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A Phase III randomised trial of the immunogenicity and safety of quadrivalent versus trivalent inactivated subunit influenza vaccine in adult and elderly subjects, assessing both anti-haemagglutinin and virus neutralisation antibody responses

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ABSTRACT

Background: Trivalent influenza vaccines (TIVs) offer substantial protection against matching B-strains, however, protection against alternate-lineage B-strains may be enhanced by adding a second B-strain in quadrivalent influenza vaccines (QIVs). In this Phase III, double-blind, multicentre, randomised study, the immunogenicity and safety of subunit inactivated QIV versus TIV was assessed in adult (aged ≥18 to ≤60 years) and elderly (aged ≥61 years) subjects by analysing a combination of haemagglutinin inhibition (HI) and virus neutralisation (VN).

Methods: Subjects (n = 1980) were recruited off season (2015/2016) from 20 centres in five European countries and randomised to receive either QIV (n = 1538), TIV with B-strain of the Victoria lineage (n = 221) or TIV with B-strain of the Yamagata lineage (n = 221). The primary aim was to demonstrate non-inferiority of QIV to TIV for immunogenicity against matched influenza strains based on post-vaccination HI titres. Secondary aims were to show superiority of QIV to TIV for immunogenicity against alternate-lineage B-strains and to characterise the immune response by reverse cumulative distribution (RCD) curves of antibody titres and derived serological parameters for HI and VN. Reactogenicity and occurrence of adverse events were assessed post-vaccination.

Results: QIV elicited a non-inferior immune response for matched strains (upper limit of 95% CI for HI geometric mean ratios [GMRs] <1.5) and a superior response for alternate-lineage B-strains (HI GMRs < 1; $p < 0.0001$) versus TIV. RCD curves demonstrated that post-vaccination HI and VN titres were higher for QIV versus TIV for both alternate-lineage B-strains. Seroconversion rates and geometric mean fold increases of the VN assay were consistent with the HI assay for all strains in QIV. Reporting rates of local and systemic reactions were similar in both vaccine groups.

Conclusions: QIV was non-inferior in immunogenicity to TIV for matched strains and superior to the alternate-lineage B-strains in TIV. Safety and tolerability profiles of QIV and TIV were comparable.

1. Introduction

Influenza is a respiratory infection caused by influenza viruses that affects all ages, which can lead to serious complications in high-risk individuals [1]. In temperate climates, seasonal influenza epidemics occur during the winter months and are associated with significant morbidity, mortality and economic burden [1–3]. Influenza A/H1N1, A/H3N2 and B viruses are currently circulating and can cause seasonal influenza outbreaks, with B viruses responsible for a median of 17% of influenza cases between 2000 and 2015 in Europe [4]. The disease burden of both A and B viruses is substantial, and B viruses have been estimated to be associated with 25% of all influenza related mortality [3,5]. Two antigenically distinct lineages of influenza B viruses (Victoria and Yamagata) co-circulate globally with levels of each lineage varying in different regions within the same influenza season [6–8]. Vaccination remains the most effective method of preventing influenza; further

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to this, the World Health Organization's Global Influenza Surveillance and Response System recommends the composition of vaccines based on the circulating influenza strains each influenza season both in the northern and southern hemisphere [1].

Trivalent influenza vaccines (TIVs), such as Influvac® (influenza vaccine, surface antigen, inactivated; Abbott Biologicals B.V., Weesp, The Netherlands), are currently widely available and contain two subtypes of influenza A (A/H1N1 and A/H3N2) and a single-type influenza B-strain (either Victoria or Yamagata lineage). TIVs have been shown to offer a high level of protection against matched-lineage influenza B-strains with proven immunogenicity and safety [9–11]. However, as TIVs contain only one B-strain, mismatch can occur between the recommended lineage for TIV and the circulating B-strain [5]. Although TIVs provide some cross-protection for alternate-lineage strains, quadrivalent influenza vaccines (QIVs) containing B-strains from both the Victoria and Yamagata lineages have been developed to prevent mismatch [12]. Recent Phase III randomised clinical trials have demonstrated superiority of QIVs versus TIVs for immunogenicity of alternate-lineage B-strains and non-inferiority of QIVs versus TIVs for immunogenicity of shared-lineage vaccine strains [13–17].

Traditionally, the haemagglutinin inhibition (HI) assay has been the most important serological method used to investigate the immunogenicity of influenza vaccines [18,19]. It measures the antibodies against the haemagglutination antigen and correlates with the ability of antibodies to inhibit virus infection of host cells [19,20]. A serum HI titre of ≥40 is associated with a 50% reduction in susceptibility to influenza. However, as the HI assay detects antibodies solely against the influenza haemagglutinin protein, testing additional serological antigens or serological parameters may better assess vaccine effectiveness [20,21]. Consequently, the European Medicines Agency (EMA) now recommends the use of the virus neutralisation (VN) assay, which measures the levels of functional systemic antibodies to inhibit the cytopathic effects of the virus [20,22]. This assay provides valuable additional information, because it is more sensitive for some influenza strains, and detects additional virus antigens when compared with the HI assay [20]. To our knowledge, our study is the first to date to report VN as supplementary data to HI non-inferiority or superiority analyses in a Phase III clinical trial of QIV versus TIV in accordance with the new EMA guidelines on influenza vaccines [13–17].

In this Phase III, double-blind, multicentre, randomised study, the immunogenicity and safety of Abbott's candidate subunit inactivated QIV versus Influvac®, Abbott's subunit inactivated TIV, was assessed in adult (≥18 to ≤60 years of age) and elderly (≥61 years of age) subjects. The primary aim was to demonstrate the non-inferiority of QIV to TIV for shared-lineage influenza strains based on the post-vaccination HI-titres. Secondary aims were to show the superiority of QIV to TIV for alternate-lineage influenza B-strains and to characterise the immune responses in detail by means of reverse cumulative distribution (RCD) curves of antibody titres and derived serological parameters for HI and VN. The safety profile of QIV compared to that of TIV was assessed by analysing reactogenicity as well as the occurrence of unsolicited adverse events (AEs).

2. Materials and methods

2.1. Study design

This was a Phase III, randomised, double-blind, active-controlled, three-arm, multicentre study (EudraCT number: 2014-001042-24). Eligible subjects were randomly assigned to vaccination with QIV, TIV with B-strain of the Victoria lineage ($TIV_{(Vic)}$) or TIV with B-strain of the Yamagata lineage ($TIV_{(Yam)}$). Immunogenicity and safety were assessed at Day 22 (21 days post-vaccination) with a long-term safety follow-up period of 6 months. Written approval of the study was obtained from the relevant Independent Ethics Committee/Institutional Review Board. The study was conducted in compliance with Good Clinical Practice and all applicable laws and guidelines consistent with ethical principles of the Declaration of Helsinki [23].

2.2. Study subjects

Study subjects were adults (≥18 to ≤60 years of age) and elderly (≥61 years of age) stratified 1:1. The study included 20 centres in five European countries (Belgium, Germany, Hungary, Latvia and Lithuania) and consisted of two visits and two phone contacts per subject between May 2015 and January 2016. Male and female subjects were included if they could give informed consent, were ≥18 years of age on the day of vaccination and were in stable health. Exclusion criteria included: allergy to vaccine components; history of Guillain Barré syndrome; treatment with any vaccine within 4 weeks prior to the study vaccination or influenza vaccine within the 6 months preceding enrolment; immunocompromisation; a history of known drug or alcohol abuse or any other characteristic that, in the investigator's opinion, prohibited the inclusion of the subject into the study. Any medication that influenced the immune system was not permitted 4 weeks prior to the start of, or during, the study until the Day 22 assessment.

2.3. Randomisation and blinding

Subjects were randomly assigned to the three vaccine groups through an interactive web response system (randomisation scheme provided by Abbott Biologicals B.V.); randomisation was by country and age group. Vaccines were supplied as pre-filled syringes, and to achieve double-blindness, all syringes were identical in appearance. All study investigators and subjects remained blinded throughout the duration of the study; for emergency unblinding, an interactive web response system could be used.

2.4. Vaccines and vaccination schedule

Each subject received one 0.5 ml dose containing 15 μg of haemagglutinin for each virus strain by intramuscular injection (deltoid) using a 25 mm needle (QIV, 1095939-G54A; $TIV_{(Vic)}$, 1095937-G52; $TIV_{(Yam)}$, 1095938-G53). All study vaccines were manufactured by Abbott Biologicals B.V. Blood samples were taken at Day 1 (before vaccination) and at Day 22 (21 days post-vaccination).

2.5. Immunogenicity endpoints

The immunogenicity endpoints were Day 22 post-vaccination HI antibody titres and Day 22 post-vaccination VN antibody titres against the four virus strains.

2.5.1. Primary immunogenicity analysis

The non-inferiority of QIV to TIV with respect to induced immunogenicity against the shared strains was tested by comparing the Day 22 HI geometric mean titres (GMTs) of the shared strains between the QIV and TIV formulations in subjects ≥18 years of age (per-protocol sample [PPS]).

2.5.2. Secondary immunogenicity analysis

The superiority of QIV to TIV with respect to induced immunogenicity against the alternate-lineage B-strains was tested by comparing the Day 22 HI GMTs of the alternate-lineage B-strains between QIV and TIV formulations in subjects ≥18 years of age.

Immune responses were further characterised in both age cohorts by analysing pre- and post-vaccination HI and VN GMTs, seroprotection rates (SPRs), seroconversion rates (SCRs) and geometric mean fold increases (GMFIs) in HI and VN titres. SPRs were defined as the proportion of subjects achieving an HI titre ≥40 (appropriate VN titres to derive SPRs are currently unknown). SCRs for HI and VN were both defined as becoming seropositive if seronegative at enrolment, or a ≥4-fold rise in titre if seropositive at enrolment.

2.5.3. Safety endpoints

The safety endpoints were the frequency and severity of solicited local and systemic reactions reported as per home diary for a period of 7 days after vaccination. In addition, a general question at the end of the 7-day reporting period was asked regarding overall inconvenience after the vaccination. Unsolicited AEs (both non-serious and serious) were recorded up to the Day 22 visit, and only serious unsolicited AEs (SAEs) and new chronic illnesses (NCIs) were recorded between Day 22 and Day 183 (6 months).

2.5.4. Sample size determination and statistical analyses

The sample size was confirmed by scientific advice obtained from an EU National Competent Authority to collect safety data for approximately 1500 subjects vaccinated with QIV. Given this, a sample size of 1540 subjects vaccinated with QIV and two times 220 subjects vaccinated with TIV secured an overall statistical power of >95% to demonstrate the non inferiority of QIV to TIV with respect to the immunogenicity against the shared strains.

Log-transformed HI titres were compared between the groups by means of analysis of variance (ANOVA), with country and age group as covariates. Non-inferiority of QIV to TIV could be concluded if all four two-sided 95% confidence intervals (CIs) for the geometric mean ratios (GMRs) would fall below the predefined non-inferiority margin of 1.5. The non inferiority margin of 1.5 was set in accordance with current regulatory guidance [24]. The PPS was used for the primary non-inferiority analysis, in accordance with the International Conference on Harmonisation E9 guideline Statistical Principles for Clinical Trials [25].

Superiority of QIV over TIV (with regard to inducing higher immune responses) was investigated using the ANOVA model described above. Superiority could be concluded if the two-sided 95% CI fell below 1. Derived serological parameters and RCD curves were summarised for each of the four strains in each of the three formulations and each age cohort. The RCD plot is a graphical tool to display the distribution of immunogenicity values and is useful for visual comparisons of distributions between vaccine groups [26]. The x-axis represents the immunogenicity values, the y-axis the proportion of subjects having at least that immunogenicity level. The curve begins at 100% and then descends from left to right. If the RCD curve for one vaccine is above the curve for another vaccine, then the vaccine with the highest curve induced the greatest immune responses. If two RCD curves coincide it demonstrates comparable immune responses.

3. Results

3.1. Demographics and baseline characteristics

A total number of 1980 subjects were randomised to receive QIV (n = 1538), $TIV_{(Vic)}$ (n = 221) or $TIV_{(Yam)}$ (n = 221) (Fig. 1). Overall, 11 subjects prematurely withdrew from the study; the reasons were: withdrawal of consent (n = 4), AEs (n = 3), lost to follow-up (n = 3), or administrative (n = 1). Subjects were predominantly white (99.5%) and mean age at screening was 55.7 years (SD ± 17.7 years) (Table 1). The proportion of male to female subjects was 43.4–56.6%.

3.2. Immunogenicity

3.2.1. Primary efficacy

For all four influenza strains in the PPS, the upper limit of the 95% CI for the adjusted HI GMR fell below the non-inferiority margin of 1.5, demonstrating non-inferiority of QIV versus TIV for shared strains (Fig. 2a). The non-inferiority analysis was repeated for the full analysis sample and the results were comparable (data not shown).

3.2.2. Secondary efficacy

For both B-strain influenza lineages, the post-vaccination HI GMT of the TIV group with the alternate B-strain lineage was less than half that of the QIV group. The HI GMTs in the QIV group for B/Victoria and B/Yamagata lineages were 153.1 and 101.9 versus 64.1 and 47.2 for the alternate lineages in the TIV groups, which translated to an adjusted HI GMR of 0.41 and 0.45, respectively (Fig. 2b, $p < 0.0001$ for both comparisons). The HI antibody responses elicited by the B-strain antigens were, therefore, superior to the antibody responses elicited by cross-reactivity antigens of the alternate-lineage B-strains.

In the QIV group, SPRs were ≥91.6% in adult subjects and ≥73.3% in elderly subjects for all four influenza strains (Supplemental Material Table 1). SCRs based on HI titres were ≥51.3% in adult subjects and ≥39.3% in elderly subjects for all four influenza strains. SCRs based on VN titres were in line with the HI titres, with values of ≥42.5% in adult subjects and ≥34.1% in elderly subjects for all four strains. GMFIs in HI titres varied between 6.3 and 11.4 in adult subjects versus 4.2 and 5.5 in elderly subjects. Across all subjects, GMFIs in VN titres were in line with the HI titres, although differences were less pronounced: between 3.1 and 5.4 in adult subjects versus 2.4 and 3.4 in elderly subjects. HI and VN GMTs pre- and post-vaccination in adult and elderly subjects are shown in Supplemental Material Table 2.

In all vaccine groups pre vaccination, the HI and VN RCD curves were similar for each of the tested influenza strains, indicating a lack of baseline serological bias between study groups (data not shown). Post-vaccination, the proportion of subjects reaching higher HI and VN titres increased. While the post-vaccination RCD curves were similar for the shared-lineage influenza strains, for the alternate-lineage B-strains, there was a shift in the TIV RCD curves demonstrating that a lower proportion of subjects achieved HI titres ≥10.0 and VN titres ≥28.3 versus QIV (Fig. 3).

3.3. Safety and tolerability

Local and systemic reaction profiles were comparable between QIV and TIV 7 days post vaccination across all subjects; most reactions were mild or moderate in severity and lasted for 1–3 days for the majority of subjects. In adult subjects, reporting rates for systemic reactions were generally low (<10%), except for headache and fatigue (Fig. 4A). Headache was the most common systemic reaction reported by 12.4% and 13.1% of adult subjects in the QIV and TIV groups, respectively (Fig. 4A). Reporting rates of local reactions were also generally low (<10%), except for vaccination-site pain, which was reported by 24.9% and 18.5% of adult subjects in the QIV and TIV groups, respectively (Fig. 4A). Reporting rates of local and systemic reactions in elderly subjects followed a similar profile to that of adult subjects, although overall reporting rates were lower. The only local or systemic reaction occurring in >10% of elderly subjects in either vaccine group was fatigue, which was reported by 10.6% and 6.8% of subjects in the QIV and TIV groups, respectively (Fig. 4B). The vast majority of subjects did not experience any overall inconvenience during the 7 days after vaccination and the frequency of overall inconvenience was similar between the QIV and TIV groups (data not shown).

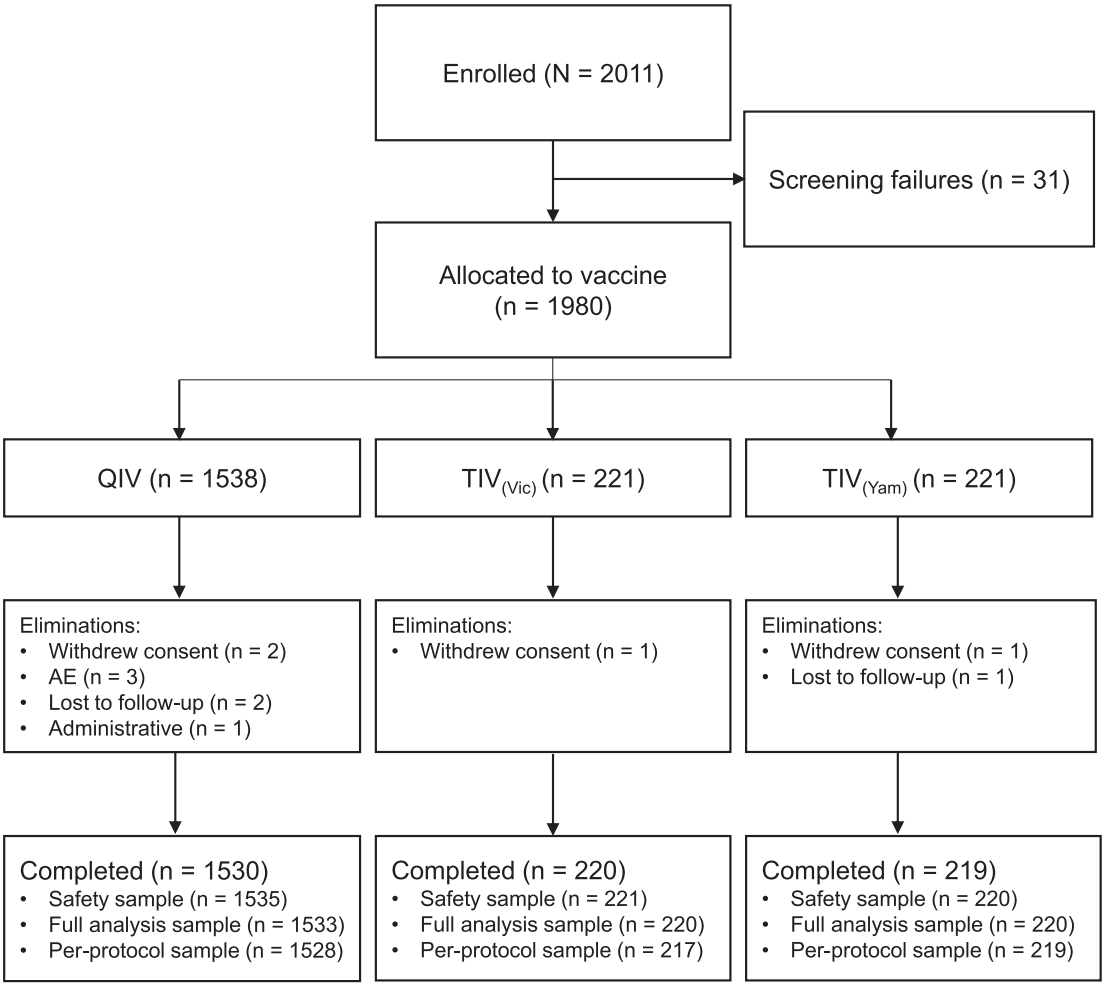


Fig. 1. Subject disposition (CONSORT flow diagram). AE, adverse event; QIV, quadrivalent influenza vaccine; TIV, trivalent influenza vaccine; Vic, Victoria lineage; Yam, Yamagata lineage.

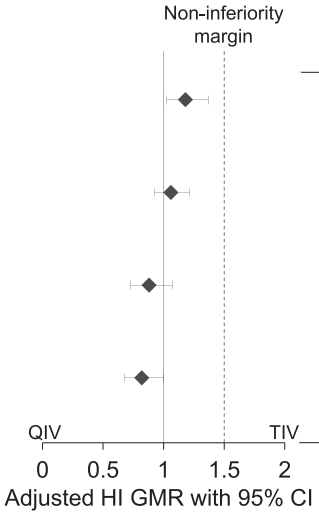
Table 1
Subject demographics at baseline.

	Statistic	QIV (N = 1538)	$TIV_{(Vic)}$ (N = 221)	$TIV_{(Yam)}$ (N = 221)	All subjects (N = 1980)
Age (years)	Mean (SD)	55.9 (17.6)	55.4 (18.0)	55.0 (17.6)	55.7 (17.7)
	Median	61.0	61.0	60.0	61.0
	Min/Max	18/91	18/88	18/86	18/91
Age group					
Adult subjects (≥18 to ≤60 years)	n (%)	769 (50.0)	110 (49.8)	112 (50.7)	991 (50.1)
Elderly subjects (≥61 years)	n (%)	769 (50.0)	111 (50.2)	109 (49.3)	989 (49.9)
Gender					
Male	n (%)	664 (43.2)	100 (45.2)	95 (43.0)	859 (43.4)
Female	n (%)	874 (56.8)	121 (54.8)	126 (57.0)	1121 (56.6)
Race					
White	n (%)	1529 (99.4)	221 (100.0)	221 (100.0)	1971 (99.5)
Asian	n (%)	3 (0.2)	0	0	3 (0.2)
Black or African American	n (%)	3 (0.2)	0	0	3 (0.2)
Other	n (%)	3 (0.2)	0	0	3 (0.2)

Max, maximum; Min, minimum; QIV, quadrivalent influenza vaccine; SD, standard deviation; TIV, trivalent influenza vaccine; Vic, Victoria lineage; Yam, Yamagata lineage.

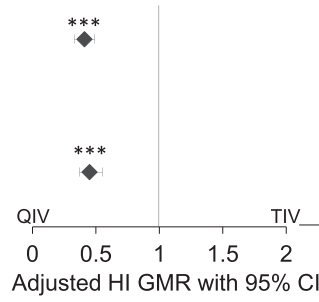
The frequency of unsolicited AEs was low and comparable between the QIV and TIV groups; the vast majority of subjects did not report any AEs up to the Day 22 visit (Table 2). The frequency of adult subjects reporting at least one AE was 4.8% and 3.6% in the QIV and TIV groups, respectively. Similarly, the frequency of elderly subjects reporting at least one AE was 3.8% and 2.7% in the respective QIV and TIV groups (Table 2). The most frequent AEs were in the category of infections and infestations across all subjects (data not shown). The proportion of subjects experiencing vaccine-related AEs was low (<1%) for both vaccine groups across all subjects. Both QIV and TIV had favourable long-term safety profiles (Day 22 to Day 183), with the incidence of both SAEs

A



HI analysis strain	GMT (n)		Adjusted HI GMR (95% CI)
	TIV	QIV	TIV/QIV
A/H1N1	220.9 (433)[a]	186.6 (1511)	1.18 (1.02–1.37)
A/H3N2	413.5 (436)[a]	393.1 (1524)	1.06 (0.93–1.21)
B/Victoria	142.0 (215)	152.9 (1521)	0.88 (0.73–1.07)
B/Yamagata	86.1 (215)	102.1 (1520)	0.82 (0.68–1.00)

B



HI analysis strain	GMT (n)		Adjusted HI GMR (95% CI)
	$TIV_{(Yam)}$	QIV	$TIV_{(Yam)}$/QIV
B/Victoria	64.1 (218)	153.1 (1526)	0.41 (0.33–0.49)
	$TIV_{(Vic)}$	QIV	$TIV_{(Vic)}$/QIV
B/Yamagata	47.2 (220)	101.9 (1525)	0.45 (0.37–0.55)

Fig. 2. (A) Non-inferiority analysis of QIV versus TIV for the immunogenicity of shared influenza strains in subjects ≥18 years of age (per-protocol sample). Adjusted HI GMR and 95% CI were calculated using ANOVA on the log-transformed titres at the Day 22 visit. The non-inferiority margin of 1.5 (dashed line) was pre-determined in accordance with current regulatory guidance [24]. (B) Superiority analysis of QIV versus TIV for the immunogenicity of alternate-lineage influenza B-strains in subjects ≥18 years of age (full analysis sample). Superiority was analysed using an ANOVA model for the log-transformed titres and superiority of QIV was demonstrated if the two-sided 95% CI for the adjusted GMR fell below 1 (solid grey line). *** $p < 0.0001$ (two-sided ANOVA). [a]HI data for the two A-strains in TIV were pooled. ANOVA, analysis of variance; CI, confidence interval; FDA, Food and Drug Administration; GMT, geometric mean titre; GMR, geometric mean ratio; HI, haemagglutinin inhibition; QIV, quadrivalent influenza vaccine; TIV, trivalent influenza vaccine; Vic, Victoria lineage; Yam, Yamagata lineage.

and NCIs low in adult (<2%) and elderly (<5%) subjects, but none with a reasonable possibility for a causal relationship with the study vaccine. Overall, the safety profile of QIV was similar to that of TIV and both vaccine types were generally well tolerated.

4. Discussion

In this Phase III, randomised, double-blind clinical trial in adult (≥18 to ≤60 years of age) and elderly (≥61 years of age) subjects, the immune response, as defined by post-vaccination HI titres to QIV, was non-inferior to that of TIV for shared influenza strains. These data demonstrate that the presence of a second influenza B-strain in QIV does not negatively affect immune response to the other strains. Moreover, the immune response to QIV was superior to cross-reactive titres against the alternate-strain lineage in TIV, which is consistent with other Phase III trials comparing inactivated QIVs versus TIVs [13–17]. Collectively, these data indicate that the addition of a second B-strain in QIV may enhance the protective efficacy of influenza vaccines on occasions where mismatch would occur between the recommended B-strain for TIV and the one predominantly circulating. As influenza B is associated with substantial mortality and economic burden, the use of QIVs over TIVs may further reduce the burden of influenza [3,5]. In addition, modelling studies suggest that QIVs could be more cost-effective than TIVs in protecting children and the elderly against influenza, further suggesting a positive impact on healthcare for QIVs [27].

In this study, the HI data showed a strong serological response for each of the shared influenza strains in the QIV and TIV groups, and the percentage of subjects in the QIV group achieving a serum HI titre ≥ 40 (SPR) was high both for adult and elderly subjects. Importantly, the HI immunogenicity data were supplemented with

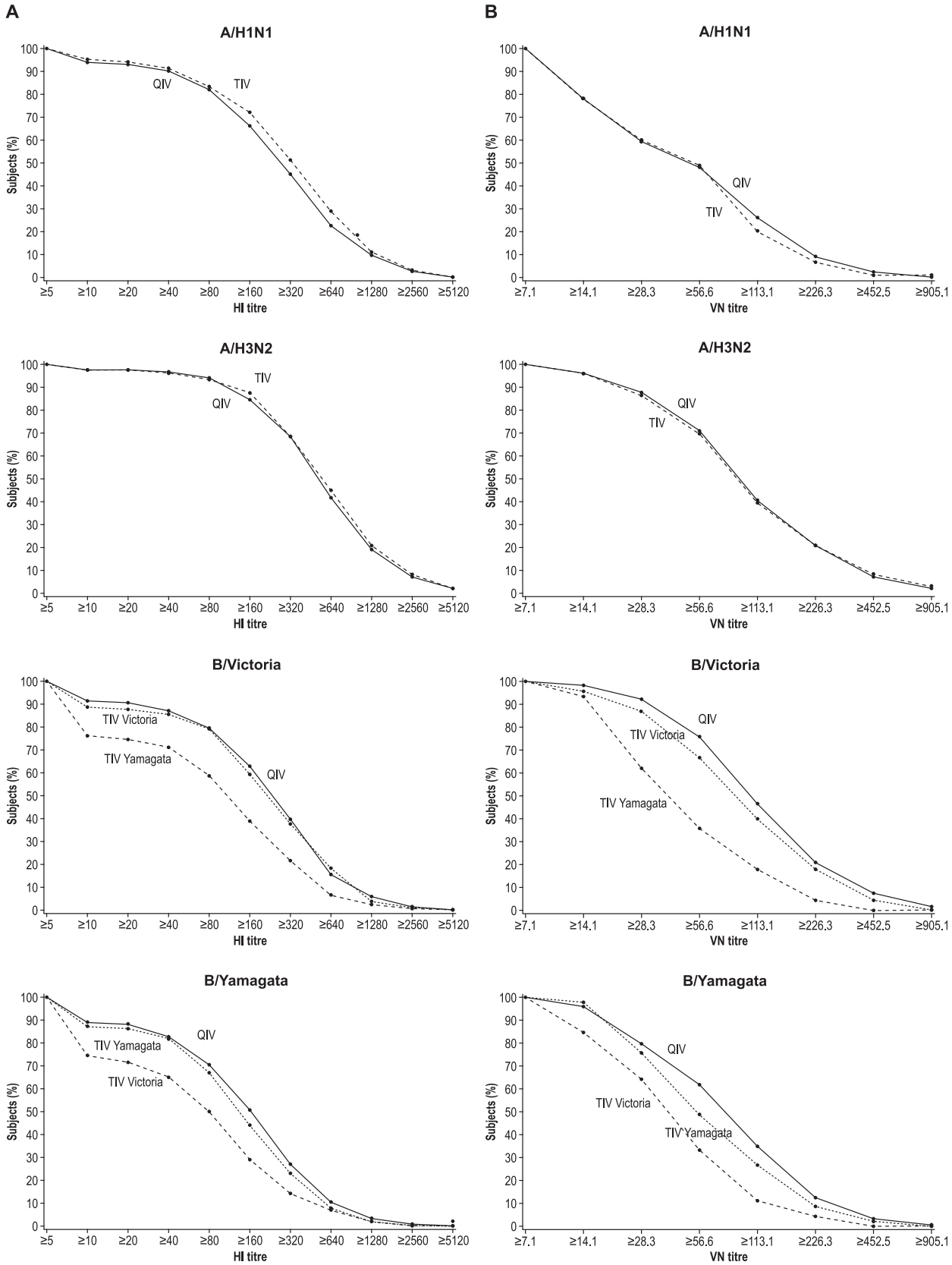


Fig. 3. Reverse cumulative distribution curves of HI titres (A) and VN titres (B) in subjects ≥18 years of age pre- and post-vaccination with QIV or TIV for each of the four strains. When the QIV and the TIV curves overlap, the two vaccines induced comparable immune responses. If the curve for QIV is above the curve for TIV, QIV induced the higher immune response. HI, haemagglutinin inhibition; QIV, quadrivalent influenza vaccine; TIV, trivalent influenza vaccine; VN, virus neutralisation.

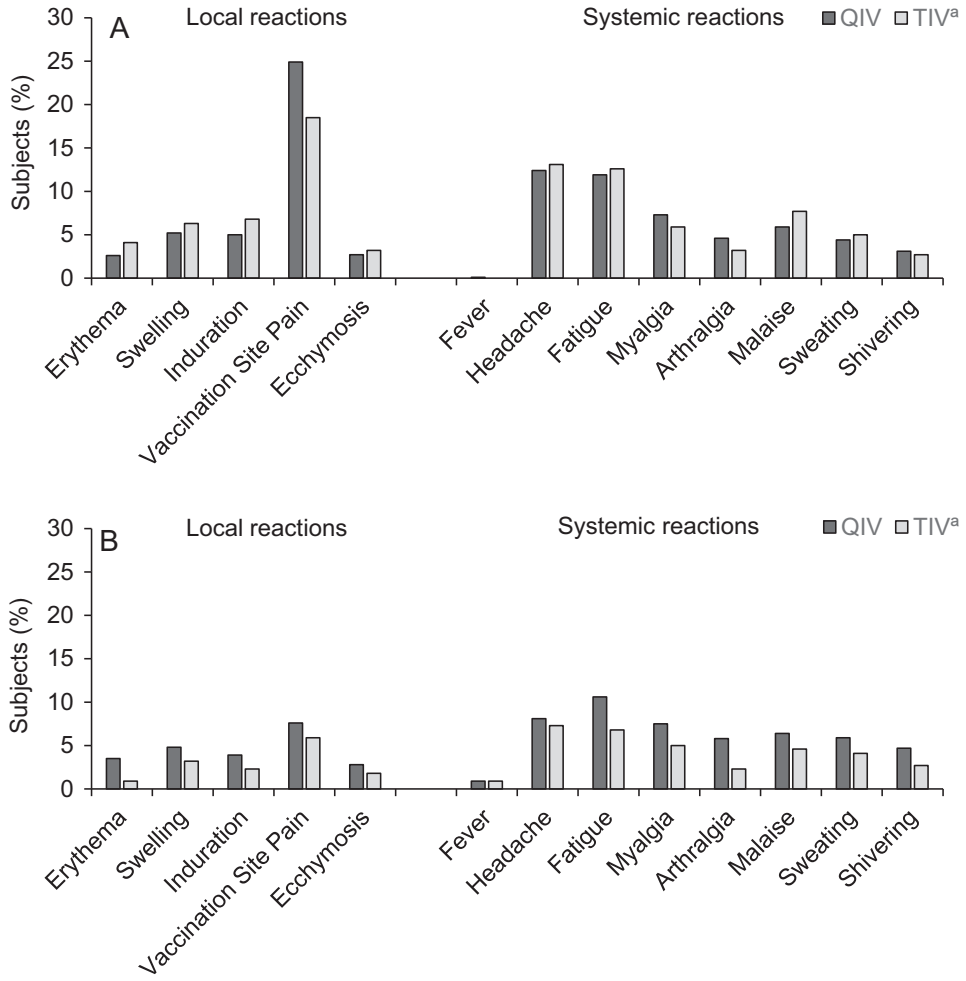


Fig. 4. Local and systemic reactions 7 days after vaccination in (A) adult (≥18 to ≤60 years of age) and (B) elderly (≥61 years of age) subjects (safety sample). Data are presented as percentage of subjects experiencing a vaccination reaction. [a]The safety data of the subjects vaccinated with TIV formulations were pooled. QIV, quadrivalent influenza vaccine; TIV, trivalent influenza vaccine.

Table 2
Subjects with ≥1 AE during the immunisation phase (Days 1–22 post-vaccination) and the long-term safety follow-up (Days 22–183).

	Adult subjects (≥18 to ≤60 years)		Elderly subjects (≥61 years)	
	QIV (N = 768)	TIV[a] (N = 222)	QIV (N = 767)	TIV[a] (N = 219)
Immunisation phase				
Unsolicited AE, n (%)	37 (4.8)	8 (3.6)	29 (3.8)	6 (2.7)
SAE, n (%)	2 (0.3)	0	4 (0.5)	1 (0.5)
AE with possibility for a causal relationship, n (%)	4 (0.5)	2 (0.9)	6 (0.8)	2 (0.9)
AE leading to study termination, n (%)	0	0	0	0
Death, n (%)	0	0	0	0
Long-term safety follow-up				
SAE, n (%)	10 (1.3)	4 (1.8)	30 (3.9)	9 (4.1)
NCI, n (%)	10 (1.3)	3 (1.4)	31 (4.0)	5 (2.3)
SAE/NCI with possibility for a causal relationship, n (%)	0	0	0	0
AE leading to study termination, n (%)	1 (0.1)	0	2 (0.3)	0
Death, n (%)	1 (0.1)	0	4 (0.5)	0

AE, adverse event; NCI, new chronic illness; QIV, quadrivalent influenza vaccine; SAE, serious adverse event; TIV, trivalent influenza vaccine.
[a] Safety data of the subjects vaccinated with TIV were pooled.

VN assay analyses, a combination that has not been reported in other recent Phase III clinical trials of QIV versus TIV [13–17]. This study demonstrated that the GMFIs and SCRs of the VN assay were consistent with those of the HI assay, indicating the effective immunogenicity of QIV and validating the use of VN testing for future investigation into influenza vaccines. These data were consistent with the results of a smaller Phase I/II single-centre, single-blind study investigating QIV versus TIV [28] and a Phase

III trial that was not powered to assess non inferiority or superiority of QIV responses versus TIV from HI analyses [29].

HI and VN assays have different strengths and limitations. Whilst an HI titre of ≤40 is currently the only universal indicator of a 50% reduction in the risk of influenza disease, HI titres only correlate with the ability of antibodies to inhibit virus infection of host cells [19,20]. In contrast, the VN assay measures the antibody titres needed to inhibit the cytopathic effects of the virus and more closely mirrors the physiology of the influenza disease process compared with the HI assay [20]. The VN assay is generally thought to be more sensitive than the HI assay, but it is also more expensive and time-consuming, and harder to standardise across different laboratories [20]. Because of these strengths and limitations, assessing both anti-HI and VN antibody responses may enhance the overall accuracy of vaccine testing.

The recent EMA guidelines also recommend the reporting of RCD curves, to provide valuable insight into the distribution of antibody titres between study populations and vaccination groups pre- and post-vaccination [22]. Importantly, the RCD data demonstrated that pre vaccination titres were similarly distributed for each of the tested influenza strains for all vaccine groups, indicating an absence of serological imbalances pre-vaccination with QIV or TIV. For both alternate-lineage B-strains, post-vaccination HI and VN titres were higher for QIV versus TIV, further validating the addition of the extra B-strain in QIVs.

Overall, the results of this study demonstrate that the additional B-strain in QIV did not compromise safety compared to TIV. Reporting rates of unsolicited AEs were low and similar between the vaccine groups up to Day 22 and none of the reported SAEs or NCIs during the long-term follow-up period had a reasonable possibility for a causal relationship with the study vaccine. Local and systemic reactogenicity profiles were also similar between vaccine groups; the majority of reactions were mild or moderate in severity and lasted for 1–3 days. These data are consistent with the results of a meta-analysis of five randomised clinical trials, demonstrating that there was no significant difference between QIV and TIV in terms of the frequency of aggregated local and systemic AEs within 7 days post-vaccination [30].

Influvac® subunit TIV has been used for over 30 years with a well-established immunogenicity and safety profile [9]. In this study, we demonstrated that the immunogenicity of the influenza strains in QIV was non-inferior to the shared strains and superior to the alternate-lineage B-strains in TIV. Furthermore, the safety profiles of the two vaccine groups were comparable. Collectively, these data support the use of Influvac® QIV for seasonal vaccination in adult and elderly subjects, which may enhance the protection against influenza and decrease the burden associated with influenza complications.

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Role of the sponsor

Abbott Biologicals B.V. was involved in the study design, data collection, data analysis and preparation of the manuscript.

Role of the contributors

All authors attest they meet the ICMJE criteria for authorship. All authors participated in the design, implementation, analysis and/or interpretation of the study. All authors were involved in the drafting of the article or revising it critically for important intellectual content and final approval of the manuscript.

Conflict of interest

S. van de Witte and J. Nauta are employees of Abbott Healthcare Products B.V. J. Weckx was coordinating investigator and received grant support from Abbott Biologicals B.V. to conduct this study. No other conflict of interest was declared for the work presented in this article.

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Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.vaccine.2018.04.043.

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