8%)
B/Phuket/3073/2013	Day 0	29	2.5	0.91	1.6	1.6	2.3	3.0	5.1	21 (72.4%)
	Day 58	25	4.6	0.93	2.3	4.4	4.4	5.1	6.5	25 (100.0%)

10.2.2 Geometric mean titres

TABLE 52: GEOMETRIC MEAN TITRES FOR THE HAI TITRES AGAINST INFLUENZA STRAINS INCLUDED IN THE VAXIGRIP TETRA® VACCINE IN THE ACCORDING-TO-PROTOCOL (ATP) POPULATION

Strain	Timing	N	GMT	GMT (95% CI)		GMT (98.75% CI)	
				LL	UL	LL	UL
A/Victoria/2570/2019/H1H1	Day 0	29	5.8	4.8	7.1	4.7	7.3
	Day 58	25	23.3	15.2	35.7	14.3	37.9
A/Tasmania/503/2020/H3N2	Day 0	29	7.7	5.4	10.9	5.1	11.5
	Day 58	25	55.7	33.5	92.7	31.1	99.7
B/Washington/02/2019	Day 0	29	6.3	5.0	8.1	4.8	8.4
	Day 58	24	46.2	31.2	68.3	29.5	72.3
B/Phuket/3073/2013	Day 0	29	12.1	8.7	16.8	8.3	17.7
	Day 58	25	98.5	68.3	142.0	64.8	149.6

10.2.3 Mean geometric increase

TABLE 63: MEAN GEOMETRIC INCREASE FOR THE HAI TITRES AGAINST INFLUENZA STRAINS INCLUDED IN THE VAXIGRIP TETRA® VACCINE IN THE ACCORDING-TO-PROTOCOL (ATP) POPULATION

Strain	Timing	N	MGI	MGI (95 % CI)		MGI (98.75 % CI)	
				LL	UL	LL	UL
A/Victoria/2570/2019/H1H1	Day 58	25	6.7	5.1	8.9	4.9	9.3
A/Tasmania/503/2020/H3N2	Day 58	25	8.9	6.6	12.1	6.4	12.6
B/Washington/02/2019	Day 58	24	9.1	7.2	11.5	7.0	11.9
B/Phuket/3073/2013	Day 58	25	8.2	5.9	11.3	5.6	11.9

10.2.4 Seroprotection rate

TABLE 74: SEROPROTECTION RATES FOR THE HAI TITRES AGAINST INFLUENZA STRAINS INCLUDED IN THE VAXIGRIP TETRA® VACCINE IN THE ACCORDING-TO-PROTOCOL (ATP) POPULATION

Strain	Timing	N	Response (n, %)	Response (95 % CI)		Response (98.75 % CI)	
				LL	UL	LL	UL
A/Victoria/2570/2019/H1H1	Day 0	29	1 (3.4%)	0.0%	10.1%	0.0%	11.0%
	Day 58	25	9 (36.0%)	17.2%	54.8%	14.5%	57.5%
A/Tasmania/503/2020/H3N2	Day 0	29	3 (10.3%)	0.0%	21.4%	0.0%	23.0%
	Day 58	25	19 (76.0%)	59.3%	92.7%	56.9%	95.1%
B/Washington/02/2019	Day 0	29	2 (6.9%)	0.0%	16.1%	0.0%	17.4%
	Day 58	25	18 (75.0%)	57.7%	92.3%	55.2%	94.8%
B/Phuket/3073/2013	Day 0	29	4 (13.8%)	1.2%	26.3%	0.0%	28.1%
	Day 58	25	23 (92.0%)	81.4%	100.0%	79.8%	100.0%

10.2.5 Seroconversion rate

TABLE 85: SEROCONVERSION RATES FOR THE HAI TITRES AGAINST INFLUENZA STRAINS INCLUDED IN THE VAXIGRIP TETRA® VACCINE IN THE ACCORDING-TO-PROTOCOL (ATP) POPULATION

Strain	Timing	N	Response (n, %)	Response (95 % CI)		Response (98.75 % CI)	
				LL	UL	LL	UL
A/Victoria/2570/2019/H1H1	Day 58	25	9 (36.0%)	17.2%	54.8%	14.5%	57.5%
A/Tasmania/503/2020/H3N2	Day 58	25	19 (76.0%)	59.3%	92.7%	56.9%	95.1%
B/Washington/02/2019	Day 58	24	18 (75.0%)	57.7%	92.3%	55.2%	94.8%
B/Phuket/3073/2013	Day 58	25	19 (76.0%)	59.3%	92.7%	56.9%	95.1%

10.3 Total vaccinated population

10.3.1 Descriptive statistics

Descriptive statistics for HAI antibodies against influenza strains included in the Vaxigrip Tetra® vaccine are presented in tables 9 and 10.

TABLE 9: DESCRIPTIVE STATISTICS FOR THE HAI TITRES AGAINST INFLUENZA STRAINS INCLUDED IN THE VAXIGRIP TETRA® VACCINE IN THE TOTAL VACCINATED POPULATION (TVP)

Strain	Timing	N	Avg	SD	Min	q25	q50	q75	Max	Pos
A/Victoria/2570/2019/ H1N1	Day 0	31	7.8	13.2	5	5	5	5	80	4 (12.9%)
	Day 58	25	54.0	123.7	5	10	20	40	640	22 (88.0%)
A/Tasmania/503/2020/ H3N2	Day 0	31	16.0	32.1	5	5	5	5	160	7 (22.6%)
	Day 58	25	151.2	285.5	5	40	40	80	1280	24 (96.0%)
B/Washington/02/2019	Day 0	31	10.3	15.5	5	5	5	5	80	6 (19.4%)
	Day 58	25	72.6	74.3	5	40	40	80	320	24 (96.0%)
B/Phuket/3073/2013	Day 0	31	21.5	30.3	5	6	10	20	160	23 (74.2%)
	Day 58	25	146.4	144.0	10	80	80	160	640	25 (100.0%)

TABLE 10: DESCRIPTIVE STATISTICS FOR THE LOG10-TRANSFORMED HAI TITRES AGAINST INFLUENZA STRAINS INCLUDED IN THE VAXIGRIP TETRA® VACCINE IN THE TOTAL VACCINATED POPULATION (TVP)

Strain	Timing	N	Avg	SD	Min	q25	q50	q75	Max	Pos
A/Victoria/2570/2019/ H1H1	Day 0	31	1.75	0.5	1.6	1.6	1.6	1.6	4.4	4 (12.9%)
	Day 58	25	3.15	1.1	1.6	2.3	3.0	3.7	6.5	22 (88.0%)
A/Tasmania/503/2020/ H3N2	Day 0	31	2.13	1.1	1.6	1.6	1.6	1.8	5.1	7 (22.6%)
	Day 58	25	4.02	1.3	1.6	3.7	3.7	4.4	7.2	24 (96.0%)
B/Washington/02/2019	Day 0	31	1.90	0.7	1.6	1.6	1.6	1.6	4.4	6 (19.4%)
	Day 58	25	3.85	1.0	1.6	3.7	3.7	4.4	5.8	24 (96.0%)
B/Phuket/3073/2013	Day 0	31	2.56	0.9	1.6	1.8	2.3	3.0	5.1	23 (74.2%)
	Day 58	25	4.59	0.9	2.3	4.4	4.4	5.1	6.5	25 (100.0%)

10.3.2 Geometric mean titres

TABLE 116: GEOMETRIC MEAN TITRES FOR THE HAI TITRES AGAINST INFLUENZA STRAINS INCLUDED IN THE VAXIGRIP TETRA® VACCINE IN THE TOTAL VACCINATED POPULATION (TVP)

Strain	Timing	N	GMT	GMT (95% CI)		GMT (98.75% CI)	
				LL	UL	LL	UL
A/Victoria/2570/2019/H1H1	Day 0	31	5.8	4.8	6.9	4.7	7.1
	Day 58	25	23.3	15.2	35.7	14.3	37.9
A/Tasmania/503/2020/H3N2	Day 0	31	7.6	5.8	12.3	5.5	13.0
	Day 58	25	55.7	33.5	92.7	31.1	99.7
B/Washington/02/2019	Day 0	31	6.7	5.2	8.6	5.0	9.0
	Day 58	25	47.2	32.3	68.9	30.6	72.7
B/Phuket/3073/2013	Day 0	31	12.9	9.3	17.8	8.9	18.7
	Day 58	25	98.5	68.3	142.0	64.8	149.6

10.3.3 Mean geometric increase

TABLE 127: MEAN GEOMETRIC INCREASE FOR THE HAI TITRES AGAINST INFLUENZA STRAINS INCLUDED IN THE VAXIGRIP TETRA® VACCINE IN THE TOTAL VACCINATED POPULATION (TVP)

Strain	Timing	N	MGI	MGI (95 % CI)		MGI (98.75 % CI)	
				LL	UL	LL	UL
A/Victoria/2570/2019/H1H1	Day 58	25	1.91	1.63	2.19	1.59	2.23
A/Tasmania/503/2020/H3N2	Day 58	25	2.19	1.89	2.49	1.85	2.53
B/Washington/02/2019	Day 58	25	2.21	1.98	2.44	1.94	2.47
B/Phuket/3073/2013	Day 58	25	2.10	1.77	2.43	1.72	2.48

10.3.4 Seroprotection rate

TABLE 138: SEROPROTECTION RATES FOR THE HAI TITRES AGAINST INFLUENZA STRAINS INCLUDED IN THE VAXIGRIP TETRA® VACCINE IN THE TOTAL VACCINATED POPULATION (TVP)

Strain	Timing	N	Response (n, %)	Response (95 % CI)		Response (98.75 % CI)	
				LL	UL	LL	UL
A/Victoria/2570/2019/H1H1	Day 0	31	1 (3.2%)	0.0%	9.4%	0.0%	10.3%
	Day 58	25	9 (36.0%)	17.2%	54.8%	14.5%	57.5%
A/Tasmania/503/2020/H3N2	Day 0	31	3 (9.7%)	0.0%	20.1%	0.0%	21.6%
	Day 58	25	19 (76.0%)	59.3%	92.7%	56.9%	95.1%
B/Washington/02/2019	Day 0	31	3 (9.7%)	0.0%	20.1%	0.0%	21.6%
	Day 58	25	19 (76.0%)	59.3%	92.7%	56.9%	95.1%
B/Phuket/3073/2013	Day 0	31	5 (16.1%)	3.2%	29.1%	1.3%	30.9
	Day 58	25	23 (92.0%)	81.4%	100.0%	79.8%	100.0%

10.3.5 Seroconversion rate

TABLE 149: SEROCONVERSION RATES FOR THE HAI TITRES AGAINST INFLUENZA STRAINS INCLUDED IN THE VAXIGRIP TETRA® VACCINE IN THE TOTAL VACCINATED POPULATION (TVP)

Strain	Timing	N	Response (n, %)	Response (95 % CI)		Response (98.75 % CI)	
				LL	UL	LL	UL
A/Victoria/2570/2019/H1H1	Day 58	25	9 (36.0%)	17.2%	54.8	14.5%	57.5%
A/Tasmania/503/2020/H3N2	Day 58	25	19 (76.0%)	59.3%	92.7%	56.9%	95.1%
B/Washington/02/2019	Day 58	25	18 (72.0%)	54.4%	89.6%	51.9%	92.1%
B/Phuket/3073/2013	Day 58	25	19 (76.0%)	59.3%	92.7%	56.9%	95.1%

11. SAFETY RESULTS

For the analyses of the vital signs and the physical examinations, refer to section 9.4.3. None of the detected clinically significant abnormalities became serious during the study. No other serious adverse events (SAEs) were reported during the study period.

12. OVERALL CONCLUSIONS

The vaccine was well tolerated in all study participants; no serious adverse events were reported. Significant increases in HAI antibodies were elicited between D0 and D58 after vaccination with Vaxigrip Tetra®.

13. STUDY REPORT AUTHORS

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14. ANNEXES

Annex I: Informed consent form version 4.0

Annex II: Investigators' signature page

Annex III: Summary of product characteristics

Annex IV: Summary of viral strains included in vaccines and HAI testing