Influenza vaccine effectiveness in Spain 2013/14: subtype-specific early estimates using the cycEVA study

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Adjusted early estimates of the 2013/14 influenza vaccine effectiveness (VE) in Spain for all age groups was 35% (95% CI: −9 to 62), 33% (95% CI: −33 to 67) and 28% (95% CI: −33 to 61) against any influenza virus type, A(H1N1)pdm09 and A(H3N2) viruses, respectively. For the population targeted for vaccination, the adjusted VE was 44% (95% CI: −11 to 72), 36% (95% CI: −64 to 75) and 42% (95% CI: −29 to 74), respectively. These preliminary results in Spain suggest a suboptimal protective effect of the vaccine against circulating influenza viruses.

Early assessment of influenza vaccine effectiveness in Spain at national level

In the current influenza season, Spain has experienced a relatively early influenza epidemic compared with other European countries [1]. We present here nationwide early estimates of the effectiveness of the 2013/14 seasonal trivalent influenza vaccine in Spain in preventing medically attended laboratory-confirmed influenza-like illness (ILI) infections, by virus type and subtype in all age groups and in the population targeted for vaccination, during the time when the influenza epidemic in Spain was increasing (9 December 2013 to 26 January 2014). Our early estimates suggest a suboptimal protective effect of the vaccine in preventing medically attended A(H1N1)pdm09 and A(H3N2) laboratory-confirmed influenza.

Background

Since 2008, Spain has been providing interim influenza vaccine effectiveness (VE) results using the cycEVA study – casos y controles para la Efectividad de la Vacuna Antigripal [cases and controls for monitoring influenza vaccine effectiveness], the Spanish component of the I-MOVE (Monitoring Vaccine Effectiveness in Europe) network [2,3]. The agreement between interim and final influenza VE estimates supports the use of interim assessments as a proxy for final VE results [4,5].

In February 2013, the Vaccine Strain Selection Committee of the World Health Organization (WHO) formally received for the first time a compilation of preliminary influenza VE estimates for the 2012/13 season from Europe, Canada and the United States (US) [6]. Interim estimates 2013/14 and final 2013 estimates of influenza VE from countries in the northern and southern hemisphere, respectively, together with results of the characterisation of influenza viruses and vaccine serological studies, have contributed again this year to the decision of the Committee in February on the recommended composition of influenza vaccines for the forthcoming (2014/15) northern hemisphere influenza season [7]. Interim 2013/14 VE estimates have shown substantial protection against laboratory-confirmed A(H1N1)pdm09 illness in Canada and US [8,9] but suboptimal protection against this subtype in the Spanish Navarre region [10].

The results presented here at national level – the first at national level in Europe, in a scenario of multiple Spanish regions with probable differences in some epidemiological features or circulating viruses – might

Table 1

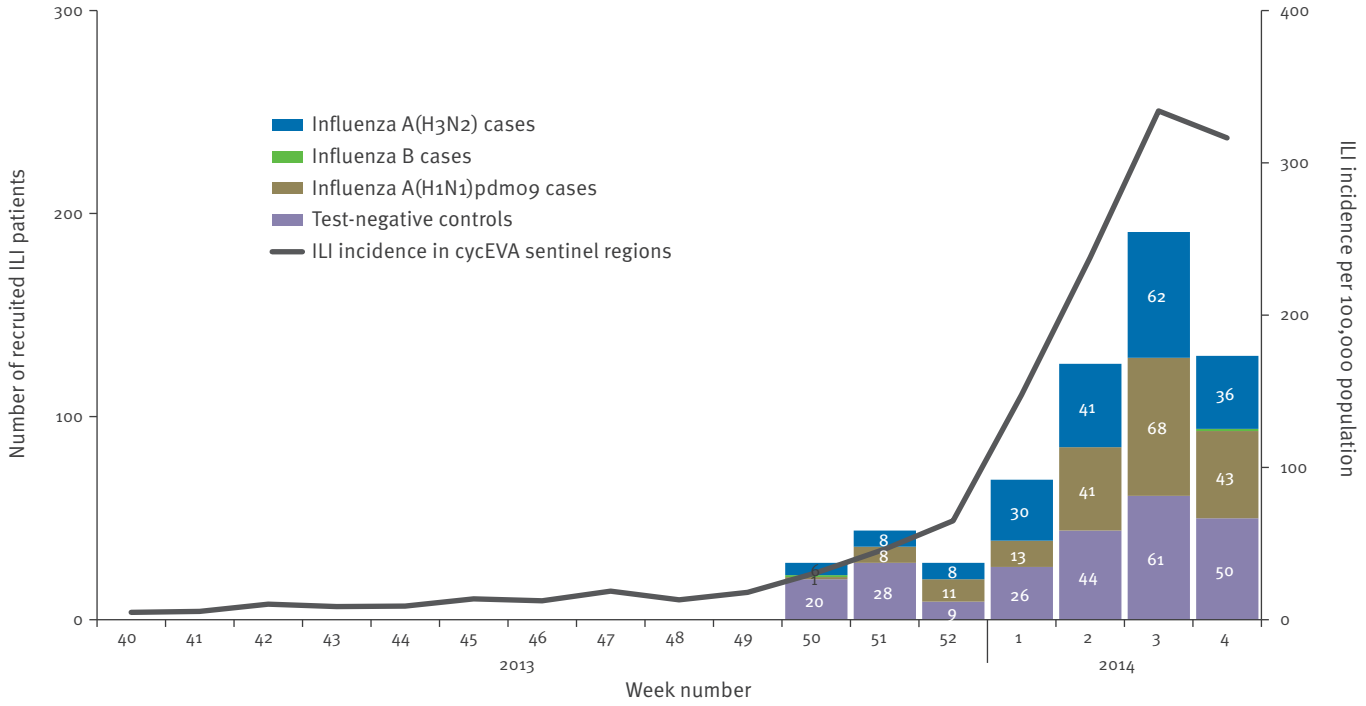
Reference haemagglutinin sequences obtained from the EpiFlu database of the Global Initiative on Sharing Avian Influenza Data (GISAID) and used in phylogenetic analysis, Spanish Influenza Surveillance System, week 40 2013–week 4 2014 (30 September 2013–26 January 2014)

Segment ID	Segment	Country	Collection date	Isolate name	Originating laboratory	Submitting laboratory	Authors
EPI353304	HA	Australia	2007-02-06	A/Brisbane/10/2007	Queensland Health Scientific Services	WHO Collaborating Centre for Reference and Research on Influenza	Iannello, P; Komadina, N
EPI182941	HA	Australia	2009-04-07	A/Perth/16/2009	Pathwest QE II Medical Centre	WHO Collaborating Centre for Reference and Research on Influenza	Deng, Y-M; Iannello, P; Erneste, J; Komadina, N
EPI272062	HA	Australia	2009-06-02	A/Victoria/208/2009	Victorian Infectious Diseases Reference Laboratory	WHO Collaborating Centre for Reference and Research on Influenza	Deng, Y-M; Iannello, P; Caldwell, N; Leang, S-K; Komadina, N
EPI335923	HA	United States	2010-12-30	A/Iowa/19/2010	Iowa State Hygienic Laboratory	Centers for Disease Control and Prevention	Garten, R
EPI279881	HA	Hong Kong (SAR)	2010-07-05	A/Hong Kong/2121/2010	Government Virus Unit	National Institute for Medical Research	Gregory, V
EPI278805	HA	United States	2010-07-13	A/Alabama/05/2010	U.S. Air Force School of Aerospace Medicine	Centers for Disease Control and Prevention	Garten, R
EPI318272	HA	Sweden	2011-02-21	A/Stockholm/18/2011	Swedish Institute for Infectious Disease Control	Swedish Institute for Infectious Disease Control	Brytting, M
EPI358885	HA	Greece	2012-02-01	A/Athens/112/2012	Hellenic Pasteur Institute	National Institute for Medical Research	Gregory, V
EPI450687	HA	Madagascar	2012-06-01	A/Mahajanga/3628/2012	Institut Pasteur de Madagascar	National Institute for Medical Research	Gregory, V
EPI466963	HA	Madagascar	2013-02-25	A/Maevatanana/563/2013	Institut Pasteur de Madagascar	National Institute for Medical Research	Gregory, V
EPI335697	HA	Slovenia	2011-01-25	A/Slovenia/537/2011	Laboratory for Virology, National Institute of Public Health	National Institute for Medical Research	Gregory, V
EPI349103	HA	Australia	2011-10-24	A/Victoria/361/2011	Melbourne Pathology	WHO Collaborating Centre for Reference and Research on Influenza	Deng, Y-M; Caldwell, N; Iannello, P; Komadina, N
EPI377499	HA	United States	2012-04-15	A/Texas/50/2012	Texas Department of State Health Services-Laboratory Services	Centers for Disease Control and Prevention	Garten, R
EPI360950	HA	Germany	2011-07-03	A/Berlin/93/2011	National Institute for Medical Research	Centers for Disease Control and Prevention	Garten, R
EPI397362	HA	United States	2012-07-09	A/Hawaii/22/2012	State of Hawaii Department of Health	Centers for Disease Control and Prevention	Garten, R
EPI467994	HA	Ireland	2013-04-02	A/Ireland/M28390/2013	National Virus Reference Laboratory of Ireland	National Institute for Medical Research	Gregory, V
EPI460558	HA	Russian Federation	2013-03-12	A/Samara/73/2013	WHO National Influenza Centre	National Institute for Medical Research	Gregory, V

We gratefully acknowledge the authors, originating and submitting laboratories of the sequences, other than those coming from our surveillance network, retrieved from GISAID's EpiFlu database.

Figure 1

Recruited influenza cases (n=445) and test-negative controls (n=229) and influenza-like illness incidence in sentinel regions, cycEVA study, Spain, week 50 2013–week 4 2014 (9 December 2013–26 January 2014)



cycEVA: casos y controles para la Efectividad de la Vacuna Antigripal [cases and controls for monitoring influenza vaccine effectiveness]; ILI: influenza-like illness.

add substantial value to the previous estimates from Navarre regarding the suboptimal protective effect of the vaccine. By sharing these results with the scientific community, we are providing evidence that will help to fill the current gaps in knowledge of the relationship between antigenic match and the reported effectiveness of the vaccine.

Estimating vaccine effectiveness and determining virus type

In the 2013/14 influenza season, six of the 17 regional networks belonging to the Spanish Influenza Sentinel Surveillance System participated in the cycEVA study. The methods used were similar to those carried out in previous seasons in the cycEVA study [11].

Influenza cases were ILI patients who tested positive for influenza virus using real-time reverse-transcription polymerase chain reaction or virus culture. Controls were ILI patients with swabs testing negative for any type of influenza virus.

The WHO National Influenza Centre in Madrid selected a subset of influenza isolates from the entire sentinel surveillance system for genetic characterisation by sequencing the amplified HA1 fragment of the viral haemagglutinin gene. Isolates were selected in order to be as representative as possible of viruses circulating in all Spanish regions. Thus they included viruses collected in every phase of the influenza season (beginning, epidemic peak and end of the season). They were also selected to include all ages, irrespective of the vaccination status of the patients. Phylogenetic analysis and molecular evolutionary analyses of the HA1 sequences was conducted using MEGA version 5 [12] in order to characterise the influenza A strains. Reference haemagglutinin nucleotide sequences were obtained from the Global Initiative on Sharing Avian Influenza Data (GISAID) [13] (Table 1).

We used logistic regression to calculate influenza VE from week 50 (starting 9 December) 2013 to week 4 (starting 26 January) 2014, including in the model potential confounding factors and restricting the analysis to those swabbed within seven days of symptom onset. In a sensitivity analysis, we calculated influenza VE in the population targeted for vaccination (individuals over six months-old with chronic conditions, people with risk factors (pregnancy, in women aged 15–44 years, or morbid obesity (body mass index ≥40 kg/m^2), people aged over 59 years (over 64 years in some regions), healthcare workers and caregivers).

Table 2

Characteristics of laboratory-confirmed cases with influenza A(H1N1)pdm09 or A(H3N2) viruses and test-negative controls, cycEVA study, Spain, week 50 2013–week 4 2014 (9 December 2013–26 January 2014) (n=601)

Variables	Test-negative controls, n=229[a]	Influenza A(H1N1)pdm09 cases, n=184[a]	P value[b,c]	Influenza A(H3N2) cases n=188[a]	P value[c,d]
	Number/total number (%)[e]	Number/ total number (%)[e]		Number/ total number (%)[e]	
Age group in years					
0–4	22/229 (9.6)	7/184 (3.8)	0.005	13/188 (6.9)	0.617
5–14	30/229 (13.1)	30/184 (16.3)		20/188 (10.6)	
15–64	153/229 (66.8)	140/184 (76.1)		135/188 (71.8)	
≥65	24/229 (10.5)	7/184 (3.8)		20/188 (10.6)	
Median age in years (range)	36 (0–92)	37 (1–80)	0.868[f]	37 (0–89)	0.778[e]
Male	121/229 (52.8)	102/184 (55.4)	0.599	94/188 (50.0)	0.564
Any chronic condition reported	50/228 (21.9)	32/184 (17.4)	0.251	47/187 (25.1)	0.443
Any risk factor reported[g]	6/211 (2.8)	5/184 (2.7)	0.947	5/165 (3.0)	0.915
Any hospitalisation for chronic conditions in previous year	0/229 (0)	3/184 (1.6)	0.052	0/188 (0)	0.100
Median number of visits to a GP or pediatrician in previous year per patient (range)	3 (0–44)	3 (0–36)	0.450[f]	3 (0–27)	0.775[f]
Smoker	38/227 (16.7)	23/184 (12.5)	0.229	24/186 (12.9)	0.277
Interval between symptom onset and swabbing less than 4 days	223/229 (97.4)	178/184 (96.7)	0.700	184/188 (97.9)	0.744
Population targeted for vaccination	78/218 (35.8)	43/184 (23.4)	0.027	66/172 (38.4)	0.598
Vaccination status					
All ages					
Received seasonal 2013/14 vaccine[h]	38/229 (16.6)	21/184 (11.4)	0.085	30/188 (16.0)	0.681
Received both seasonal 2013/14 and 2012/13 vaccines	35/229 (15.3)	20/184 (10.9)	0.278	28/188 (14.9)	0.623
Targeted for vaccination					
Received seasonal 2013/14 vaccine[h]	28/78 (35.9)	12/43 (27.9)	0.371	17/66 (25.8)	0.191
Received both seasonal 2013/14 and 2012/13 vaccines	24/78 (30.8)	11/43 (25.6)	0.760	16/66 (24.2)	0.424

GP: general practitioner; ILI: influenza-like illness.

[a] Cases and controls recruited during the specified time period and with an interval between ILI symptom onset and swabbing of less than eight days.

[b] P value for A(H1N1)pdm09 cases versus controls.

[c] Chi-squared test or Fisher's exact test.

[d] P value for A(H3N2) cases versus controls.

[e] Unless otherwise indicated. The denominator changes for variables in which the information was missing for some patients.

[f] Non-parametric test of the median.

[g] Defined as pregnancy (in women aged 15–44 years) and/or morbid obesity (body mass index ≥40 kg/m^2).

[h] Vaccination at least 14 days before the onset of influenza like illness symptoms.

Early national vaccine effectiveness estimates

Description of the 2013/14 influenza season in Spain

The 2013/14 influenza season in Spain started in week 1 (30 December 2013–5 January 2014) and reached the epidemic peak in week 4 (20–26 January 2014) at both the national level and in the six regions participating in the cycEVA study [14]. It was a medium-intensity influenza season, clearly dominated by mixed circulation of influenza A viruses: 61% (571/929) A(H1N1) pdm09 and 39% (358/929) A(H3N2) influenza [14].

Participants' characteristics

Among the 217 participating sentinel physicians in the study, 167 (77%) recruited at least one ILI patient. Of the 687 ILI patients recruited, 202 (29%) belonged to the population targeted for influenza vaccination. After excluding 15 patients swabbed more than seven days

Table 3

Crude and adjusted seasonal vaccine effectiveness estimates against laboratory-confirmed influenza by virus type/subtype, overall and among the target population for influenza vaccination, cycEVA study, Spain, week 50 2013–week 4 2014 (9 December 2013–26 January 2014)

Population included	All influenza viruses	Influenza A(H1N1)pdm09	Influenza A(H3N2)
All patients			
Number of patients for the analysis: cases + controls	674	413	417
Number of cases/controls	445/229	184/229	188/229
Number of vaccinated cases/vaccinated controls	53/38	21/38	30/38
Crude VE % (95% CI)	32 (−7 to 56)	35 (−15 to 63)	5 (−61 to 43)
Adjusted VE[a] %(95% CI)	35 (−9 to 62)	33 (−33 to 67)	28 (−33 to 61)
Population targeted for vaccination			
Number of patients for the analysis: cases + controls	299	121	144
Number of cases/controls	121/78	43/78	66/78
Number of vaccinated cases/vaccinated controls	30/27	12/27	17/27
Crude VE % (95% CI)	38 (−16 to 67)	27 (−65 to 98)	34 (−35 to 68)
Adjusted VE[a] % (95% CI)	44 (−11 to 72)	36 (−64 to 75)	42 (−29 to 74)

CI: confidence interval; VE: vaccine effectiveness.

[a] Adjusted for age (age groups adjusted for: 0–4, 5–14, 15–64 and ≥65 years), sex, severity, number of general practitioner visits, smoking history (had ever smoked), chronic conditions, pregnancy (in women aged 15–44 years), morbid obesity (body mass index ≥40 kg/m²) and week of swabbing.

after symptom onset, 674 ILI patients were included in the study, comprising 445 influenza cases – 188 with influenza A(H3N2) virus, 184 A(H1N1)pdm09, 71 influenza A not subtyped and two with influenza B virus– and 229 test-negative controls (Figure 1).

The percentage of the population targeted for vaccination was higher in the controls (35.8%, 78/218,) than in the A(H1N1)pdm09 cases (23.4%, 43/184) (Table 2). Vaccine coverage with the 2013/14 influenza vaccine was not statistically different among controls and A(H1N1)pdm09 or A(H3N2) cases, in all age groups and among the population targeted for vaccination. The majority of cases (96.7–97.9%, 178/184–184/188) and controls (97.4%, 223/229) were swabbed less than four days after symptom onset.

Of the 89 people vaccinated, there were 54 vaccine failures: 30 were positive for influenza A(H3N2) virus, 21 for influenza A(H1N1)pdm09 virus and three with an unknown influenza virus. Of the 54 vaccine failures, 30 were cases belonging to the target population for vaccination.

Vaccine effectiveness estimates

The adjusted influenza VE for all age groups was 35% (95% CI: −9 to 62), 33% (95% CI: −33 to 67) and 28% (95% CI: −33 to 61) against any influenza virus type, A(H1N1)pdm09 and A(H3N2) viruses, respectively (Table 3).

Among the population targeted for vaccination, the adjusted influenza VE against any influenza virus type, A(H1N1)pdm09 and A(H3N2) viruses was 44% (95% CI: −11 to 72), 36% (95% CI: −64 to 75) and 42% (95% CI: −29 to 74), respectively (Table 3).

Genetic analysis of selected isolates

Sequence analysis of the amplified HA1 genome fragments showed that all 93 influenza A(H1N1)pdm09 viruses studied clustered into the group 6B [15] represented by A/Norway/2417/2013 and defined by D97N, K163Q, S185T, S203T, A256T and K283E amino acid mutations compared with the vaccine virus A/California/07/2009.

Regarding influenza A(H3N2) virus, all 61 viruses studied clustered into the group 3C [15] which includes the A/Texas/50/2012 vaccine virus strain, but harboured some amino acid changes that make it possible to differentiate them into two subsets (named 3C.2 and 3C.3) (representative isolates are shown in Figure 2, including viruses collected in past seasons for a better understand the genetic drift of influenza A viruses). Six of the 61 viruses clustered within subgroup 3C.2 represented by A/Ireland/M28390/2013, defined by the HA1 amino acid substitution N128T. The remaining 55/61 viruses (90%) clustered within the subgroup 3C.3 represented by A/Samara/73/2013 and defined by N128A and R142G amino acid substitutions. Interestingly, we could differentiate 23 viruses within the 3C.3 subgroup with an additional L157S change, most of them (20 of 23) harbouring a second N122D mutation. Another subset of six viruses harbouring the K160R amino acid substitution could be identified within the 3C.3 subgroup. Changes in influenza A(H3N2) viruses were referred to the A/Texas/50/2012 vaccine virus strain.

Figure 2

Phylogenetic tree showing genetic differences in HA1 fragment of the haemagglutinin of influenza A(H3N2) circulating viruses, Spanish Influenza Surveillance System, Spain, week 40 2013–week 4 2014 (30 September 2013–26 January 2014)

Phylogenetic relationships were inferred using the MEGA5 programme applying the neighbor-joining method and the Kimura 2-parameter model [12]. Representative isolates are shown, including viruses collected in past seasons to illustrate genetic drift. Viruses in bold are representative of groups 3C.1, 3C.2 and 3C.3, according to the European Centre for Disease Prevention and Control's *Influenza virus characterisation* [15].

Discussion

Our interim point estimate in preventing A(H1N1) pdm09 infections was 33% in a 2013/14 season with circulating A(H1N1)pdm09 strains antigenically similar and genetically well conserved at the European level, as of week 4/2014 [15,16]. Suboptimal protective effects against well-conserved A(H1N1)pdm09 virus were previously described in Europe during the 2011/12 season by the I-MOVE network [17]. In Spain, during the 2010/11 season, early VE estimates against well-matched A(H1N1)pdm09 virus were also found to be lower than 50% (49%; 95% CI: 3 to 73) [2], which were highly consistent with the final estimates, 46% (95% CI: 0 to 72) [18]. Estimates recently published by Canada and the US for the 2013/14 season [8,9] against A(H1N1)pdm09 were higher than our results. A higher protective effect of the vaccine against A(H1N1) pdm09 in North America compared with Spain could be due to different characteristics of the circulating A(H1N1)pdm09 viruses: most of the viruses analysed were shown to be antigenically similar to the vaccine strain in Canada and the US. In Spain, antigenic tests for A(H1N1)pdm09 virus are unfortunately not yet available. In addition, in light of the positive effect of previous influenza vaccination described in Canada [19] and Spain [18], a higher proportion of the population previously vaccinated with the 2009 monovalent pandemic vaccine in Canada (about 40%) [8] compared with that in Spain (<10%) [18,20] could partly explain the higher VE estimates observed in Canada. The use of different types of influenza vaccine could also contribute to the differences between the results of both studies. However, our results were in line (VE below 50%) with those recently published by the Navarre region [10], a Spanish region that also participates in the cycEVA study and I-MOVE network. In the Navarre study, patients recruited in primary healthcare and in hospitals were included, giving similarly low influenza VE estimates in both settings. These observations were in accordance with the evolution of the influenza epidemic in Spain this season: a considerably higher number of severe hospitalised laboratory-confirmed cases were seen than in the two previous seasons. Of these cases, 40% had received the seasonal influenza vaccine [13]. The reasons behind these highly variable estimates of VE are still unclear.

Subtype-specific estimates of VE for influenza A(H3N2) were also in the lower range of VE points described in previous seasons (range: 25–60% [11,21-23]), with adjusted estimates of 28% and 42% for all age groups and population targeted for vaccination, respectively. Reduced protection from influenza A(H3) infection has been described in previous seasons worldwide, including in Spain and the rest of Europe during the 2011/12 season, when A(H3) last circulated as the predominant virus, but was poorly matched to the vaccine [11,23]. The importance of the amino acid changes we describe in the circulating A(H3N2) virus in Spain will be studied at the end of the season once the haemagglutination inhibition assays have been carried out. However, it is important to highlight that the L157S and N122D mutations identified are located in the HA1 antigenic sites B and A, respectively, of A(H3N2) viruses: this could indicate a suboptimal protective effect of the current vaccine against A(H3N2) virus in Spain.

For the 2014/15 northern hemisphere influenza season, WHO has recommended the inclusion of the same strains included in the current seasonal influenza vaccine [24]. Final estimates in Spain with a larger sample size will allow us to confirm the extent of the protective value of the 2013/14 influenza vaccine in Spain and could give an indication of what could be expected in other countries in the northern hemisphere

Although VE estimates are subject to change over time, some studies have demonstrated agreement between interim and final influenza VE estimates, with early estimates within five to seven percentage points of final estimates [5,22]. Using the cycEVA study, the early [2,3] and final estimates [11,18] of the influenza VE in the 2010/11 and 2011/12 seasons in Spain have been similar.

The main limitation of our study was the sample size, which makes estimates for virus subtypes imprecise; therefore, final estimates should be obtained at the end of the influenza season.

Early estimates of influenza VE can help to guide health authorities in influenza prevention and provide useful information for the WHO strain selection process. Future influenza VE studies worldwide are necessary to gain more knowledge about which virus amino acid changes could be influencing the protective effect of the current influenza vaccines. Although our results indicate the protection against A(H1N1)pdm09 and A(H3N2) viruses was suboptimal, the VE was higher among those at risk of severe influenza complications, underlying the importance of annual influenza vaccination. The suboptimal protective effect of the vaccine should also lead to a clear public health message underlying the importance of early antiviral treatment for patients at high risk of influenza complications, and the adoption of non-pharmacological preventive measures to avoid influenza infection.

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Conflict of interest

None declared.

Authors' contributions

Silvia Jiménez-Jorge and Amparo Larrauri designed the study. Silvia Jiménez-Jorge wrote the first draft of the manuscript and undertook the statistical analysis. Silvia Jiménez-Jorge, Salvador de Mateo and Amparo Larrauri participated in data analysis, writing and interpretation of the results. Francisco Pozo and Inmaculada Casas were responsible for the virus characterisation and contributed with the interpretation of the virological data. All authors participated in the interpretation of the data, contributed to the revision of the draft manuscript and approved the final version.

References

1. European Centre for Disease Prevention and Control (ECDC). Weekly influenza surveillance overview. 31 January 2014. Main surveillance developments in week 4/2013 (20–26 January 2014). Stockholm: ECDC; 2014. Available from: http://ecdc.europa.eu/en/publications/Publications/influenza-surveillance-overview-31-jan-2014.pdf
2. Savulescu C, Jiménez-Jorge S, de Mateo S, Ledesma J, Pozo F, Casas I, et al. Effectiveness of the 2010/11 seasonal trivalent influenza vaccine in Spain: preliminary results of a case-control study. Euro Surveill. 2011;16(11):pii=19820.
3. Jimenez-Jorge S, de Mateo S, Pozo F, Casas I, Garcia CM, Castilla J, et al. Early estimates of the effectiveness of the 2011/12 influenza vaccine in the population targeted for vaccination in Spain, 25 December 2011 to 19 February 2012. Euro Surveill. 2012;17(12):pii: 20129.
4. Belongia EA, Kieke BA, Donahue JG, Coleman LA, Irving SA, Meece JK, et al. Influenza vaccine effectiveness in Wisconsin during the 2007-08 season: comparison of interim and final results. Vaccine. 2011;29(38):6558-63. http://dx.doi.org/10.1016/j.vaccine.2011.07.002
5. Sullivan SG, Kelly H. Late season interim estimates of influenza vaccine effectiveness reliably predict end of season estimates in Victoria, Australia, 2007 to 2012. Euro Surveill. 2013;18(41):pii=20605. http://dx.doi.org/10.2807/1560-7917.ES2013.18.41.20605
6. European Centre for Disease Control and Prevention (ECDC). WHO Recommendation on influenza virus vaccines for the Northern Hemisphere 2013-2014 season. Stockholm: ECDC; 15 March 2013. Available from: http://www.ecdc.europa.eu/en/activities/sciadvice/_layouts/forms/Review_DispForm.aspx?ID=737&List=a3216f4c-f040-4f51-9f77-a96046dbfd72
7. World Health Organization (WHO). Recommended composition of influenza virus vaccines for use in the northern hemisphere 2014-15 influenza season and development of candidate vaccine viruses for pandemic preparedness. Geneva: WHO; 2014. Questions and answers. Available from: http://www.who.int/influenza/vaccines/virus/recommendations/201402_qanda_recommendation.pdf
8. Skowronski DM, Chambers C, Sabaiduc S, De Serres G, Dickinson JA, Winter AL, et al. Interim estimates of 2013/14 vaccine effectiveness against influenza A(H1N1)pdm09 from Canada's sentinel surveillance network, January 2014. Euro Surveill. 2014;19(5):pii=20690. http://dx.doi.org/10.2807/1560-7917.ES2014.19.5.20690
9. Flannery B, Thaker SN, Clippard J, Monto AS, Ohmit SE, Zimmerman RK, et al. Interim estimates of 2013-14 seasonal influenza vaccine effectiveness - United States, February 2014. MMWR Morb Mortal Wkly Rep. 2014;63(7):137-42.
10. Castilla J, Martinez-Baz I, Navascues A, Fernandez-Alonso M, Reina G, Guevara M, et al. Vaccine effectiveness in preventing laboratory-confirmed influenza in Navarre, Spain: 2013/14 mid-season analysis. Euro Surveill. 2014;19(6):pii=20700. http://dx.doi.org/10.2807/1560-7917.ES2014.19.6.20700
11. Jiménez-Jorge S, de Mateo S, Delgado-Sanz C, Pozo F, Casas I, Garcia-Cenoz M, et al. Effectiveness of influenza vaccine against laboratory-confirmed influenza, in the late 2011-2012 season in Spain, among population targeted for vaccination. BMC Infect Dis. 2013;13(1):441. http://dx.doi.org/10.1186/1471-2334-13-441
12. Tamura K, Peterson D, Peterson N, Stecher G, Nei M, Kumar S. MEGA5: molecular evolutionary genetics analysis using

maximum likelihood, evolutionary distance, and maximum parsimony methods. Mol Biol Evol. 2011;28(10):2731-9. http://dx.doi.org/10.1093/molbev/msr121

13. The GISAID Initiative. GISAID. Available from: http://www.gisaid.com/
14. Instituto de Salud Carlos III. Informes anuales y semanales de Vigilancia de la Gripe en Espa-a. Sistema de Vigilancia de la Gripe en Espa-a. [Annual and weekly national influenza reports. Spanish Influenza Surveillance System]. Madrid: Centro Nacional de Epidemiología Instituto de Salud Carlos III; 2012. [Accessed 15 January 2014]. Spanish. Available from: http://www.isciii.es/cne-gripe-infsemanal
15. European Centre for Disease Prevention and Control (ECDC). Influenza virus characterisation. Summary Europe December 2013. Stockholm: ECDC; 2014. p. 1. Available from: http://www.ecdc.europa.eu/en/publications/Publications/influenza-virus-characterisation-dec-2013.pdf
16. Instituto de Salud Carlos III. Informe semanal de Vigilancia de la gripe en Espa-a. Semana 05/2014 (27 de enero - 2 de febrero 2014). Nº 379. Comienza la fase de descenso de la onda epidémica 2013/14 en Espa-a, una vez sobrepasado el pico máximo de incidencia de gripe. Sistema de Vigilancia de la Gripe en Espa-a. [Influenza activity is decreasing in Spain after reaching the 2013/14 influenza epidemic peak]. Madrid: Instituto de Salud Carlos III; 2014. Spanish. Available from: http://vgripe.isciii.es/gripe/documentos/20132014/boletines/grno52014.pdf
17. Kissling E, Valenciano M. Early estimates of seasonal influenza vaccine effectiveness in Europe, 2010/11: I-MOVE, a multicentre case-control study. Euro Surveill. 2011;16(11):pii=19818.
18. Jiménez-Jorge S, Savulescu C, Pozo F, de Mateo S, Casas I, Ledesma J, et al. Effectiveness of the 2010-11 seasonal trivalent influenza vaccine in Spain: cycEVA study. Vaccine. 2012;30(24):3595-602. http://dx.doi.org/10.1016/j.vaccine.2012.03.048
19. Skowronski DM, Janjua NZ, Sabaiduc S, De Serres G, Winter AL, Gubbay JB, et al. Influenza A/subtype and B/lineage effectiveness estimates for the 2011-12 trivalent vaccine: cross-season and cross-lineage protection with unchanged vaccine. J Infect Dis. 2014; Jan 19. [Epub ahead of print]. http://dx.doi.org/10.1093/infdis/jiu048
20. Savulescu C, Jiménez-Jorge S, De Mateo S, Pozo F, Casas I, Perez-Bre-a P, et al. Using surveillance data to estimate pandemic vaccine effectiveness against laboratory confirmed influenza A(H1N1)2009 infection: two case control studies, Spain, season 2009-2010. BMC Public Health. 2011;11:899. http://dx.doi.org/10.1186/1471-2458-11-899
21. Fielding JE, Grant KA, Papadakis G, Kelly HA. Estimation of type- and subtype-specific influenza vaccine effectiveness in Victoria, Australia using a test negative case control method, 2007-2008. BMC Infect Dis. 2011;11:170. http://dx.doi.org/10.1186/1471-2334-11-170
22. Belongia EA, Kieke BA, Donahue JG, Greenlee RT, Balish A, Foust A, et al. Effectiveness of inactivated influenza vaccines varied substantially with antigenic match from the 2004-2005 season to the 2006-2007 season. J Infect Dis. 2009;199(2):159-67. http://dx.doi.org/10.1086/595861
23. Kissling E, Valenciano M, Larrauri A, Oroszi B, Cohen JM, Nunes B, et al. Low and decreasing vaccine effectiveness against influenza A(H3) in 2011/12 among vaccination target groups in Europe: results from the I-MOVE multicentre case-control study. Euro Surveill. 2013;18(5):pii=20390.
24. World Health Organization (WHO). Recommended composition of influenza virus vaccines for use in the 2014-2015 northern hemisphere influenza season. Geneva: WHO; 20 February 2014. Available from: http://www.who.int/influenza/vaccines/virus/recommendations/2014_15_north/en/