Vaccine: X 12 (2022) 100220

Contents lists available at ScienceDirect

Vaccine: X

journal homepage: www.elsevier.com/locate/jvacx

Differentiation of influenza B lineages circulating in different regions of Brazil, 2014–2016, using molecular assay

Daniela Bernardes Borges da Silva, Katia Corrêa de Oliveira Santos, Margarete Aparecida Benega, Terezinha Maria de Paiva *

Respiratory Virus Laboratory/NDR/VC, Institute Adolfo Lutz, Brazil/Nacional Influenza Centre/World Health Organization

ARTICLE INFO

Article history:
Received 7 April 2022
Received in revised form 27 September 2022
Accepted 28 September 2022
Available online 30 September 2022

Keywords:
Influenza B lineages
Virological surveillance
RT- qPCR
Vaccine composition
Southern hemisphere
Brazil

ABSTRACT

Background: Two antigenically and genetically distinct lineages of influenza B viruses (B/Victoria and B/Yamagata) have been co-circulating worldwide since 2002. Virological surveillance is essential to differentiate between both lineages with a view to the annual updating of the B component for the trivalent or quadrivalent influenza vaccine composition.

Methods: The samples analyzed in the present study were collected by influenza sentinel units located in the Southeast, Midwest, North, and Northeast regions of Brazil, part of the National Influenza Virus Surveillance Network, coordinated by the Ministry of Health of Brazil. A total of 870 influenza B positive samples by reverse transcription real – time polymerase chain reaction (RT-qPCR), collected during 2014, 2015, and 2016 influenza seasons, were submitted to the influenza B lineage genotyping panel for characterization as B/Yamagata or Victoria lineages using RT-qPCR.

Results: Of the 197 samples analyzed in 2014, a total of 160 (81 %) corresponded to the B/Yamagata lineage, 19 (10 %) to the B/Victoria lineage, and 18 (9 %) to indeterminate lineages. Of the 190 samples analyzed in 2015, a total of 124 (65 %) corresponded to the B/Yamagata lineage; 55 (29 %) to the B/Victoria lineage, whereas 11 (6 %) were of indeterminate lineages. Of the 483 samples analyzed in 2016, a total of 297 (62 %) corresponded to the B /Victoria lineage; 174 (36 %) to the B/Yamagata lineage and 12 (2 %) to indeterminate lineages. This cross-sectional study revealed influenza B virus (IBV) infection in all age groups, and among them, the highest prevalence was observed in individuals between 11 and 49 years of age Our findings demonstrate the match between influenza B virus lineages recommended by the World Health Organization (WHO) for the trivalent vaccine composition to be used in the Southern Hemisphere (SH) and the predominant circulating viruses during the 2014, 2015, and 2016 seasons.

© 2022 The Author(s). Published by Elsevier Ltd. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

1. Background

Influenza viruses are recognized as a significant threat to global public health due to annual seasonal epidemics and pandemic events. The burden of respiratory diseases caused by influenza viruses is revealed by an estimated 1 billion cases, 3–5 million severe cases, and 290,000–650,000 deaths around the world yearly [1,2].

Influenza viruses belong to the *Orthomyxoviridae* family, composed of 7 genera and nine species. Influenza is grouped into four genera with one species per genus: Alphainfluenzavirus, Betainfluenzavirus, Deltainfluenzavirus, and Gammainfluenzavirus, represented by the following species types: Influenza A virus, Influenza B virus, Influenza C virus, and the recently discovered influenza D virus, respectively [3–5]. Influenza A virus is classified into subtypes based on two proteins on the virus's surface: hemagglutinin (H) and neuraminidase (N). To date, 18 different hemagglutinin subtypes (H1–H18), and 11 different neuraminidase subtypes (N1–N11) have already been identified [6]. The biological properties and the diversity of the host range, presented by the influenza A virus, are a constant concern for the global public health context due to the favorable scenarios for the emergence of a pandemic strain [6,7]. Influenza B viruses are restricted to human beings, although rare spillover has already been identified in seals [8,9]. In addition, influenza B viruses are not divided into subtypes, such as influenza A viruses; instead, they are two antigenically and genetically distinct lineages, known as B/Victoria/2/87-like and B/Yamagata/16/88-like, which have been co-circulating in human populations since 1983 [10].

Curiously, from 1991 to 2000, the influenza B/Victoria lineage circulating virus was limited to Southeastern Asia. Outside Asia,

* Corresponding author.

https://doi.org/10.1016/j.jvacx.2022.100220
2590-1362/© 2022 The Author(s). Published by Elsevier Ltd.
This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

its first occurrence was observed in Hawaii during May-June 2001 [11]. In Brazil, the reemerging B/Victoria lineage variant (B/Hong Kong/330/2002-like) was first identified in the city of São Paulo and during an outbreak of acute respiratory illness in Araraquara, state of São Paulo, during the 2002 influenza season [12,13].

The epidemiology of influenza B viruses has changed since the reemerging of the B/Victoria lineage in 2001, allowing the co-circulation of the two distinct lineages. This scenario triggered the development of epidemiologic studies to obtain the actual incidence of IBV infections during the seasonality of influenza viruses worldwide. In this regard, a global influenza B study (GIBS) project was created in 2012 to collect data on the epidemiology and global burden of respiratory diseases caused by IBV to support prevention policies [24].

Considering the absence of cross-protection between both lineages, there is a global concern about the influenza prevention and control strategies provided by trivalent vaccines. In 2013, the WHO presented its first recommendation regarding the annual influenza vaccine composition, including both lineages [14,15].

Fifteen years after the reemerging of the B/Victoria lineage, a new challenge for influenza vaccine composition arises due to the evolutionary pattern of the B/Victoria lineage, presenting a double deletion and designated as subclade V1A.1 (previously V1A – 2 DEL). The emerging V1A.1 subclade is represented by the B/Colorado/06/2017, identified in the USA, during the 2016/2017 influenza season, which is antigenically distinguishable from the B/Brisbane/60/2008 reference virus [16].

Following the evolutionary process, viruses presenting triple deletion in the HA gene were identified in Asia, Africa, Europe, and America [17]. This new emerging subclade was designated as V1A.3 (previously V1A–3DEL) represented by the B/Washington/02/2019, which is antigenically distinguishable from the B/Colorado/06/2017.

The B/Yamagata lineage viruses are classified into three clades: 1, 2, and 3. To date, the evolutionary pattern of this lineage has not been implicated in generating a subclade [18,22]**.** Influenza C and D have not represented a global public health concern until now [19,20]. However, from an epidemiological point of view, influenza A virus is of public concern due to its ability to rearrange genes with different species of birds and animals, generating potential pandemic strains; in contrast, human species is the host of IBV making the rearrangements of genes with different species unfeasible. Although global surveillance shows that influenza B viruses are responsible for approximately 20–25 % of infections during annual influenza seasons, in some seasons, they represent more than 50 % of infections, contributing to the overall mortality and morbidity, mainly in children <5 years old [21–24]. Considering the evolutionary pattern of influenza B viruses, our study aims to provide information regarding influenza B lineages circulating in different regions of Brazil, using RT-qPCR, instead of the hemagglutination inhibition assay, for the rapid identification of both antigenically and phylogenetically distinct influenza B viruses to obtain the benefits of quadrivalent vaccines.

2. Materials and methods

2.1. Study design: Cross-sectional study

Influenza virus surveillance in Brazil comprises sentinel surveillance of influenza-like illness (ILI), severe acute respiratory illness in patients hospitalized in intensive care units (SARI – ICU), and universal SARI surveillance.

The universal SARI surveillance monitors hospitalized cases and deaths intending to identify the behavior of the Influenza virus in the country to guide decision-making in a scenario that requires the positioning of the Ministry of Health and Municipal and State Health Departments.

Routine influenza-like illness (ILI) surveillance consisted of receiving systematically collected nasopharyngeal aspirates or combined oropharyngeal plus nasopharyngeal swabs from individuals presenting clinical history of ILI, ie sudden onset of fever, accompanied by cough and/ or sore throat and at least one of the following symptoms: headache, myalgia or arthralgia with no other diagnosis. Respiratory specimens were collected at selected sentinel units located in the state of São Paulo (SP, Southeast region) and other states located in the Midwest: Mato Grosso (MT), Mato Grosso do Sul (MS), Goiás (GO) and Distrito Federal (DF); the Northeast: Piauí (PI); and the North: Rondônia (RO) and Tocantins (TO). These states are part of the regions covered by the Instituto Adolfo Lutz – a National Influenza Centre (NIC) recognized by the WHO – in the National Influenza Virus Surveillance Network (NIVSN), sponsored by the Brazilian Ministry of Health.

From 2014 to 2016 years the influenza sentinel surveillance in the different states were organized as follows: MT surveillance included respiratory specimens collected in the context of Universal SARI; MS and DF conducted influenza virus surveillance collecting samples from two ILI sentinel units and universal SARI; GO included five ILI sentinel units and universal SARI; RO collected sample from two ILI s and two SARI -ICU sentinel units; PI included three ILI and two SARI-ICU sentinel units; TO collected samples from two ILI and one SARI – ICU sentinel units. Therefore, 21 ILI sentinel units are distributed among the São Paulo metropolitan region; among those, 8 ILI and 9 SARI –ICU sentinel units are located in the city of São Paulo.

A total of five respiratory specimens per week and their respective epidemiological data were collected until five days of the onset of clinical symptoms after obtaining informed consent. For children, the informed consent was provided by the parents or guardians following the Brazilian Ministry of Health protocol and with the approval of the Institute Adolfo Lutz review board.

The samples collected by the sentinel units were preliminarily sent to the regional Central Public Health Laboratory (CPHL), in each state, towards to investigate influenza viruses. Influenza positive samples were sent to the National Influenza Center/Institute Adolfo Lutz (NIC/IAL) for virus isolation attempts in Madin Darby Canine Kidney (MDCK) cell culture, antigenic and genetic analysis. Representative A(H1)pdm09, A(H3) influenza strains, and IBV were sent to the Centers for Diseases Control and Prevention (CDC) according to the WHO/NICs terms of reference for further characterization. Thus, we received confirmation about influenza B antigenic characterization using hemagglutination inhibition (HI) from the CDC. Timely shipments of the most recently circulating viruses provide information to the WHO for the Southern Hemisphere (SH) influenza vaccine composition.

2.2. Methods

We conducted a cross-sectional study analyzing data from the routine viral surveillance, performed using RT-qPCR assay for influenza A(H1N1) pdm09, A(H3N2), and influenza virus B following procedures developed by the CDC [25]. In addition, Influenza B positive samples were submitted to the influenza B lineage genotyping panel developed by the CDC and distributed to the NICs by the International Reagent Resources (IRR) to obtain their characterization as B/Yamagata or Victoria lineages using the One-step RT-qPCR assay.

Ribonucleic acid (RNA) was extracted from 140 µL of respiratory specimens using the QIAamp viral RNA mini-Kit (Qiagen, Hilden, Germany) following the manufacturer's instructions. Sixty µl of eluted RNA were aliquoted and retained at −80 °C until testing. Five µl were used to identify influenza B lineages (Yamagata, Vic-

toria). All respiratory specimens were tested using RT-qPCR for the human RNase P (RP) gene, a constitutive gene that serves as an internal positive control for human nucleic acid to assess specimen quality and extraction [25].

Real - time RT-PCR was performed using an Invitrogen Superscript™ III Platinum™.

One-Step Quantitative kit according to the manufacturer's instructions, on a Roche LightCycler® 480 II instrument4 (Roche, Basel, Switzerland). The sets of primers and probes available in the influenza B lineage genotyping panel were prepared and stored according to the CDC's instructions for use. Briefly, the master mix containing appropriate primers and probe were dispensed into a reaction tube in a volume of 20 μL, and 5 μL of the extracted RNA were added to each well. All runs included positive and negative virus template controls to monitor assay performance. Respiratory specimens are considered of acceptable quality when RP curves cross the threshold line at or before 35 cycles [25].

Amplification was carried out on a Roche LC480 II instrument with the following conditions: reverse transcription 50 °C for 30 min, taq inhibitor activation 95 °C for 10 min, PCR amplification (45 cycles) 95 °C for 15 sec and extension 55 °C for 30 sec.

A test was considered positive when a well-defined fluorescence curve crossed the threshold within 40 cycles [25].

2.3. Statistical analysis

We have estimated the prevalence of H1, H3, and B strains. All analyses were performed with the software Microsoft Excel.

3. Results

Influenza viruses identified during 2014, 2015, and 2016 influenza seasons demonstrate the higher prevalence of H3 strains during two consecutive years (2014 and 2015), as well as the high prevalence of H1 strains during the 2016 influenza season. On the other hand, a lower prevalence of influenza B virus was observed during 2016 influenza season; furthermore, 2016 was the season with the lowest frequency of influenza B virus among the positive tests (576/5937 = 9.7 %) (Table 1).

Among the 342, 197, and 576 influenza B viruses identified during 2014, 2015, and 2016, 197, 190, and 483 were submitted to lineage characterization, respectively.

Of the 197 samples analyzed in 2014, a total of 160 (81 %) corresponded to the B/Yamagata lineage antigenically like to the B/Massachusetts/20/2012; 19 (10 %) corresponded to the B/Victoria lineage antigenically like to the B/Brisbane/60/2008, and 18 (9 %) to indeterminate lineages. Of the 190 samples analyzed in 2015, 124 (65 %) corresponded to the B/Yamagata strain antigenically like the B/Phuket/30/2013; 55 (29 %) matched the B/Victoria lineage antigenically like the B/Brisbane/60/2008 and 11 (6 %) were of indeterminate lineage. Of the 483 samples analyzed in 2016, 297 (62 %) corresponded to the B/Victoria strain antigenically like to the B/Brisbane/60/2008; 174 (36 %) matched the B/Yamagata lineage antigenically like to the B/Phuket/3073/2013 and 12 (2 %) were of indeterminate lineage as shown in Fig. 1.

The influenza B/Yamagata lineage antigenically-like to the B/Massachusetts/02/2012 predominated during the influenza season 2014. This lineage which belongs to genetic group 2, was the influenza B component of the trivalent vaccine for the 2014 influenza season in the South Hemisphere.

As illustrated in Fig. 2 the influenza B/Yamagata lineage was identified in the epidemiological week 6 (EW-6), and it was sporadically detected up to EW-20. An increase in circulation was observed in the EW-36, 37, 38, peaking in the EW-39 and decreasing in the EW-40, 41, 42, and 43.

The influenza B/Victoria lineage antigenically-like to the B/Brisbane /60/2008 virus was the component for the quadrivalent vaccine in the 2014 Southern Hemisphere influenza season.

Table 1
Influenza virological surveillance 2014–2016 seasons.

Year	Samples processed n	H1 n (%)	H3 n (%)	B n (%)	Positive by year n (%)
2014	10,631	284 (2.7)	758 (7.1)	342 (3.2)	1384 (13.0)
2015	7215	56 (0.8)	428 (5.9)	197 (2.7)	681 (9.4)
2016	24,135	5306 (22.0)	55 (0.2)	576 (2.4)	5937 (24.6)

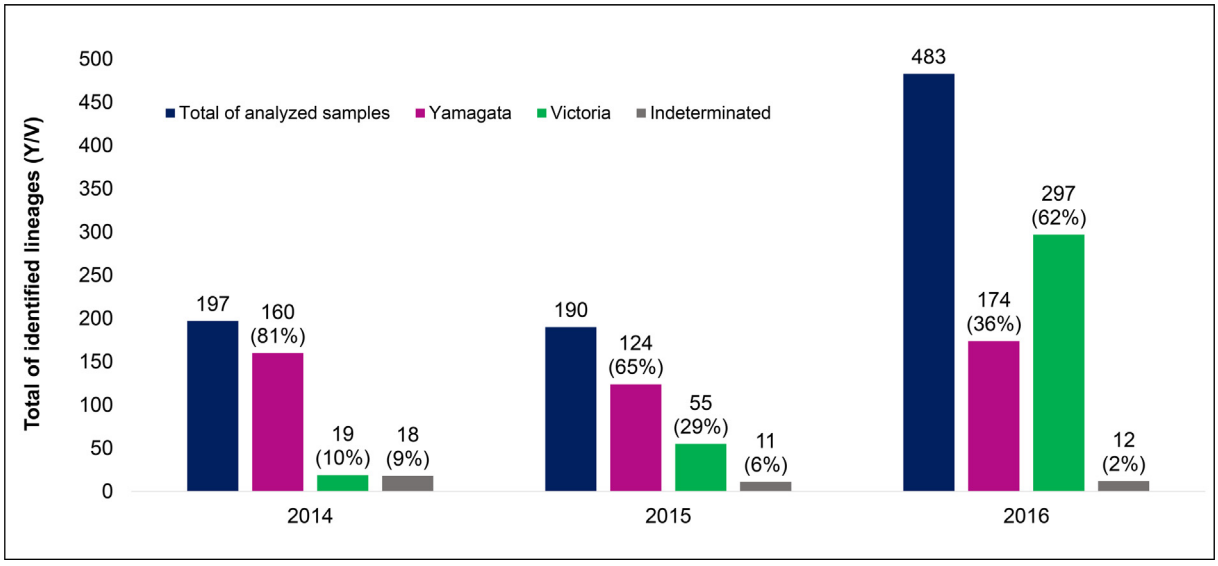

Fig. 1. Distribution of 870 influenza B viruses by lineages (Yamagata and Victoria) by years.

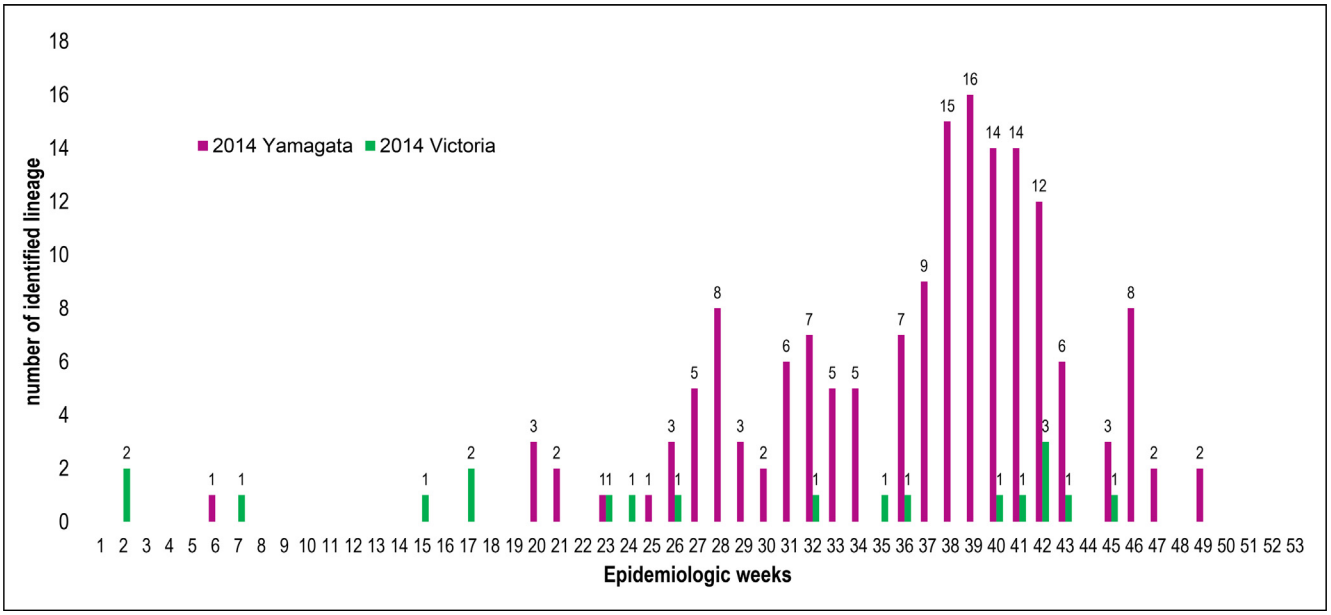

Fig. 2. Seasonality of influenza B/lineages (Yamagata, Victoria) during influenza season 2014.

As illustrated in Fig. 3 the influenza B/Yamagata lineage was the predominant circulating virus during the 2015 influenza season, although the B/Yamagata lineage, belonging to genetic group 3, antigenically related to the B/Phuket /3073/2013, predominated and was the influenza B component of trivalent vaccines for the 2015 Southern Hemisphere influenza season.

An increase in B/Yamagata circulation was observed in the EW-42, peaking in the EW-43, 44, 45 and decreasing in the EW- 46, after a peak in the EW-47, following a decrease in circulation from EW-48. Its peak of circulation was later than the one observed in the 2014 influenza season, as shown in Fig. 2. Fig. 3.

The WHO recommended the Influenza B/Brisbane/60/2008 circulating virus for the composition of the quadrivalent vaccine for the 2015 Southern Hemisphere influenza season.

Regarding the seasonality of the influenza B /Victoria lineage, an increase in circulation was observed from EW-48 to EW-53, concomitantly with the decrease of the B/Yamagata lineage during the 2015 influenza season (Fig. 3). The Influenza B Victoria lineage was antigenically similar to the B/Brisbane/60/2008 predominating during the 2016 influenza season, as shown in (Fig. 4). The WHO recommended this lineage as the component of the trivalent vaccine for the 2016 Southern Hemisphere influenza season. Its circulation was observed all year round, peaking in the EW-13, EW-14, EW-41, and EW-49.

The Influenza B/Yamagata lineage antigenically like B/Phuket/3073/2013 circulated during the 2016 influenza season and was the WHO-recommended B component for the quadrivalent vaccine for the Southern Hemisphere influenza seasons.

Fig. 3. Seasonality of influenza B/lineages (Yamagata, Victoria) during influenza season 2015.

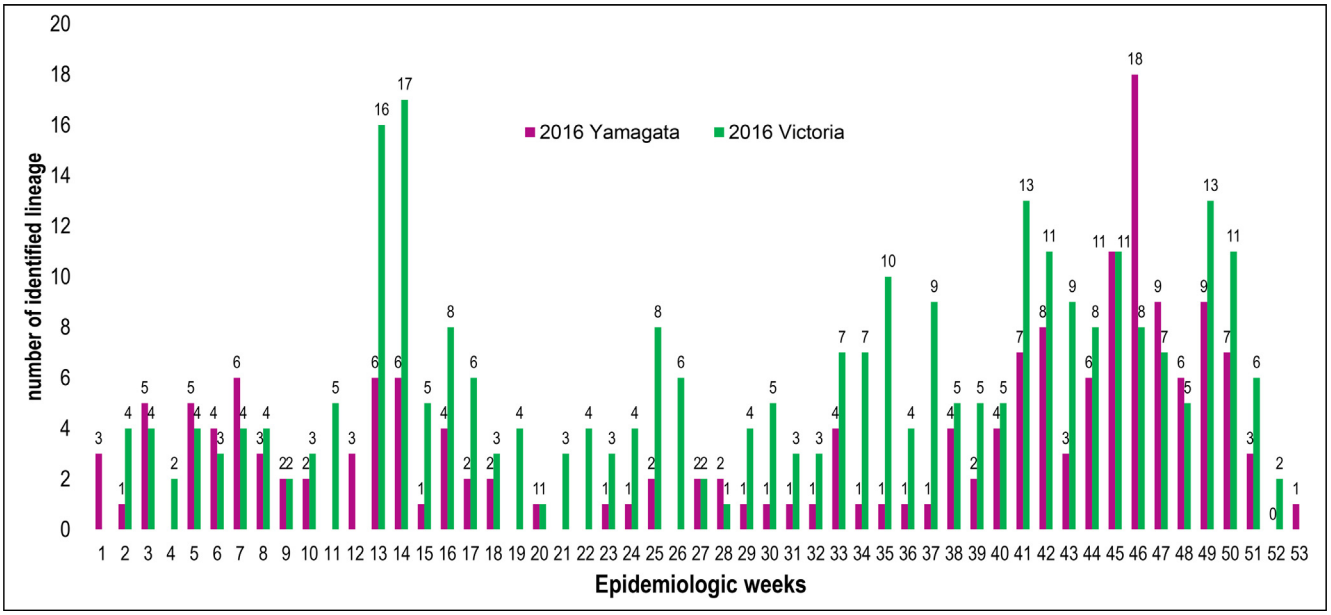

Fig. 4. Seasonality of influenza B/lineages (Yamagata, Victoria) during influenza season 2016.

This study revealed changes in the circulation pattern of the influenza B virus during the 2016 influenza season, that is, one peak earlier in the EW-13 and EW-14, corresponding to the B/Victoria lineage, and another in EW- 46, corresponding to the Yamagata lineage, as shown in Fig. 4.

The present study revealed the co-circulation of both IBV lineages year-round during the 2014, 2015, and 2016 influenza seasons, as illustrated in Figs. 2, 3, and 4, respectively.

4. Discussion

Our results demonstrate the match between IBV lineages preconized by the WHO for the trivalent vaccine composition used in the Southern Hemisphere and circulating viruses during the 2014, 2015, and 2016 seasons.

Our findings clearly demonstrated the co-circulation of both lineages in children up to 5 years of age. This age group is more susceptible to IBV infections presenting high hospitalization rates mainly in children ≤2 years of age [26–28]. Due to the impact of infection by the IBV in this age group, it would be advised to prioritize the quadrivalent vaccine, containing both lineages, to promote better effectiveness of the vaccination campaign in the face of trivalent mismatch outcomes [22,23]. Data obtained from the South region of Brazil demonstrate the occurrence of SARI because of IBV infection, mainly in infants ≤1 year of age and patients ≥60 yo [23].

Previous studies showed IBV infection prevalence in school children and in adolescents: (5–14 yo) [29,30,32]; (5–17 yo) [31] and (13–18 yo) [33].

Our findings revealed IBV infection in the age groups of school children, adolescents, and young adults. The relevant co-circulation of both lineages among 6–59 years age groups, not included in the vaccine campaign schedule, raises concerns about the real impact of circulating lineages in different age groups Fig. 5. This scenario emphasizes the crucial role of virologic surveillance throughout the year.

The present study also showed the IBV prevalence in the adolescent group, which is an important target for vaccination because of the high susceptibility of these individuals regardless of the predominant lineage. In addition, although elderly persons (≥60 years old) are already included in vaccination campaign schedules worldwide, the availability of a quadrivalent vaccine for this population can improve their quality of life, considering the risk of complications related to infections by the IBV and the presence of comorbidities.

Our findings revealed that, apart 2016 influenza season, infections by the influenza B virus were laboratory-confirmed in 24.7 % (2014) and 28.9 % (2015) of the positive samples received from sentinel units in the context of the National Influenza Surveillance Network similar to findings from studies conducted in countries located in both hemispheres (Northern and Southern) [33–42].

One limitation of our study was the inability to retrieve respiratory samples from the beginning of 2014 and to provide information on the severity of the disease resulting from infection by the distinct IBV lineages. On the other hand, our results highlight this information's importance in guiding further investigations and the low representativeness of respiratory samples from some Brazilian states except for São Paulo. Furthermore, at least one B/lineage was identified in different Brazilian states, except for MS, PI and TO, during the 2014 influenza season, and GO and MS during the 2016 influenza season (Table 2).

The differentiation of Influenza B lineages is routinely carried out using the HI assay, which requires virus isolation in cell culture and serological tests, both time-consuming. A rapid assay as RT-qPCR contributes to understanding how the prevalence of both lineages varies in different geographical regions of the world and provides information that could guide vaccine strain selection. However, it does not replace the HI test in selecting viruses for the influenza virus vaccine composition. Nowadays, there are five genetically and antigenically distinct IBV co-circulating: B/Yamagata, B/Victoria V1A, B/Victoria V1A – 2 DEL, and two groups of B/Victoria V1A – 3DE L [18,22]. May IBV lineage Victoria predominates in the future? Victoria lineage variants will increase the severity of the disease in younger age groups and in the general population? Recently evolutionary pattern of Victoria lineage reinforces the mission of the Global Influenza Surveillance Response

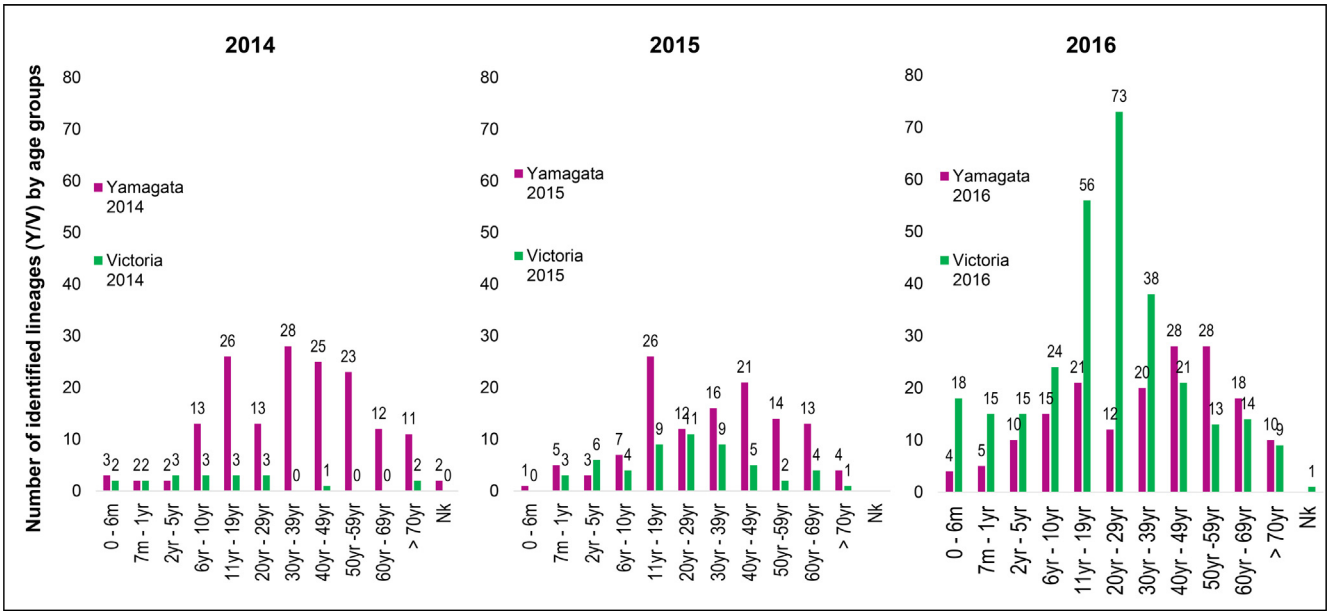

Fig. 5. Distribution of influenza B virus lineages (Yamagata and Victoria) by age groups during 2014, 2015 and 2016 influenza seasons.

Table 2
Distribution of influenza B lineages (Yamagata and Victoria) in selected Brazilian states according to year, 2014–2016.

UF	2014				2015				2016			
	Yamagata	Victoria	Indetermined	Total	Yamagata	Victoria	Indetermined	Total	Yamagata	Victoria	Indetermined	Total
DF	1	1	1	3	1	2	0	3	0	1	0	1
GO	2	3	0	5	8	1	0	9	0	0	0	0
MS	0	0	0	0	2	0	0	2	0	0	0	0
MT	0	5	2	7	2	0	2	4	1	4	1	6
PI	0	0	0	0	1	1	0	2	0	8	0	8
RO	0	1	0	1	5	0	0	5	1	1	0	2
SP	157	9	15	181	93	51	9	153	170	283	11	464
TO	0	0	0	0	12	0	0	12	2	0	0	2
Total	160	19	18	197	124	55	11	190	174	297	12	483

Systems (GISRS) towards influenza virus strains selection aiming prevention strategies by vaccination campaigns. This scenario urges a rapid and specific identification of the circulating lineage using molecular assays.

5. Conclusion

The differentiation of lineages of influenza B viruses by RT-qPCR accelerates the identification of the most prevalent circulating lineages; these results have direct implications in the lineage selection for vaccine composition. Nowadays, more attention is required regarding IBV surveillance facing the Victoria lineage evolutionary pattern from 2016 and the co-circulation of both lineages (Victoria and Yamagata) worldwide since 2002.

The virological surveillance demonstrated the match between influenza B virus lineages preconized by the WHO for the trivalent vaccine composition used in the Southern Hemisphere influenza season and the predominate circulating lineages during the 2014, 2015, and 2016 seasons.

The relevant prevalence of influenza B virus in the age range 6–59 years, not currently included in the vaccine campaign schedule, highlights the need to review vaccine policies.

Considering the co-circulation of both influenza B lineages (Yamagata, Victoria) during the 2014–2016 influenza seasons, a quadrivalent vaccine would be advisable.

Data availability

Data will be made available on request.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

Our thanks to Nilva Matias dos Reis, Ana Lúcia de Carvalho Avelino, Lincoln Spinazzola do Prado, Pamela Arrebola de Sousa, Renato de Sousa Paulino for technical support; Respiratory Diseases Center/ Virology Center/ Instituto Adolfo Lutz/SP/Brazil; Sentinel Units Health Professionals; Municipal and State Secretariat of Health of São Paulo, SP, Brazil; Center for Epidemiological Surveillance "Prof. Alexandre Vranjac" São Paulo/SP; Central Public Health Laboratory of the states of São Paulo, Mato Grosso, Mato Grosso do Sul, Goiás, Piauí, Tocantins, Rondônia and Distrito Federal; Ministry of Health of Brazil, Pan American Health Organization, Washington-DC, USA; Centers for Disease Control and Prevention, Atlanta, GA, USA; World Health Organization – Genève – Switzerland.

Authors contributions

Conceived and designed the experiment: DBBdS, KCOS, MAB, TMd.P. Performed the experiments: DBBdS, KCOS, MAB. Analyzed the data: DBBdS, KCOS, MAB, TMdP. Wrote the paper; TMdP.

Funding

Collaborative study sponsored by Institute Adolfo Lutz and funded by Sanofi Pasteur.

References

[1] World Health Organization. Global influenza strategy 2019–2030. World Health Organization; 2019. <https://apps.who.int/iris/handle/10665/311184>. License: CC BY-NC-SA 3.0 IGO, 31p – ISBN 978924151520/en/.

[2] Paget J, Spreeuwenberg P, Charu V, Taylor RJ, Iuliano AD, Bresee J, et al. Global seasonal influenza-associated mortality collaborator network and GLaMOR collaborating teams*. Global mortality associated with seasonal influenza epidemics: new burden estimates and predictors from the GLaMOR Project. J Glob Health 2019;9(2):020421. https://doi.org/10.7189/jogh.09.020421. PMID: 31673337; PMCID: PMC6815659.

[3] Virus Taxonomy: 2019 Release. EC 51, Berlin, Germany, July 2019. E mail ratification March 2020 (MSL#35). Orthomyxoviridae: 7 genera, 9 species.

[4] New species. <http://talk.ictvonline.orgICTV/proposal/2014.009a-dM.A.v2>. InfluenzavirusD.pdf; 2018 influenza D, <http://www.science daily.com/releases/2016/09/1609011405433.htm>. [accessed 03 March 2017] [accessed 13 august 2018].

[5] Su S, Fu X, Li G, Kerlin F, Veit M. Novel Influenza D virus: epidemiology, pathology, evolution and biological characteristics. Virulence 2017;8(8):1580–91. https://doi.org/10.1080/21505594.2017.1365216.

[6] Kuchipudi Suresh V, Nissly Ruth H. Novel flu viruses in bats and cattle: pushing the envelope. Vet Sci 2018;5(3):71. https://doi.org/10.3390/vetsci5030071.

[7] Parrish CR, Murcia PR, Holmes EC, Schultz-Cherry S. Influenza virus reservoirs and intermediate hosts: dogs, horses, and new possibilities for influenza virus exposure of humans. J Virol 2015;89(6):2990–4.

[8] Osterhaus AD, Rimmelzwaan GF, Martina BE, Bestebroer TM, Fouchier RA. Influenza B virus in seals. Science 2000;288:1051–3. https://doi.org/10.1126/science.288.5468.1051.

[9] Bodewes R, Morick D, Mutsert G, Osinga N, Bestebroer T, van der Vliet S, et al. Recurring influenza B virus infections in Seals. Emerg Infect Dis 2013;19(3):511–2. https://doi.org/10.3201/eid1903.120965. PMID: 23750359.

[10] Rota PA, Wallis TR, Harmon MW, Rota JS, Kendal AP, Nerome K. Cocirculation of two distinct evolutionary lineages of influenza type B virus since 1983. Virology 1990;175(1):59–68.

[11] Hampson A. Emergence of influenza B/Hong Kong/330/2001-like strains. Victorian Infect Dis Bull 2002;5:22–3.

[12] Paiva TM, Ishuida MA, Maria Gisele Gonçalez MG, Benega MA, Sousa MCO, Cruz AS. Occurrence of influenza/Hong Kong – like strains in São Paulo, Brazil during 2002. Rev Inst Med trop S Paulo 2003;45(1):51–2.

[13] Paiva TM, Benega MA, Silva DB, Santos KC, Cruz AS, Hortenci MF, et al. Evolutionary pattern of the reemerging influenza B/Victoria lineage viruses in São Paulo, Brazil, 1996–2012: implications for vaccine compositions strategy. J Med Virol 2013;85:1983–9.

[14] Ambrose CS, Levin MJ. The rationale for quadrivalent influenza vaccines PMID: 22252006. Hum Vaccin Immunother 2012;8(1):81–8. htpps://doi.org/10.4161/hv.8.1.17623.

[15] Tisa V et al. Quadrivalent influenza vaccine: a new opportunity to reduce the influenza burden. J Prev Med Hyg 2016;57:E28–33.

[16] Blanton L, Alabi N, Mustaquin D, Taylor C, Kniss K, Kramer N, et al. Update influenza activity in the United States during the 20–2017 season and composition on the 20177–2018 influenza vaccine. MMWR Morb Mortal Wkly Rep 2017;66(25):668–76. https://doi.org/10.15585/mmwr.mm6625a3. PMID 28662019.

[17] World Health Organization (WHO). Recommended composition of influenza virus vaccines for use in the 2018–2019 northern hemisphere influenza season. Geneva: WHO; 2018. <https://www.who.int/influenza/vaccines/virus/recommendations/201802_recommendations.pdf>.

[18] Virk RK, Jayakumar J, Mendenhall IH, Moorthy M, Lam P, Linster M, et al. Divergent evolutionary trajectories of influenza B viruses underlie their contemporaneous epidemic activity. Proc Natl Acad Sci USA 2020;117(1):619–28.

[19] Sederdahl BK, Williams JV. Epidemiology and clinical characteristics of influenza C virus. Viruses 2020;12(1):89.

[20] Shuo Su, Xinliang Fu, Li G, Kerling F, Veit M. Influenza D virus: epidemiology, pathology, evolution and biological characteristics. Virulence 2017;8(8):1580–91. Https://doi.org/10.180/21505594.2017.1365216.

[21] Falleiros Arlant LH, Bricks LF. Influenza B burden in latin america and potential benefits of the new quadrivalent vaccines. J Pediatric Infect Dis Soc 2016;5(1):1–2. https://doi.org/10.1093/jpids/piv107. Epub 2016 Jan 23 PMID: 26803330.

[22] Bhat YR. Influenza B infection in children: a review. World J Clin Pediatr 2020;9(3):42–52. https://doi.org/10.5409/wjcp.v9.i3.44.

[23] Duarte MB, Gregianini TS, Martins LG, Veiga ABG. Epidemiology of influenza B infection in the state of Rio Grande do Sul, Brazil, from 2003 to 2019. J Med Virol 2021;93(8):4756–62. https://doi.org/10.1002/jmv.26822. Epub 2021 Feb 1 PMID: 33501655.

[24] Caini S, Huang QS, Ciblak MA, et al. Epidemiological and virological characteristics of Influenza B: results of the Global Influenza B Study. Influenza Other Resp Viruses 2015;9(Suppl. 1):3–12.

[25] Center for Disease Control and Prevention – CDC. CDC Protocol for realtime RTPCR for influenza A (H1N1) [documento na internet] <http://www.who.int/csr/resources/publications/swineflu/CDCRealtimeRTPCR_SwineH1Assay-2009_20090430.pdf>.

[26] Eşki A, Öztürk GK, Gülen F, Çiçek C, Demir E. Risk factors for influenza virus related severe lower respiratory tract infection in children. Pediatr Infect Dis J 2019;38:1090–5. https://doi.org/10.1097/INF0000000000002447. PMID: 31469782.

[27] Paul Glezen W. Editorial commentary: changing epidemiology of influenza B virus. Clin Infect Dis 2014;59(11):1525–6.

[28] Barr IG, VijayKrishna D, Sullivan SG. Differential age susceptibility to influenza B/Victoria lineages viruses in the 2015 Australian influenza season pii=30118. Euro Surveill 2016;21(4). https://doi.org/10.2807/1560-7917.ES.2016.21.4.30118.

[29] Rivas MJ, Alegretti M, Coppola L, et al. Epidemiology and genetic variability of circulating influenza B viruses in Uruguai, 2012–2019. Microorganisms 2020;8:591. https://doi.org/10.3390/microorganisms.

[30] Beauté J, Zucs P, Korsun N, Bragstad K, Enouf V, Kossyvakis A, et al. Age-specific differences in influenza virus type and subtype distribution in the 2012/2013 season in 12 European countries. Epidemiol Infect 2015;143(14):2950–8.

[31] Olson DR, Heffernan RT, Paladini M, Konty K, Weiss D, Mostashari F, et al. Monitoring the impact of influenza by age: emergency department fever and respiratory complaint surveillance in New York City. PLoS Med 2007;4(8):e247.

[32] Perosa AN, Granato C, Bellei N. Detection of influenza B lineages from 2001 to 2013 in a tertiary hospital in the city of São Paulo. Mem Inst Oswaldo Cruz 110(5):606–10. doi:10.590/0074-02760150044.

[33] Paul Glesen W, Scmier JK, Kuehn CM, Ryan KJ, Oxford J. The burden of influenza B: a structures literature review. Am J Public Health 2013;103(3):e43–5. https://doi.org/10.2105/AJPH.2012.301137. PMID: 23327249.

[34] Barros EN, Cintia O, Rosseto E, Freitas L, Colindres R. Patterns of influenza B circulation in Brazil and its relevance to seasonal vaccine composition. Bras J Infect Dis 2016;20(1):81–90. https://doi.org/10.1016/j.bjid.2015.09.009. PMID:26626166.

[35] Palekar R, Rodriguez A, Avila C, Barrera G, Barrear M, Brenes H, et al. Patterns of influenza B circulation in Latin America and the Caribbean, 2010–2017. PLoS ONE 2019;14(8). https://doi.org/10.1371/journal.pone.0219595.

[36] Caini S, Kusznierz G, Garate W, Wangchuk S, Thapa B, de Paula Júnior FJ, et al. The epidemiological signature of influenza B virus and its B/Victoria and B/Yamagata lineages in the 21st century. PloS ONE 2019;14(9). https://doi.org/10.1371/journal.pone.0222381.

[37] Jennings L, Huang QS, Barr I, Lee PI, Kim WJ, Buchy P, et al. Literature review of the epidemiology of influenza B disease in 15 countries in the SIA Pacific region. Influenza Other Resp Viruses 2018;12(3):383–411. https://doi.org/10.1111/irv.12522. PMDI 29127742.

[38] Seleka M, Treurnicht FK, Tempias S, Hellferscee O, Mtshali S, Al Cohen, et al. Epidemiology of influenza B/Yamagata and Victoria line GES in South Africa 2005–2014. PLoS One 2017;12(5):e0177655. https://doi.org/10.1371/journal.pone.0177655. PMID 28542324.

[39] Moa AM, Muscatello DJ, Turner RM, MacIntyre CR. Epidemiology of influenza B in Australia: 2001-2014 influenza seasons. Influenza Other Respi Virus 2017;11(2):102–9.

[40] Mosnier A, Caini S, Daviaud I, Jl Bensoussan, Stoll-Keller F, Bui TT, et al. Ten influenza season in France: distribution and timing of influenza A and B circulation, 2003–2013. BMC Infect Dis 2019;15:357. https://doi.org/10.1186/s12879-015-1056-z. PMDI: 26289794.

[41] Tan J, Asthagiri-Arunkumar G, Krammer F. Universal influenza virus vaccines and therapeutics: Where do we stand with influenza B virus? Curr Opin Immunol 2018;53:45–50. https://doi.org/10.1016/j.coi.2018.04.002.

[42] Caini S, El-Guerche Séblain C, Ciblak MA, Paget J. Epidemiology of seasonal influenza in the Middle East and North Africa regions, 2010–2016: circulating influenza A and B viruses and spatial timing of epidemics. Influenza Other Resp Viruses 2018;12(3):344–52.