Cross-protective efficacy of HPV-16/18 AS04-adjuvanted vaccine against cervical infection and precancer caused by non-vaccine oncogenic HPV types: 4-year end-of-study analysis of the randomised, double-blind PATRICIA trial

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Summary

Background We evaluated the efficacy of the human papillomavirus HPV-16/18 AS04-adjuvanted vaccine against non-vaccine oncogenic HPV types in the end-of-study analysis after 4 years of follow-up in PATRICIA (PApilloma TRIal against Cancer In young Adults).

Methods Healthy women aged 15–25 years with no more than six lifetime sexual partners were included in PATRICIA irrespective of their baseline HPV DNA status, HPV-16 or HPV-18 serostatus, or cytology. Women were randomly assigned (1:1) to HPV-16/18 vaccine or a control hepatitis A vaccine, via an internet-based central randomisation system using a minimisation algorithm to account for age ranges and study sites. The study was double-blind. The primary endpoint of PATRICIA has been reported previously; the present analysis evaluates cross-protective vaccine efficacy against non-vaccine oncogenic HPV types in the end-of-study analysis. Analyses were done for three cohorts: the according-to-protocol cohort for efficacy (ATP-E; vaccine n=8067, control n=8047), total vaccinated HPV-naive cohort (TVC-naive; no evidence of infection with 14 oncogenic HPV types at baseline, approximating young adolescents before sexual debut; vaccine n=5824, control n=5820), and the total vaccinated cohort (TVC; all women who received at least one vaccine dose, approximating catch-up populations that include sexually active women; vaccine n=9319, control=9325). Vaccine efficacy was evaluated against 6-month persistent infection, cervical intraepithelial neoplasia grade 2 or greater (CIN2+) associated with 12 non-vaccine HPV types (individually or as composite endpoints), and CIN3+ associated with the composite of 12 non-vaccine HPV types. This study is registered with ClinicalTrials.gov, number NCT00122681.

Findings Consistent vaccine efficacy against persistent infection and CIN2+ (with or without HPV-16/18 co-infection) was seen across cohorts for HPV-33, HPV-31, HPV-45, and HPV-51. In the most conservative analysis of vaccine efficacy against CIN2+, where all cases co-infected with HPV-16/18 were removed, vaccine efficacy was noted for HPV-33 in all cohorts, and for HPV-31 in the ATP-E and TVC-naive. Vaccine efficacy against CIN2+ associated with the composite of 12 non-vaccine HPV types (31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68), with or without HPV-16/18 co-infection, was 46·8% (95% CI 30·7–59·4) in the ATP-E, 56·2% (37·2–69·9) in the TVC-naive, and 34·2% (20·4–45·8) in the TVC. Corresponding values for CIN3+ were 73·8% (48·3–87·9), 91·4% (65·0–99·0), and 47·5% (22·8–64·8).

Interpretation Data from the end-of-study analysis of PATRICIA show cross-protective efficacy of the HPV-16/18 vaccine against four oncogenic non-vaccine HPV types—HPV-33, HPV-31, HPV-45, and HPV-51—in different trial cohorts representing diverse groups of women.

Funding GlaxoSmithKline Biologicals.

Introduction

Infection with oncogenic human papillomavirus (HPV) types is a necessary cause of invasive cervical cancer (ICC).[1,2] Roughly 15 HPV types have been classified as oncogenic. Among these, HPV-16 and HPV-18 are the most prevalent, and cause around 70% of ICC worldwide.[3] The next most prevalent oncogenic type is HPV-45.[3] HPV-16 (A9 species) together with HPV-18 and HPV-45 (A7 species) cause 75% of squamous-cell carcinoma (SCC) and 94% of adenocarcinoma.[3] The next five most common oncogenic HPV types are all from the A9 species (HPV-31, HPV-33, HPV-35, HPV-52, and HPV-58) and together cause another 15% of ICC.[3] The remaining oncogenic HPV types individually cause a very small proportion of ICC worldwide (<2%) and include HPV-51 (A5 species), HPV-56 (A6 species), and HPV-39 and HPV-59 (A7 species).[3–5] The possible carcinogenicity of HPV-66 (A6 species) is uncertain, whereas HPV-68 (A7 species) is probably oncogenic.[4]

Published Online November 9, 2011 DOI:10.1016/S1470-2045(11)70287-X

See Online/Comment DOI:10.1016/S1470-2045(11)70324-2

See Online/Articles DOI:10.1016/S1470-2045(11)70286-8

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Prophylactic HPV vaccines are administered in vaccination programmes targeted at young adolescent girls before sexual exposure, and in catch-up programmes for young women in some countries. Since non-vaccine HPV types account for around 30% of cervical cancers, cross-protection against these types would potentially enhance primary cervical cancer prevention efforts.

The HPV-16/18 AS04-adjuvanted vaccine (Cervarix, GlaxoSmithKline Biologicals) and HPV-6/11/16/18 vaccine (Gardasil, Merck) consist of virus-like particles (VLPs) composed of relatively well conserved L1 capsid proteins. The neutralising antigenic sites (epitopes) defined so far are mainly situated on one of five variable loops of the L1 capsomer. These are exposed on virion surfaces and should be readily accessible to neutralising antibodies.[6–8] In theory, aminoacid sequence or conformational differences determine the type-specificity of any HPV-neutralising epitope. Some oncogenic HPV types that are phylogenetically related to vaccine types presumably share epitopes that can elicit cross-reactive immune responses, although cross-neutralising antibodies might be induced by HPV vaccination at much lower levels than type-specific antibodies.[9]

This report summarises cross-protection data with the HPV-16/18 vaccine in the end-of-study analysis of the PApilloma TRIal against Cancer In young Adults (PATRICIA). In general, cervical intraepithelial neoplasia grade 2 or greater (CIN2+) is the accepted clinical endpoint to evaluate HPV vaccine efficacy. However, analyses can be biased if a lesion is co-infected with both a vaccine and a non-vaccine oncogenic HPV type, since definitive causality to a single HPV type cannot be readily assigned.[10–12] This confounding bias particularly applies to analyses of cross-protection, because HPV-16 and HPV-18 are common co-infections and are more prevalent than other HPV types in cervical lesions. Additionally, as a

	Vaccine			Control			Efficacy (95% CI)
	N	Cases	Rate	N	Cases	Rate	
6-month persistent infection							
Non-vaccine A9 species (composite HPV-31/33/35/52/58)	7671	608	2·49	7656	770	3·19	22·0% (13·2 to 30·0)
HPV-31	7400	58	0·24	7414	247	1·01	76·8% (69·0 to 82·9)
HPV-33	7534	65	0·26	7513	117	0·47	44·8% (24·6 to 59·9)
HPV-35	7579	67	0·27	7569	56	0·22	–19·8% (–74·1 to 17·2)
HPV-52	7289	346	1·46	7237	374	1·59	8·3% (–6·5 to 21·0)
HPV-58	7518	144	0·58	7511	122	0·49	–18·3% (–51·8 to 7·7)
Non-vaccine A7 species (composite HPV-39/45/59/68)	7672	419	1·69	7656	472	1·91	11·6% (–1·0 to 22·7)
HPV-39	7429	175	0·71	7428	184	0·75	4·8% (–17·7 to 23·1)
HPV-45	7594	24	0·09	7556	90	0·36	73·6% (58·1 to 83·9)
HPV-59	7536	73	0·29	7530	68	0·27	–7·5% (–51·8 to 23·8)
HPV-68	7450	165	0·67	7424	169	0·69	2·6% (–21·5 to 21·9)
Other							
HPV-51	7190	349	1·50	7165	416	1·79	16·6% (3·6 to 27·9)
HPV-56	7467	226	0·92	7451	215	0·88	–5·3% (–27·5 to 13·1)
HPV-66	7412	211	0·87	7375	215	0·89	2·3% (–18·7 to 19·6)
CIN2+ with or without co-infection with HPV-16/18							
Non-vaccine A9 species (composite HPV-31/33/35/52/58)	7854	55	0·21	7846	108	0·42	49·1% (29·0 to 63·9)
HPV-31	7575	5	0·02	7592	40	0·16	87·5% (68·3 to 96·1)
HPV-33	7712	13	0·05	7700	41	0·16	68·3% (39·7 to 84·4)
HPV-35	7760	3	0·01	7757	8	0·03	62·5% (–56·5 to 93·6)
HPV-52	7455	24	0·10	7409	33	0·14	27·6% (–26·3 to 59·1)
HPV-58	7701	15	0·06	7696	21	0·08	28·5% (–45·5 to 65·7)
Non-vaccine A7 species (composite HPV-39/45/59/68)	7855	18	0·07	7846	43	0·17	58·2% (25·9 to 77·3)
HPV-39	7602	4	0·02	7608	16	0·06	74·9% (22·3 to 93·9)
HPV-45	7774	2	0·01	7738	11	0·04	81·9% (17·0 to 98·1)
HPV-59	7713	1	0·00	7716	5	0·02	80·0% (–79·1 to 99·6)
HPV-68	7626	11	0·04	7606	15	0·06	26·8% (–70·7 to 69·6)
Other							
HPV-51	7356	21	0·09	7341	46	0·19	54·4% (22·0 to 74·2)
HPV-56	7638	7	0·03	7631	13	0·05	46·1% (–45·2 to 81·8)
HPV-66	7583	7	0·03	7559	16	0·06	56·4% (–12·1 to 84·8)

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	Vaccine			Control			Efficacy (95% CI)
	N	Cases	Rate	N	Cases	Rate	
(Continued from previous page)							
CIN2+ excluding co-infection with HPV-16/18							
Non-vaccine A9 species (composite HPV-31/33/35/52/58)	7854	55	0·21	7846	78	0·30	29·5% (–0·9 to 51·0)
HPV-31	7575	5	0·02	7592	32	0·13	84·3% (59·5 to 95·2)
HPV-33	7712	13	0·05	7700	32	0·13	59·4% (20·5 to 80·4)
HPV-35	7760	3	0·01	7757	5	0·02	39·9% (–208·9 to 90·7)
HPV-52	7455	24	0·10	7409	19	0·08	–25·8% (–142·9 to 34·0)
HPV-58	7701	15	0·06	7696	14	0·06	–7·3% (–139·9 to 51·7)
Non-vaccine A7 species (composite HPV-39/45/59/68)	7855	16	0·06	7846	23	0·09	30·4% (–37·5 to 65·7)
HPV-39	7602	4	0·02	7608	7	0·03	42·7% (–125·4 to 87·7)
HPV-45	7774	2	0·01	7738	4	0·02	50·1% (–247·9 to 95·5)
HPV-59	7713	1	0·00	7716	3	0·01	66·6% (–316·1 to 99·4)
HPV-68	7626	9	0·04	7606	10	0·04	10·1% (–146·2 to 67·7)
Other							
HPV-51	7356	19	0·08	7341	22	0·09	13·7% (–67·1 to 55·8)
HPV-56	7638	6	0·02	7631	10	0·04	40·0% (–82·3 to 82·1)
HPV-66	7583	7	0·03	7559	11	0·04	36·5% (–79·3 to 79·1)

Women could be infected with multiple HPV types (therefore the number of cases for the composite endpoints might not equal the sum of the cases for each individual type included in the composite). Types tested for were HPV-16, HPV-18, HPV-31, HPV-33, HPV-35, HPV-39, HPV-45, HPV-51, HPV-52, HPV-56, HPV-58, HPV-59, HPV-66, and HPV-68. Women included in the analysis were HPV DNA negative for the HPV type under consideration at month 0 and month 6, irrespective of initial serostatus, and had negative or low-grade cytology at month 0. For the composite endpoints, women had to be infected with, or have a lesion associated with, at least one of the HPV types in the composite, and had to be HPV DNA negative for the corresponding HPV type at month 0 and month 6. CIN2+ was defined histologically as CIN2, CIN3, adenocarcinoma in situ, or invasive carcinoma. CIN=cervical intraepithelial neoplasia. HPV=human papillomavirus. ATP-E=according-to-protocol for efficacy. N=number of evaluable women in each group. Cases=number of evaluable women reporting at least one event. Rate=number of cases divided by sum of follow-up period (per 100 woman years); follow-up period began the day after the third vaccine dose.

Table 1: **Cross-protective efficacy against 6-month persistent infection and CIN2+ associated with non-vaccine HPV types, in women who were DNA negative for the corresponding HPV type at baseline (ATP-E cohort)**

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See **Online** for webappendix

result of vaccination, HPV-16 and HPV-18 infections are differentially removed from the vaccine and control groups. To overcome these biases, we did analyses of CIN2+ and the more stringent endpoint, CIN3+, which either include or exclude cases co-infected with a vaccine type. We also did complementary analyses using virological endpoints. Persistent HPV infection usually precedes cervical cancer and its precursor lesions (CIN grade 2 and 3),[13–16] and therefore provides a relevant marker for the risk of developing these precancerous lesions.

To estimate the extent of cross-protection, we did the analyses in various cohorts: the according-to-protocol cohort for efficacy (ATP-E), the total vaccinated HPV-naive cohort (TVC-naive), and the total vaccinated cohort (TVC). The ATP-E cohort represents a population of women who at baseline had no evidence of infection with the HPV type under analysis and who received all three vaccine doses. In terms of exposure to and acquisition of HPV types, the TVC-naive approximates the current primary target of HPV vaccination programmes. The TVC includes women with evidence of current or previous infection with oncogenic HPV types, and approximates a population currently targeted by catch-up HPV vaccination programmes. Data regarding other measures of efficacy in the TVC-naive and TVC are reported in an accompanying article by Lehtinen and colleagues.[17]

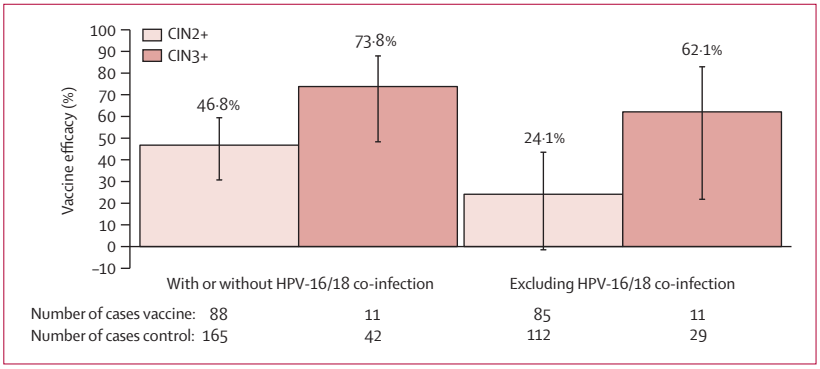

Figure 1: **Vaccine efficacy against CIN2+ and CIN3+ associated with a composite of 12 non-vaccine HPV types, with or without HPV-16/18 co-infection and excluding HPV-16/18 co-infection, in the ATP-E cohort**
Women had to have a lesion associated with at least one of the HPV types included in the composite. In the analysis of the ATP-E, women were HPV DNA negative for the corresponding HPV type at month 0 and month 6, irrespective of initial serostatus, and had negative or low-grade cytology at baseline. Types tested for were HPV-16, HPV-18, HPV-31, HPV-33, HPV-35, HPV-39, HPV-45, HPV-51, HPV-52, HPV-56, HPV-58, HPV-59, HPV-66, and HPV-68. Follow-up period started the day after the third vaccine dose. CIN2+ was defined histologically as CIN2, CIN3, adenocarcinoma in situ, or invasive carcinoma; CIN3+ did not include CIN2. Vaccine efficacy point estimates are shown above each bar, and error bars represent 95% CI. CIN=cervical intraepithelial neoplasia. HPV=human papillomavirus. ATP-E=according to protocol for efficacy.

Methods

The trial methods have been previously described in detail, and the results of event-driven analyses presented.[10,11] In this end-of-study analysis after 48 months of

follow-up, we report vaccine efficacy against types other than HPV-16 and HPV-18, using persistent infection, CIN2+, and CIN3+ as endpoints.

Participants

Healthy women aged 15–25 years with no more than six lifetime sexual partners (this exclusion criterion was not applied in Finland, in accordance with local regulatory and ethical requirements[18]) were included in the trial; full inclusion and exclusion criteria, trial locations, and dates have been described previously.[10,11] Women were included regardless of their baseline HPV DNA status, HPV-16 or HPV-18 serostatus, or cytology. Written informed consent was obtained from all adult participants. For minors, written informed assent was obtained from the participant and written informed consent from their parents. The protocol and other materials were approved by independent ethics committees or institutional review boards at each location.

	Vaccine		Control		Efficacy (95% CI)
	Cases	Rate	Cases	Rate	
6-month persistent infection (N=5427 vaccine *vs* 5399 control)					
Non-vaccine A9 species (composite HPV-31/33/35/52/58)	407	2·05	550	2·84	27·6% (17·6 to 36·5)
HPV-31	38	0·18	163	0·81	77·1% (67·2 to 84·4)
HPV-33	53	0·26	92	0·45	43·1% (19·3 to 60·2)
HPV-35	38	0·18	31	0·15	–21·8% (–102·5 to 26·2)
HPV-52	231	1·14	281	1·41	18·9% (3·2 to 32·2)
HPV-58	93	0·45	87	0·43	–6·2% (–44·0 to 21·6)
Non-vaccine A7 species (composite HPV-39/45/59/68)	263	1·31	334	1·68	22·3% (8·4 to 34·2)
HPV-39	111	0·54	139	0·69	20·9% (–2·3 to 38·9)
HPV-45	13	0·06	61	0·30	79·0% (61·3 to 89·4)
HPV-59	45	0·22	43	0·21	–3·9% (–61·7 to 33·1)
HPV-68	112	0·55	122	0·60	8·9% (–18·8 to 30·1)
Other					
HPV-51	253	1·26	334	1·68	25·5% (12·0 to 37·0)
HPV-56	147	0·72	148	0·73	1·4% (–24·8 to 22·0)
HPV-66	141	0·69	138	0·68	–1·5% (–29·3 to 20·3)
CIN2+ with or without co-infection with HPV-16/18 (N=5466 vaccine *vs* 5452 control)					
Non-vaccine A9 species (composite HPV-31/33/35/52/58)	31	0·15	71	0·35	56·6% (33·0 to 72·5)
HPV-31	3	0·01	28	0·14	89·4% (65·5 to 97·9)
HPV-33	5	0·02	28	0·14	82·3% (53·4 to 94·7)
HPV-35	1	0·00	6	0·03	83·4% (–36·6 to 99·6)
HPV-52	14	0·07	20	0·10	30·4% (–45·0 to 67·5)
HPV-58	9	0·04	14	0·07	36·1% (–58·6 to 75·6)
Non-vaccine A7 species (composite HPV-39/45/59/68)	8	0·04	28	0·14	71·6% (36·0 to 88·8)
HPV-39	3	0·01	11	0·05	72·9% (–2·7 to 95·1)
HPV-45	0	0·00	8	0·04	100% (41·7 to 100)
HPV-59	0	0·00	2	0·01	100% (–429·6 to 100)
HPV-68	5	0·02	11	0·05	54·8% (–41·2 to 87·7)
Other					
HPV-51	9	0·04	30	0·15	70·2% (35·6 to 87·6)
HPV-56	0	0·00	7	0·03	100% (31·0 to 100)
HPV-66	3	0·01	11	0·05	72·9% (–2·7 to 95·1)

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Procedures

Women were randomised in a 1:1 ratio to receive either the HPV-16/18 AS04-adjuvanted vaccine (Cervarix, GlaxoSmithKline Biologicals) or control hepatitis A vaccine (GlaxoSmithKline Biologicals) at 0, 1, and 6 months in a double-blind manner. The study protocol prescribed that both groups were to be unblinded after the month 48 visit and offered the cross-over vaccine. Cervical samples were obtained every 6 months for HPV DNA detection and typing. Gynaecological examinations were performed and cytology samples were obtained every 12 months. Collection of cytology and histopathology specimens and the clinical management algorithm for abnormal cytology results and colposcopy referral have been described elsewhere.[10,11,17] A broad spectrum PCR SPF_{10}-$LiPA_{25}$ (version 1 based on licensed Innogenetics SPF10 technology; Labo Biomedical Products, Rijswijk, Netherlands) and type-specific PCR for HPV-16 and HPV-18 DNA were used to test cervical samples and biopsy material for HPV DNA from 14 oncogenic HPV types (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68).[10,19] These types were selected based on the International Agency for Research on Cancer (IARC) list of oncogenic HPV types that was current at the time of writing the study protocol, although the evidence in humans for the carcinogenicity of HPV-66 is now considered to be limited.[20] All CIN cases were reviewed by an independent endpoint committee.

In the present analysis, we describe secondary and exploratory endpoints of vaccine efficacy against persistent infection, CIN2+, and CIN3+ associated with non-vaccine HPV types (31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68) tested by PCR. For persistent infection and CIN2+, we evaluated vaccine efficacy against individual non-vaccine types, and against composite endpoints of four HPV types from the A7 species (HPV-39/45/59/68), five HPV types from the A9 species (HPV-31/33/35/52/58), and 12 non-vaccine HPV types (all HPV types tested by PCR except HPV-16/18—ie, HPV-31/33/35/39/45/51/52/56/58/59/66/68). For CIN3+, we evaluated vaccine efficacy against the 12-type composite. For the composite endpoints, women had to be infected with, or have a lesion associated with, at least one of the HPV types included in the composite.

6-month persistent infection was defined as detection of the same HPV type in consecutive samples over a minimum of 5 months. 12-month persistent infection was defined as detection of the same HPV type in consecutive samples over a minimum of 10 months. CIN2+ was defined as CIN2, CIN3, adenocarcinoma in situ (AIS), or invasive carcinoma; CIN3+ excluded CIN2. Since multiple HPV types are often found in cervical

lesion samples and it is not always possible to assign causality to a particular type, we considered two analyses: a prespecified analysis in which all cases were included, irrespective of whether they were co-infected with HPV-16/18 (referred to hereafter as CIN2+ or CIN3+); and an additional post-hoc analysis in which all CIN2+ or CIN3+ cases that were co-infected with HPV-16/18 were excluded (referred to hereafter as CIN2+ or CIN3+ excluding co-infection with HPV-16/18).

Statistical analysis

Endpoints were evaluated in three cohorts: the ATP-E, the TVC-naive, and the TVC cohorts.[17] The ATP-E cohort included women who were evaluable for efficacy (ie, had a baseline PCR or cytology sample and one further sample available), met all eligibility criteria, complied with the protocol, received all three vaccine doses, and had negative or low-grade cytology at baseline. In the ATP-E cohort, endpoints were assessed in women who were HPV DNA negative at baseline and at month 6 for the HPV type analysed. The TVC-naive included women who had received at least one vaccine dose, were evaluable for efficacy, were HPV DNA negative at baseline for all 14 HPV types tested for, seronegative for HPV-16 and HPV-18, and had negative cytology. Excluding women who were positive for any of the 14 HPV types at baseline is the main reason for the substantially lower number of women included in the TVC-naive than in the ATP-E cohort. The TVC-naive represents the least HPV-exposed analytical group. The TVC included all women who received at least one vaccine dose and were evaluable for efficacy. Endpoints were assessed in the TVC irrespective of women's baseline HPV DNA, cytological status, and serostatus. Licensure of the vaccine was based on analysis of the ATP-E cohort to fully describe the vaccine's profile. However, the TVC and TVC-naive are more relevant from a public health perspective, and we have therefore included all three cohorts in the end-of-study analysis of cross-protection.

The end-of-study analysis was intended to support the efficacy results of the final event-driven analysis.[11] Vaccine efficacy and 95% CIs were calculated using a conditional exact method (webappendix p 8). 95% CIs were calculated for the end-of-study analysis, whereas 97·9% and 96·1% CIs were used for the interim and final event-driven analyses, respectively.[10,11] Results were considered to support statistically significant vaccine efficacy observed in the final event-driven analysis if end-of-study estimates and their 95% CIs were above zero.

Event rates were calculated as the number of cases divided by the total follow-up in years and were expressed per 100 woman years. In the TVC and the TVC-naive, follow-up started the day after the first vaccine dose. In the ATP-E, follow-up started the day after the third vaccine dose. Follow-up for each outcome ended at the time the outcome occurred or at the last available sample (up to month 48). Statistical analyses were done with SAS version 9.1 and Proc StatXact-7 on Windows XP.

	Vaccine		Control		Efficacy (95% CI)
	Cases	Rate	Cases	Rate	
(Continued from previous page)					
CIN2+ excluding co-infection with HPV-16/18 (N=5466 vaccine *vs* 5452 control)					
Non-vaccine A9 species (composite HPV-31/33/35/52/58)	31	0·15	43	0·21	28·3% (–16·5 to 56·3)
HPV-31	3	0·01	18	0·09	83·4% (43·3 to 96·9)
HPV-33	5	0·02	21	0·10	76·3% (35·5 to 93·0)
HPV-35	1	0·00	3	0·01	66·8% (–313·0 to 99·4)
HPV-52	14	0·07	6	0·03	–132·3% (–637·5 to 16·2)
HPV-58	9	0·04	8	0·04	–11·9% (–233·4 to 61·7)
Non-vaccine A7 species (composite HPV-39/45/59/68)	8	0·04	10	0·05	20·4% (–124·0 to 72·7)
HPV-39	3	0·01	5	0·02	40·3% (–206·8 to 90·7)
HPV-45	0	0·00	2	0·01	100% (–429·7 to 100)
HPV-59	0	0·00	1	0·00	100% (–3779·6 to 100)
HPV-68	5	0·02	3	0·01	–65·8% (–967·9 to 67·7)
Other					
HPV-51	9	0·04	9	0·04	0·5% (–182·9 to 65·0)
HPV-56	0	0·00	2	0·01	100% (–429·7 to 100)
HPV-66	3	0·01	7	0·03	57·4% (–86·8 to 92·9)

Women could be infected with multiple HPV types (therefore the number of cases for the composite endpoints might not equal the sum of the cases for each individual type included in the composite). Types tested for were HPV-16, HPV-18, HPV-31, HPV-33, HPV-35, HPV-39, HPV-45, HPV-51, HPV-52, HPV-56, HPV-58, HPV-59, HPV-66, and HPV-68. Women included in the analysis were DNA negative for all 14 HPV types tested for, seronegative for HPV-16 and HPV-18, and had negative cytology at month 0. CIN2+ was defined histologically as CIN2, CIN3, adenocarcinoma in situ, or invasive carcinoma. CIN=cervical intraepithelial neoplasia. HPV=human papillomavirus. TVC-naive=total vaccinated HPV-naive cohort. N=number of evaluable women in each group (vaccine *vs* control). Cases=number of evaluable women reporting at least one event. Rate=number of cases divided by sum of follow-up period (per 100 woman years); follow-up period began the day after the first vaccine dose.

Table 2: **Cross-protective efficacy against 6-month persistent infection and CIN2+ associated with non-vaccine HPV types, in women who were HPV-naive at baseline (TVC-naive)**

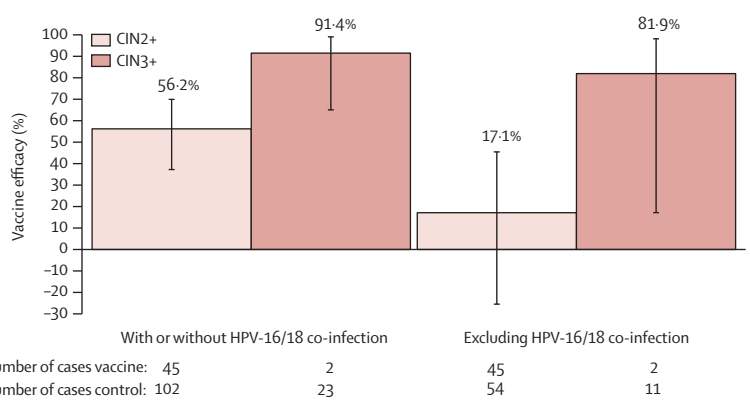

Figure 2: **Vaccine efficacy against CIN2+ and CIN3+ associated with a composite of 12 non-vaccine HPV types, with or without HPV-16/18 co-infection and excluding HPV-16/18 co-infection, in the TVC-naive**

Women had to have a lesion associated with at least one of the HPV types included in the composite. Women included in the analysis of the TVC-naive were HPV DNA negative for the 14 HPV types tested for, seronegative for HPV-16/18, and had negative cytology at month 0. Types tested for were HPV-16, HPV-18, HPV-31, HPV-33, HPV-35, HPV-39, HPV-45, HPV-51, HPV-52, HPV-56, HPV-58, HPV-59, HPV-66, and HPV-68. Follow-up period started the day after the first vaccine dose. CIN2+ was defined histologically as CIN2, CIN3, adenocarcinoma in situ, or invasive carcinoma; CIN3+ did not include CIN2. Vaccine efficacy point estimates are shown above each bar, and error bars represent 95% CI. CIN=cervical intraepithelial neoplasia. HPV=human papillomavirus. TVC-naive=total vaccinated HPV-naive cohort.

The trial is registered with ClinicalTrials.gov, number NCT00122681.

Role of the funding source

The trial was funded by GlaxoSmithKline Biologicals, who designed the study in collaboration with investigators, and coordinated collection, analysis, and interpretation of data. Investigators from the HPV PATRICIA Study Group collected data for the trial and cared for the participants. The authors had full access to all the trial data and had final responsibility for the decision to submit for publication.

Results

A total of 16 114, 11 644, and 18 644 women were included in the ATP-E (vaccine n=8067, control n=8047), TVC-naive (vaccine n=5824, control n=5820), and TVC cohorts (vaccine n=9319, control n=9325), respectively. Compliance was high. Roughly 16% of participants (3034 of 18 644) were lost to follow-up by the end of the study; the number of participants who did not complete the study was balanced between the vaccine and control groups.[17] In the ATP-E cohort, mean and median follow-up times after dose 3 were 39·8 months (SD 8·0) and 41·6 months (range 0–55·5), respectively (3·3 and 3·5 years).

	Vaccine		Control		Efficacy (95% CI)
	Cases	Rate	Cases	Rate	
6-month persistent infection (N=8863 vaccine *vs* 8870 control)					
Non-vaccine A9 species (composite HPV-31/33/35/52/58)	1179	4·00	1364	4·67	14·5% (7·5 to 20·9)
HPV-31	235	0·73	433	1·37	46·3% (36·9 to 54·3)
HPV-33	156	0·48	211	0·66	26·3% (8·9 to 40·4)
HPV-35	128	0·40	106	0·33	–21·1% (–58·2 to 7·1)
HPV-52	643	2·07	698	2·25	8·1% (–2·4 to 17·6)
HPV-58	245	0·76	216	0·67	–13·7% (–37·2 to 5·7)
Non-vaccine A7 species (composite HPV-39/45/59/68)	769	2·50	838	2·73	8·5% (–1·1 to 17·1)
HPV-39	340	1·07	347	1·09	2·0% (–14·2 to 15·8)
HPV-45	70	0·22	153	0·47	54·5% (39·2 to 66·2)
HPV-59	130	0·40	117	0·36	–11·2% (–44·0 to 14·1)
HPV-68	284	0·89	296	0·92	4·0% (–13·3 to 18·7)
Other					
HPV-51	636	2·05	732	2·37	13·7% (3·8 to 22·5)
HPV-56	351	1·10	357	1·12	1·6% (–14·4 to 15·3)
HPV-66	345	1·08	358	1·12	3·7% (–11·9 to 17·2)
CIN2+ with or without co-infection with HPV-16/18 (N=8694 vaccine *vs* 8708 control)					
Non-vaccine A9 species (composite HPV-31/33/35/52/58)	133	0·41	191	0·59	30·4% (12·7 to 44·6)
HPV-31	36	0·11	68	0·21	47·0% (19·5 to 65·7)
HPV-33	31	0·10	64	0·20	51·5% (24·5 to 69·5)
HPV-35	9	0·03	16	0·05	43·7% (–35·3 to 78·1)
HPV-52	55	0·17	61	0·19	9·8% (–32·1 to 38·5)
HPV-58	33	0·10	37	0·11	10·7% (–46·7 to 45·9)
Non-vaccine A7 species (composite HPV-39/45/59/68)	34	0·10	71	0·22	52·1% (27·0 to 69·2)
HPV-39	13	0·04	24	0·07	45·8% (–10·8 to 74·7)
HPV-45	2	0·01	21	0·06	90·5% (61·0 to 98·9)
HPV-59	5	0·02	8	0·02	37·4% (–116·9 to 83·9)
HPV-68	15	0·05	23	0·07	34·7% (–30·6 to 68·3)
Other					
HPV-51	38	0·12	76	0·23	50·0% (25·3 to 67·1)
HPV-56	11	0·03	23	0·07	52·2% (–2·1 to 79·0)
HPV-66	13	0·04	25	0·08	48·0% (–5·6 to 75·6)

(Continues on next page)

In the ATP-E cohort, vaccine efficacy was consistently high against all endpoints associated with HPV-33: 44·8% (95% CI 24·6 to 59·9) for 6-month persistent infection, 68·3% (39·7 to 84·4) for CIN2+, and 59·4% (20·5 to 80·4) for CIN2+ excluding HPV-16/18 co-infection (table 1). Cross-protective efficacy against all endpoints associated with HPV-31 was also observed: 76·8% (69·0 to 82·9) against 6-month persistent infection, 87·5% (68·3 to 96·1) against CIN2+, and 84·3% (59·5 to 95·2) against CIN2+ excluding HPV-16/18 co-infection (table 1). Few events associated with HPV-45 were observed; however, vaccine efficacy was 73·6% (58·1 to 83·9) for 6-month persistent infection and 81·9% (17·0 to 98·1) for CIN2+. Corresponding values against HPV-51 were 16·6% (3·6 to 27·9) and 54·4% (22·0 to 74·2; table 1). In the ATP-E cohort, vaccine efficacy against the composite of 12 non-vaccine HPV types was 46·8% (30·7 to 59·4) for CIN2+, 24·1% (–1·5 to 43·5) for CIN2+ excluding co-infection with HPV-16/18, 73·8% (48·3 to 87·9) for CIN3+, and 62·1% (21·8 to 82·9) for CIN3+ excluding co-infection with HPV-16/18 (figure 1).

In the TVC-naive, vaccine efficacy estimates with 95% CIs above zero were consistently noted for all endpoints associated with HPV-33 and HPV-31 (table 2). For HPV-45, vaccine efficacy was 79·0% (95% CI 61·3 to 89·4) for 6-month persistent infection, 100% (41·7 to 100) for CIN2+, and 100% (–429·7 to 100) for CIN2+ excluding HPV-16/18 co-infection, although the number of events was again small (table 2). Vaccine efficacy with 95% CIs above zero was also found against HPV-51 for 6-month persistent infection (25·5% [12·0 to 37·0]) and CIN2+ (70·2% [35·6 to 87·6]), but not for CIN2+ excluding HPV-16/18 co-infection (0·5% [–182·9 to 65·0]; table 2). For the composite endpoint of 12 non-vaccine HPV types, vaccine efficacy was 56·2% (37·2 to 69·9) against CIN2+, and 17·1% (–25·5 to 45·4) when CIN2+ cases co-infected with HPV-16/18 were excluded. Corresponding values for CIN3+ were 91·4% (65·0 to 99·0) and 81·9% (17·1 to 98·1; figure 2).

The pattern of vaccine efficacy across endpoints in the TVC was similar to the ATP-E and TVC-naive, with lower point estimates as expected in this broader cohort, which included sexually active women with previous or current HPV type-specific infections or lesions under consideration at study entry. Compared with the ATP-E and TVC-naive, more moderate vaccine efficacy, albeit with 95% CIs above zero, was seen in the TVC for 6-month persistent infection with HPV-33, HPV-31, HPV-45, and

HPV-51 (26·3% [8·9 to 40·4], 46·3% [36·9 to 54·3], 54·5% [39·2 to 66·2], and 13·7% [3·8 to 22·5], respectively; table 3). Vaccine efficacy against CIN2+ associated with HPV-33 was 51·5% (24·5 to 69·5), and 50·0% (14·7 to 71·4) against CIN2+ excluding co-infection with HPV-16/18. Vaccine efficacy against CIN2+ was 47·0% (19·5 to 65·7) for HPV-31, 90·5% (61·0 to 98·9) for HPV-45, and 50·0% (25·3 to 67·1) for HPV-51; in the analysis of CIN2+ excluding co-infection with HPV-16/18, the lower limits of the 95% CIs were below zero for these HPV types (table 3). Once again, few events associated with HPV-45 were observed. Vaccine efficacy against the composite of 12 non-vaccine HPV types was 34·2% (20·4 to 45·8) for CIN2+ and 6·2% (–18·1 to 25·6) for CIN2+ excluding co-infection with HPV-16/18 (figure 3). Vaccine efficacy estimates with 95% CIs above zero were consistently seen for CIN3+ associated with the 12-type composite: 47·5% (22·8 to 64·8) and 40·0% (1·1 to 64·2) including and excluding HPV-16/18 co-infection, respectively (figure 3).

Negative vaccine efficacy with both 95% CIs below zero was seen for CIN2+ associated with HPV-52 excluding HPV-16/18 co-infection in the TVC (table 3), and for 12-month persistent infection with HPV-58 in the ATP-E and TVC (webappendix p 3). For other endpoints and cohorts, results for these two HPV types were inconsistent, with both positive and negative vaccine efficacy point estimates.

Table 4 summarises vaccine efficacy estimates across the three different cohorts (ATP-E, TVC-naive, and TVC). Vaccine efficacy estimates for 12-month persistant infection and CIN3+ associated with individual HPV types are shown in the webappendix p 3 and p 5, respectively.

	Vaccine		Control		Efficacy (95% CI)
	Cases	Rate	Cases	Rate	
(Continued from previous page)					
CIN2+ excluding co-infection with HPV-16/18 (N=8694 vaccine *vs* 8708 control)					
Non-vaccine A9 species (composite HPV-31/33/35/52/58)	112	0·35	115	0·35	2·4% (–27·7 to 25·5)
HPV-31	30	0·09	46	0·14	34·7% (–5·7 to 60·2)
HPV-33	22	0·07	44	0·14	50·0% (14·7 to 71·4)
HPV-35	6	0·02	7	0·02	14·2% (–198·2 to 76·2)
HPV-52	45	0·14	24	0·07	–87·9% (–222·4 to –12·1)
HPV-58	30	0·09	22	0·07	–36·6% (–148·5 to 23·8)
Non-vaccine A7 species (composite HPV-39/45/59/68)	28	0·09	34	0·10	17·6% (–40·1 to 51·8)
HPV-39	10	0·03	13	0·04	23·0% (–90·1 to 69·8)
HPV-45	2	0·01	9	0·03	77·8% (–7·4 to 97·7)
HPV-59	5	0·02	5	0·02	–0·1% (–335·1 to 77·0)
HPV-68	12	0·04	8	0·02	–50·2% (–323·6 to 43·5)
Other					
HPV-51	30	0·09	34	0·10	11·7% (–48·7 to 47·8)
HPV-56	7	0·02	7	0·02	–0·1% (–234·5 to 70·0)
HPV-66	8	0·02	16	0·05	–50·0% (–23·9 to 81·5)

Women could be infected with multiple HPV types (therefore the number of cases for the composite endpoints might not equal the sum of the cases for each individual type included in the composite). Types tested for were HPV-16, HPV-18, HPV-31, HPV-33, HPV-35, HPV-39, HPV-45, HPV-51, HPV-52, HPV-56, HPV-58, HPV-59, HPV-66, and HPV-68. Women were included in the analysis irrespective of their HPV DNA or serostatus at month 0. CIN2+ was defined histologically as CIN2, CIN3, adenocarcinoma in situ, or invasive carcinoma. CIN=cervical intraepithelial neoplasia. HPV=human papillomavirus. TVC=total vaccinated cohort. N=number of evaluable women in each group (vaccine *vs* control). Cases=number of evaluable women reporting at least one event. Rate=number of cases divided by sum of follow-up period (per 100 woman years); follow-up period began the day after the first vaccine dose.

Table 3: **Cross-protective efficacy against 6-month persistent infection and CIN2+ associated with non-vaccine HPV types in women irrespective of their baseline HPV DNA and serostatus (TVC)**

Discussion

Data from the end-of-study analysis of PATRICIA show that the HPV-16/18 vaccine provides cross-protective efficacy against 6-month persistent infection and CIN2+ associated with HPV-33, HPV-31, HPV-45, and HPV-51. Consistent vaccine efficacy for all endpoints across all cohorts was seen only for HPV-33. As expected, estimates of vaccine efficacy were generally higher in the TVC-naive and ATP-E cohorts than in the TVC. The TVC-naive and ATP-E cohorts represent the primary target population for the vaccine, in terms of little to no genital HPV exposure, and show the potential effect of the vaccine against new infections and lesions. By contrast, the TVC includes women with pre-existing infections or lesions associated with the HPV types considered in the analyses, which are not expected to be affected by the prophylactic vaccine. Factors that might have limited the generalisability of the study results were enrolment of 80% of the 15–17 year old stratum in a single country (Finland), enrolment of 47% of the 18–25 year old stratum from Asia-Pacific, and exclusion of women with more than six lifetime sexual partners (this criterion did not apply to 15–17 year olds in Finland). Additionally, cross-protective vaccine efficacy could be different in different populations as a result of host (eg, race or ethnicity) and viral factors (eg, variant aminoacids potentially relevant to HPV cross-protective epitopes).

The assessment of protection against non-vaccine oncogenic HPV types poses a considerable challenge in clinical trials, as shown by our analyses. Although CIN2+ is generally the preferred endpoint to evaluate vaccine efficacy against vaccine types, it has several limitations for the evaluation of cross-protection; these limitations also apply to CIN3+, which is less common. First, very large sample sizes and extensive follow-up periods are needed, because lesions are less often associated with non-vaccine types than with HPV-16 or HPV-18. Second, HPV DNA of non-vaccine and vaccine types might be detected in biopsies taken from lesions, and causality of the lesion cannot be definitively assigned to one HPV type. Microdissection of lesions has been proposed as a method for attributing causality, but it is impractical, and it is unclear whether it would be useful.[21] Third, HPV-16 infections are more likely than infections with any other HPV type to progress to a detectable lesion.[22] The carcinogenicity of HPV-16 is unique, according to recognised risk criteria, including attributable fraction of prevalent CIN3 or cancer, probability of persistence, and detection of incident CIN3 or cancer.[23] CIN3+

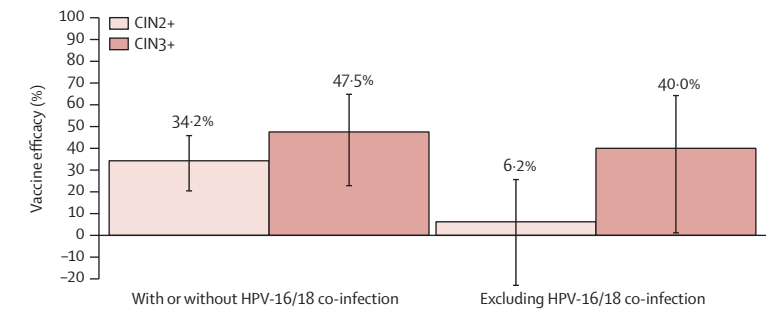

Figure 3: **Vaccine efficacy against CIN2+ and CIN3+ associated with a composite of 12 non-vaccine HPV types, with or without HPV-16/18 co-infection and excluding HPV-16/18 co-infection, in the TVC**
Women had to have a lesion associated with at least one of the HPV types included in the composite. Types tested for were HPV-16, HPV-18, HPV-31, HPV-33, HPV-35, HPV-39, HPV-45, HPV-51, HPV-52, HPV-56, HPV-58, HPV-59, HPV-66, and HPV-68. Women were included in the analysis of the TVC regardless of their HPV DNA or serostatus at month 0. Follow-up period started the day after the first vaccine dose. CIN2+ was defined histologically as CIN2, CIN3, adenocarcinoma in situ, or invasive carcinoma; CIN3+ did not include CIN2. Vaccine efficacy point estimates are shown above each bar, and error bars represent 95% CI. CIN=cervical intraepithelial neoplasia. HPV=human papillomavirus. TVC=total vaccinated cohort.

associated with HPV-16 is diagnosed much earlier than other HPV types,[24] presumably as a result of greater chromosomal instability and corresponding cellular transformation induced by deregulated HPV E6 and E7 oncogene expression.[25] Thus, the increased likelihood of progression of HPV-16 infection to detectable lesions leads to increased referral and therapy, which could differentially affect the development and detection of lesions caused by other HPV types among vaccinated versus non-vaccinated women.

We attempted to address the confounding effect of multiple infections in lesions by calculating two separate estimates of vaccine efficacy against CIN2+ and CIN3+. Our analyses of CIN2+ and CIN3+ with or without co-infection with HPV-16/18 are likely to overestimate cross-protective efficacy, since some cases counted in the analyses were probably caused by HPV-16 or HPV-18. However, our analyses excluding cases co-infected with HPV-16 or HPV-18 are very conservative. For example, cases were excluded if two independent lesions were found in the same woman, one infected with HPV-16/18 and one infected with a non-vaccine type, even if the lesions occurred at different sampling timepoints. Additionally, because of the efficacy of the vaccine against HPV-16/18 infections, fewer cases were co-infected with HPV-16/18 in the vaccine group than in the control group. As a result, most cases removed were from the control group, including some that might have been caused by a non-vaccine type. Thus, true vaccine efficacy against CIN2+ or CIN3+ associated with non-vaccine HPV types possibly lies somewhere between the two estimates. Reports of cross-protection conferred by HPV vaccines against lesions associated with oncogenic HPV types have not always taken this conservative approach.[10,26]

	ATP-E			TVC-naive			TVC		
	6-month persistent infection	CIN2+ with or without HPV-16/18 co-infection	CIN2+ excluding HPV-16/18 co-infection	6-month persistent infection	CIN2+ with or without HPV-16/18 co-infection	CIN2+ excluding HPV-16/18 co-infection	6-month persistent infection	CIN2+ with or without HPV-16/18 co-infection	CIN2+ excluding HPV-16/18 co-infection
Non-vaccine A9 species (composite HPV-31/33/35/52/58)	22·0%*	49·1%*	29·5%	27·6%*	56·6%*	28·3%	14·5%*	30·4%*	2·4%
HPV-31	76·8%*	87·5%*	84·3%*	77·1%*	89·4%*	83·4%*	46·3%*	47·0%*	34·7%
HPV-33	44·8%*	68·3%*	59·4%*	43·1%*	82·3%*	76·3%*	26·3%*	51·5%*	50·0%*
HPV-35	−19·8%	62·5%	39·9%	−21·8%	83·4%	66·8%	−21·1%	43·7%	14·2%
HPV-52	8·3%	27·6%	−25·8%	18·9%*	30·4%	−132·3%	8·1%	9·8%	−87·9%†
HPV-58	−18·3%	28·5%	−7·3%	−6·2%	36·1%	−11·9%	−13·7%	10·7%	−36·6%
Non-vaccine A7 species (composite HPV-39/45/59/66)	11·6%	58·2%*	30·4%	22·3%*	71·6%	20·4%	8·5%	52·1%*	17·6%
HPV-39	4·8%	74·9%*	42·7%	20·9%	72·9%	40·3%	2·0%	45·8%	23·0%
HPV-45	73·6%*	81·9%*	50·1%	79·0%*	100%*	100%	54·5%*	90·5%*	77·8%
HPV-59	−7·5%	80·0%	66·6%	−3·9%	100%	100%	−11·2%	37·4%	−0·1%
HPV-68	2·6%	26·8%	10·1%	8·9%	54·8%	−65·8%	4·0%	34·7%	−50·2%
Other									
HPV-51	16·6%*	54·4%*	13·7%	25·5%*	70·2%*	0·5%	13·7%*	50·0%*	11·7%
HPV-56	−5·3%	46·1%	40·0%	1·4%	100%*	100%	1·6%	52·2%	−0·1%
HPV-66	2·3%	56·4%	36·5%	−1·5%	72·9%	57·4%	3·7%	48·0%	−50·0%

ATP-E=according-to-protocol for efficacy. TVC-naive=total vaccinated HPV-naive cohort. TVC=total vaccinated cohort. CIN=cervical intraepithelial neoplasia. HPV=human papillomavirus. *Vaccine efficacy values with lower limit of the 95% CI above 0. †Negative vaccine efficacy values with entire 95% CI below 0.

Table 4: **Summary of cross-protective efficacy across endpoints and cohorts**

Virological endpoints are valuable in evaluation of cross-protective vaccine efficacy. Persistent infection with oncogenic HPV types usually precedes the detection of CIN2+[13–16] and occurs substantially more often, allowing sufficient statistical power using a realistic sample size in vaccine trials. We chose to use persistent infection of at least 6 months since this was comparable to persistent infection of at least 12 months as a predictor of developing CIN2+ (webappendix p 7),[27] although occurring at higher rates and providing additional statistical power. Unlike CIN2+ and CIN3+, persistent infection directly reflects the effect of vaccination on an individual HPV type, is presumably not biased by multiple infections, and does not pose the problem of assessing causality. Therefore, using virological endpoints as indicative of cross-protection, and clinical endpoints as confirmatory evidence, is a scientifically sound and robust approach.

In our study, cross-protection against endpoints associated with HPV-33—6-month persistent infection, CIN2+, and CIN2+ excluding HPV-16/18 co-infection—was consistently observed across cohorts. HPV-33 is the fourth most prevalent HPV type after HPV-16, HPV-18, and HPV-45 in ICC worldwide (4% prevalence),[3] and growing evidence supports the high-risk nature of HPV-33. For example, HPV-33 incident infections are at a very high risk of progression to CIN2 or CIN3,[28,29] and among women positive for HPV-33, the percent risk of carcinoma in situ or adenocarcinoma in situ, relative to HPV-16, has been reported to be 101% (95% CI 62 to 163)—ie, essentially equal to that for women positive for HPV-16.[24]

Vaccine efficacy against HPV-31 was consistently shown across most cohorts and endpoints. High efficacy estimates for 6-month persistent infection and CIN2+ associated with HPV-31 were obtained in the ATP-E and TVC-naive cohorts. As expected, estimates of vaccine efficacy were lower in the TVC, which was the only cohort where analysis of CIN2+ associated with HPV-31 excluding co-infection with HPV-16/18 resulted in an efficacy estimate with the lower limit of the 95% CI below zero. For HPV-45, the type most closely related to HPV-18, cross-protective vaccine efficacy was seen against 6-month persistent infection and against CIN2+ with and without HPV-16/18 co-infection, across all cohorts. Vaccine efficacy against the composite of tested non-vaccine A7 species, including HPV-45, was seen for some endpoints, probably driven by efficacy against HPV-45. HPV-45 is the third most prevalent type after HPV-16 and HPV-18, and causes roughly 6% of all ICC.[3] HPV-45 is more frequent in adenocarcinoma (12% of cases) than in SCC (5% of cases).[3] Although rarer than SCC, adenocarcinoma represents up to 25% of cervical cancers.[3,30]

We also noted consistent cross-protection against 6-month persistent infection and CIN2+ associated with HPV-51, although at lower levels than for HPV-33, HPV-31, and HPV-45. HPV-51 is ranked as the tenth or eleventh most common HPV type associated with ICC worldwide (about 1% prevalence),[3,5] but ranks higher in precursor cervical lesions such as high-grade squamous intraepithelial lesions (3·6% worldwide).[31] Although there was suggestion of cross-protection against other HPV types, such as HPV-52, HPV-56, and HPV-39, the results for virological and clinical endpoints were not consistent, and might therefore represent chance observations.

Negative vaccine efficacy was noted for some endpoints and cohorts, for HPV-52 and HPV-58. In theory, negative vaccine efficacy could represent a chance finding, a reduced sensitivity of the PCR for some non-vaccine HPV types in case of multiple infections that are more common in the control arm because of efficacy of the vaccine, or the occurrence of HPV type replacement; however, the latter is unlikely in the context of a clinical trial. Moreover, vaccine efficacy results for HPV-52 and HPV-58 were generally inconsistent across endpoints and cohorts. In the long term, postmarketing surveillance programmes will be important to properly characterise any changes in type-specific HPV prevalence and disease incidence, particularly among cervical precancers (ie, CIN2 and CIN3).

In addition to efficacy against the individual HPV types described above, vaccine efficacy was consistently shown across cohorts for the composite of 12 non-vaccine HPV types. Notably, removing HPV-16/18 co-infected lesions from the analysis had little effect on observed cross-protective vaccine efficacy. In the TVC-naive, the point estimates of vaccine efficacy were 91·4% for CIN3+, and 81·9% when CIN3+ cases co-infected with HPV-16/18 were excluded. In the TVC, vaccine efficacy was 47·5% for CIN3+ and 40·0% for CIN3+ excluding HPV-16/18 co-infected cases. Because non-vaccine HPV types are responsible for about 30% of cervical cancers, these data suggest that cross-protection could provide substantial additional protection against cervical cancers beyond protection conferred against HPV-16/18.

Vaccine efficacy was highest for CIN3+ and lower for CIN2+ and persistent infection. Additionally, the difference in vaccine efficacy with exclusion versus non-exclusion of HPV-16/18 co-infections was greater for CIN2+ than for CIN3+. These results are expected because the attributable proportion of A9 and A7 species (which include HPV types for which cross-protection was observed) rises from infection to increasingly severe lesions,[23] and detection of co-infections with any oncogenic HPV type progressively decreases as lesion severity increases.[32,33] In analyses excluding HPV-16/18 co-infection, cases are removed more often from the control group than from the vaccine group, because of the efficacy of the vaccine against HPV-16/18. This introduces a bias against the vaccine (ie, lowers the vaccine efficacy point estimate). Because fewer CIN3+ cases are co-infected than CIN2+ cases, fewer CIN3+ cases are removed from the analysis, so there is less effect on the vaccine efficacy point estimate for CIN3+ than for CIN2+. It is important to note that, although extra benefit offered by cross-protection has important public health value, it is impossible to predict whether individual women will be protected against vaccine or non-vaccine

Panel: Research in context

Systematic review

The present article reports part of a prophylactic HPV vaccine development programme. Studies in the programme were done to achieve licensure of the vaccine and to examine how the vaccine might be best used in real-world settings, and were developed in conjunction with leading experts in HPV vaccine research and with regulatory bodies. Literature related to HPV vaccination studies was systematically followed before the start of the study, during the trial, and during development of the publication (1997–2011). The volume of literature has now increased, and we used our knowledge and expertise to select the trials we thought were most relevant for the present report.

Interpretation

About 30% of invasive cervical cancer is caused by HPV types not included in current prophylactic HPV vaccines. The level of cross-protection against these non-vaccine HPV types is therefore an important component of the overall level of protection against cervical cancer offered by an HPV vaccine. This end-of-study analysis of PATRICIA reports a comprehensive evaluation of cross-protection conferred by the HPV-16/18 vaccine in diverse populations of women, including against the most stringent endpoint, CIN3+. Selection of appropriate endpoints to evaluate cross-protection is a challenge; we included analyses of both virological and clinical endpoints.

Our analysis showed that the HPV-16/18 vaccine offers cross-protective efficacy against HPV-33, HPV-31, HPV-45, and HPV-51. HPV-16/18 and these four types cause about 85% of cervical cancer; moreover, there is a particularly high risk of HPV-33 infections progressing to cervical lesions, and HPV-45 is over-represented in adenocarcinoma. Vaccine efficacy against the most stringent endpoint, CIN3+ associated with 12 non-vaccine HPV types excluding co-infection with HPV-16/18, was around 80% in HPV-naive women. Our results show that cross-protective efficacy might provide substantial additional protection against cervical cancer beyond protection conferred against HPV-16/18. These are important data for doctors and public health bodies when estimating the overall reduction in cervical lesions and invasive cancer likely to result from immunisation programmes using the HPV-16/18 vaccine.

HPV types. Additionally, the duration of cross-protective efficacy is unknown; this should be assessed in long-term follow-up, including population surveillance and effectiveness studies in real-world settings.[18,34]

A possible explanation for the high cross-protection seen with the HPV-16/18 vaccine is the presence of the AS04 Adjuvant System in the vaccine formulation, which enhances the overall immune response. The HPV-16/18 vaccine induces cross-neutralising antibodies for HPV-31 and HPV-45, raising the possibility that such antibodies might effect cross-protection.[35] However, it is unknown whether the levels of cross-reactive antibodies will help sustain vaccine-induced cross-protection against non-vaccine HPV types over the long term. The immune mechanisms of vaccine-induced cross-protection are not fully understood, but are most likely linked to conserved aminoacid sequences or structural similarities within shared neutralising epitopes among HPV types.[6–8,36–39] Only a few HPV types that belong to the same species as HPV-16 (A9: HPV-31 and HPV-33) or HPV-18 (A7: HPV-45) were associated with cross-protection. This suggests that minor differences in aminoacid sequences or structure could be important in the recognition of neutralising epitopes by vaccine-induced antibodies. However, the results for HPV-51 (A5 species) show that the cross-protection induced by the vaccine extends outside the A9 and A7 species, possibly due to sequence-based or functional similarities at critical aminoacids in shared (most likely conformational) epitopes, although efficacy for HPV-51 was lower than for HPV-31, HPV-33, and HPV-45.

In conclusion, our analyses show some of the challenges in evaluating cross-protective efficacy of HPV vaccines. They highlight the importance of using both virological and clinical endpoints, and of observing consistency between these endpoints before concluding on cross-protection. Our analyses also provide additional evidence for cross-protective efficacy of the HPV-16/18 vaccine against HPV-33, HPV-31, HPV-45, and HPV-51 in different cohorts representing diverse groups of women. Overall, we anticipate that the cross-protective efficacy of the HPV-16/18 vaccine when administered to HPV-naive women might provide substantial additional protection against cervical cancer over and above that achieved by efficacy against HPV-16/18 (panel), but long-term follow-up is needed to confirm this.

Contributors

CMW, ML, SMG, AS, XC, DD, FS, and GD formed the core writing team for the manuscript. All authors reviewed and commented on a draft of the manuscript and gave final approval to submit for publication. All authors contributed to study design, acquisition of data or statistical analyses, and interpretation of data. For the HPV PATRICIA Study Group, see webappendix p 9.

Conflicts of interest

DD, GD, FS, KH, and TZ are employees of GlaxoSmithKline Biologicals. DD, GD, FS, and KH own stock in GlaxoSmithKline Biologicals, and GD holds a relevant patent. All investigators at study clinical sites were funded through their institutions to do the study protocol. CMW, DMH, DA, JP, PN, HK, FYA, FXB, SRS, SMG, ML, TFS, AS, XC, JCT, WH, and BR have received funding through their institutions to do HPV vaccine studies for GlaxoSmithKline Biologicals or Merck Sharp & Dohme (Sanofi Pasteur MSD). JP received a research grant through the Helsinki University Hospital Research Institute to conduct clinical trials on HPV vaccination. SRS has also received funding through her institution from CSL to do research on school-based adolescent HPV vaccination. Through the University of New Mexico, CMW has received equipment and reagents for HPV genotyping from Roche Molecular Systems and funding for HPV vaccine studies from GlaxoSmithKline (in addition to the present study) and Merck & Co. FXB is an editor of the international newsletter (HPV TODAY) and guest editor of the journal *Vaccine* to prepare international reviews on topics related to HPV. WAJP, FXB, WH, XC, SMG, PN, JCT, BR, TFS, and AS have received consulting fees. DMH, SMG, SRS, FYA, PN, and TFS have received honoraria; TFS, BR, and FXB have been paid for expert testimony; BR, FYA, SRS, DMH, JCT, and WAJP have received payment for board membership; JCT, FYA, XC, PN, FXB, BR, and TFS have received payment for lectures, including service on speakers bureau; AS, FYA, FXB, and BR have received payment for development of educational presentations; and JS, WAJP, JCT, SRS, PN, XC, FXB, WH, UJ, FYA, JH, SMG, AS, and CMW have received travel reimbursements from GlaxoSmithKline Biologicals or Merck Sharp & Dohme (Sanofi Pasteur MSD), or both. DA has received support for travel from Väestöliitto. S-NC and GL declare that they have no conflicts of interest.

Acknowledgments

This study (NCT00122681/580299/008) was funded and coordinated by GlaxoSmithKline Biologicals. We thank study participants and their families. We thank Mary Greenacre for writing and editorial assistance, and Jenny Andersson (Cromsource) for editorial assistance and manuscript coordination on behalf of GlaxoSmithKline Biologicals, Wavre, Belgium.

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