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Flu is not always benign: 2018/19 epidemic

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FLU IS NOT ALWAYS BENIGN: 2018/19 EPIDEMIC

Artigo Científico

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Abbreviations

To facilitate the reading of this paper we present a list of abbreviations that were used throughout the text:

AOM - Acute otitis media
CHUC – Centro Hospitalar e Universitário de Coimbra
CRP - C reactive protein
DGS - Directorate General of Health
ES - Emergency service
FMUC - Faculty of Medicine of the University of Coimbra
ICU – Intensive Care Unit
LOS – Length of stay
PCR - Polymerase chain reaction
WHO - World Health Organization

Resumo

Introdução: A gripe é uma doença muito frequente, habitualmente considerada como uma condição benigna e autolimitada em crianças saudáveis. Contudo, estão descritos quadros graves, atingindo também crianças previamente saudáveis. O objetivo deste estudo é caracterizar a doença em crianças com e sem fatores de risco para esta infeção na epidemia de 2018/19.

Métodos: Um estudo observacional retrospetivo foi conduzido em 90 crianças com idades compreendidas entre os 0 e 18 anos, com infeção pelo vírus influenza confirmada por PCR multiplex durante a epidemia de 2018/19, no Hospital Pediátrico do Centro Hospitalar e Universitário de Coimbra (CHUC). Foram analisadas as manifestações clínicas, exames complementares de diagnóstico efetuados, tratamentos instituídos, evolução e estado vacinal dos doentes. Foram ainda analisadas separadamente crianças previamente saudáveis e crianças com fatores de risco para infeção por este vírus.

Resultados: O vírus Influenza A foi largamente predominante (98,9%). A maioria dos casos ocorreu em crianças com idade inferior a 5 anos (idade média 3,78 ± 3,99 anos), e que tinham menos fatores de risco (p=0.001) quando comparadas com as de idade superior. Em 51% das crianças foram observadas complicações e 32% foram internadas, constatando-se uma maior representatividade de crianças saudáveis nestes grupos. As crianças imunocompetentes apresentaram uma prevalência significativamente superior de complicações e internamento (p= 0,027 e p=0,047, respetivamente). Observámos que com o aumento da idade a cada ano, o odds ratio para internamento decrescia em 16% (p=0.041). Foi observada uma maior probabilidade da toma de oseltamivir em crianças com fatores de risco e nas internadas (ambos com p<0.001). Apenas 20% das crianças com recomendação para vacinação estavam efetivamente vacinadas.

Conclusão: As crianças do grupo etário mais jovem são as mais vulneráveis à infeção pelo vírus influenza, demonstrando taxas de complicações e internamentos superiores às das crianças mais velhas. O impacto desta doença foi observado não só nas crianças com fatores de risco, mas também nas previamente saudáveis. A taxa de vacinação das crianças pertencentes a grupos de risco foi muito baixa.

Palavras-chave: Influenza; crianças; vacinação; complicações; internamentos; mortalidade.

Abstract

Background: Influenza virus infection is a very common disease, that is usually seen as a benign and self-limited condition among healthy children. However, severe cases are described, affecting also previously healthy children. The aim of the study is to characterize the infection in children with and without risk factors for influenza virus infection, from the 2018/19 epidemics.

Methods: A retrospective observational study was conducted on 90 children aged between 0-18 years old with influenza virus infection confirmed by PCR multiplex in the 2018/19 influenza season, of the Pediatric Hospital of Centro Hospitalar e Universitário de Coimbra (CHUC). Clinical features, complementary investigation and conducted treatments, the course of the disease and the vaccination status were analyzed. We further analyzed previously healthy children and children with risk factors for influenza infection.

Results: Influenza A was found in almost all cases (98,9%). Most cases occurred in children under 5 years old (mean age of 3,78 ± 3,99 years) and in children with less risk factors (p=0.001), while comparing to the oldest children. Complications were found in 51% of the children and hospitalizations in 32%, both counting with a majority of previously healthy children. Complications and hospitalization were significantly associated to immunocompetent children (p= 0,027 and p=0,047, respectively). We observed that for every 1-year increase in age, the odds ratio for admission decreases 16% (p=0.041). The intake of oseltamivir was more likely in children with risk factors and in hospitalized children (both p<0.001). Only 20% of the children to whom the vaccine was recommended and funded have received it.

Conclusion: Younger children are particularly vulnerable to influenza infection, showing higher rates of complications and hospitalization than the older ones. An increased burden of the disease was seen not only in children with comorbidities, as expected, but also in previously healthy children. The vaccination coverage rate from the children representing risk factors was extremely low.

Keywords: Influenza; children; vaccination; complication; hospitalization; mortality.

Background

Influenza virus infection is a very common disease worldwide that affects one billion people per year globally, from which 90 million children are younger than 5 years old [1]. According to the World Health Organization (WHO), there are 3 to 5 million cases of severe illness associated with annual epidemics of influenza and around 250.000 to 500.000 deaths[2].

This disease is usually seen as a benign and self-limited condition among healthy children, presenting with the sudden onset of fever, rhinorrhea and cough, and less commonly with sore throat, headache, myalgia and gastrointestinal symptoms, which last 4-7 days. With symptomatic treatment and fluid hydration, most children are expected to have a spontaneous recovery in a few days. Nonetheless, more severe outcomes are seen mainly in children aged less than 5 years old and in children with underlying conditions, such as immunosuppression or chronic diseases (asthma, congenital heart disease, hematologic, neurologic or metabolic disorders)[2,3]. These children have greater rates of complications and hospitalizations, the highest being in those younger than 6 months[4]. Pneumonia, acute otitis media, respiratory failure, febrile seizures and encephalitis are among the most common complications[1,5].

Laboratory confirmation of influenza infection can be done through the detection of the virus in a biological sample of the respiratory tract, usually using a multiplex polymerase chain reaction (PCR)[6].

The use of antiviral therapy with oseltamivir is mainly recommended in hospitalized children, children with severe progressive illness or underlying comorbidities[7]. The aims of this treatment are decreasing symptoms' severity, shortening illness duration and preventing complications and death. An early recognition of infected children is important for the prompt use of antiviral treatment in the first 48 hours of illness onset, since it shows higher benefits than later treatment[8,7]. Although treatment of influenza virus infection is available, evidences of its effectiveness are mainly seen when the treatment is given within 48h of illness onset. However, the time since the symptoms start to the time children are brought to the emergency service is frequently longer[8,9]. This treatment also showed a small benefit in the pediatric population[10].

The best way to prevent and reduce the morbidity and mortality associated with this virus is trough vaccination. The strains included in the vaccine change according to the

influenza season and are dictated by WHO, based on global epidemiologic and virologic surveillance[2]. The effectiveness of influenza vaccine is variable, depending on the match between the circulating strains and the strains in the vaccine. A multicentric European project has been developed since 2008, monitoring vaccine effectiveness during influenza seasons and pandemics[6].
According to the Portuguese Directorate-General of Health (DGS), annual influenza immunization is highly recommended and state-provided to children older than 6 months, carrying a high risk condition or hospitalized/living in a healthcare institution for a long length of time[11].

Influenza infection in children is also a burden for the society, since the consequences of the disease go beyond the medical outcomes mentioned above. Because of their high infectiousness, children play a pivotal role in the transmission of influenza to close contacts and communities, contributing to school absenteeism, parental workdays lost and a socioeconomic burden as a result[2].

After the influenza H1N1 2009 pandemic, eleven European-Union countries started monitoring severe cases of acute respiratory disease. In the influenza season of 2011-12, a pilot study to detect severe cases of influenza infection admitted at the Intensive Care Unit (ICU) of secondary and tertiary hospitals, comprising all national districts, was started in Portugal. Every ICU has to notify all laboratory confirmed influenza virus infections to DGS. This study has been done every influenza season since then[6]. However, these data are only from ICU populations, excluding hospitalizations in other units. In this study the pediatric population was underrepresented, being half of the patients older than 65 years old.
In order to try to extend the influenza surveillance trough other units and to a different population, a new study was developed in the past season, reporting laboratory confirmed influenza cases of a pediatric ward, a pneumology ward and an intermediate care unit. However, because of the small sample size, no conclusions were taken.

In the last influenza season (2018-19), in the Pediatric Hospital of Centro Hospitalar e Universitário de Coimbra (CHUC), we observed several complicated influenza infections, leading to a high number of hospitalizations. This, along with the lack of detailed information on influenza infection in Portuguese children, triggered the interest on studying the 2018-19 influenza season. Therefore, the aim of the study is to characterize the infection in the pediatric population, namely to evaluate the clinical presentation,

presence of risk factors, rate of vaccination, antiviral and antibiotics use, and severity of the disease in healthy children and in children with risk factors for this infection.

Methods

Study design and oversight

A retrospective observational study was performed at the Pediatric Hospital of CHUC, a tertiary hospital that serves the central region of Portugal, in collaboration with the Faculty of Medicine of the University of Coimbra (FMUC), Coimbra, Portugal.

Study Population

The Paediatric Hospital admits children below 18 years of age, mainly from the city but also from the district. The municipality of Coimbra has no other 24-hour primary care service and there is no other paediatric Emergency Service (ES) in the city or surrounding area (≈50km). The ES looks after both ambulatory cases discharged home and short stay admissions for up to 48 hours. Approximately 63,000 children and adolescent are seen annually, some referred from other hospitals or general practitioners, but the majority brought directly by their parents. Care is provided free of charge and no referral is required. It has an Intensive Care Unit.

All the medical records of children aged between 0-18 years old observed or admitted to the Pediatric Hospital-CHUC, in the influenza season period from the 1st of December 2018 to the 30th of April 2019, with influenza virus infection confirmed by PCR multiplex performed in nasopharyngeal secretions samples, were analyzed. The laboratory test was requested based on the physicians' decision.

The study population was divided into two main groups: healthy children (n=55) and children with risk factors for influenza infection (n=35). The last group includes all children with chronic diseases (diabetes mellitus, renal insufficiency), immunosuppression (undergoing chemotherapy and transplanted patients), genetic conditions (21 trisomy, cystic fibrosis, alpha-1 antitrypsin deficiency), long-term hospitalization or residents in child-care centers, as defined by DGS[11].

Data Collection

An anonymized database was created in collaboration with a pediatrician. The following information was recorded: clinical symptoms and signs at presentation, investigation

preformed, complications, use of antiviral and antimicrobial agents, need of hospitalization and length of stay. Children´s medical history and comorbidities, as well as their influenza vaccination status were also registered.

Clinical symptoms and signs

The presence of fever, cough, rhinorrhea, myalgia, diarrhea and altered consciousness were registered. The altered level of consciousness was based on what was registered in the notes, that might have some variation between clinicians observing the child. This definition included: appearance, level of alertness and response to stimuli and speech, noting that these actions are child and age specific.

Chest X-ray and laboratory Investigations

Chest X-rays were analyzed for the presence of interstitial infiltrate and consolidation. From the laboratory tests, the following information was considered: leucocyte count and C reactive protein (CRP). Influenza detection, typing and subtyping, and identification of other viruses and bacteria in respiratory secretions by PCR were registered as well as blood culture results. The PCR multiplex in respiratory secretions test includes the detection of: Influenza virus A H1 pdm09, Influenza virus A H3, Influenza virus B, Parainfluenza 1, Parainfluenza 2, Parainfluenza 3, Parainfluenza 4, Adenovirus, Syncytial respiratory virus, Metapneumovirus, Rhinovirus/Enterovirus, Coronavirus HKU1, Coronavirus 229E, Coronavirus OC43, Coronavirus NL63, Coronavirus MERS-CoV, *Mycoplasma pneumoniae*, *Bordetella pertussis*, *Bordetella parapertussis* (Biomerieux, France).

Antiviral therapy

The use of oseltamivir is usually based on an individualized clinical decision taken by the physician observing the child. The main recommendations are in high risk patients, to prevent/minimize severe disease progression and patients showing severe signs and symptoms or a worsening clinical status. In Portugal, the DGS publishes guidelines for the use of antivirals that include: all patients with progressive disease or clinical severity, immunosuppressed, with severe or decompensated chronic disease, with morbid obesity, hospitalized or with long term salicylate treatment. Those were considered for this study[7].

Vaccination

According to the DGS recommendations, all children above 6 months of age with a chronic disease (cardiologic, pulmonary, renal, hepatic, hematological, metabolic, neuromuscular or immune), immunosuppression status or hospitalized/living in a healthcare institution for a long length of time[11], were considered a risk group with indication to receive influenza virus vaccine.

Complications

The following complications were registered: acute otitis media (AOM), pneumonia, encephalitis, sepsis, toxic shock, bacteremia, hypoxemia, febrile seizures, decompensation of underlying disease, hypoglycemia, and myositis.

Statistical Analysis

Qualitative data was handled by means of observed absolute (n) and relative (%) frequencies, while quantitative data was presented either by its mean ± standard deviation or median and interquartile range, according to the observed distribution of the variables (normal or other, respectively).

In order to assess the association between qualitative variables, we applied the Chi-square test or Fisher's exact test (in case there was one cell with less than 5 observations) or univariate and multivariate logistic regression whenever interactions should be evaluated.
The association of qualitative variables with continuous outcomes was studied using a two-sample t-test or Wilcoxon rank sum test for parametric or nonparametric distributions, respectively.
Statistical analysis was performed with Stata® 15 (StataCorp, College Station, TX) and was interpreted at a 5% significance level.

Ethical approval

The study was submitted to the Innovation and Development Unit of CHUC.

Results

General description of the study group

Study Population

Of the 90 children with influenza virus infection confirmed by PCR, there were 41 girls (45,56%) and 49 boys (54,44%), with a mean age of 3,78 ± 3,99 years old (range one month - 18 years old). Figure 1 shows the age distribution. The majority (68/90; 75,55%) were younger than five (including 10 children younger than six months) and 22 (24,44%) were ≥ 5 years old.

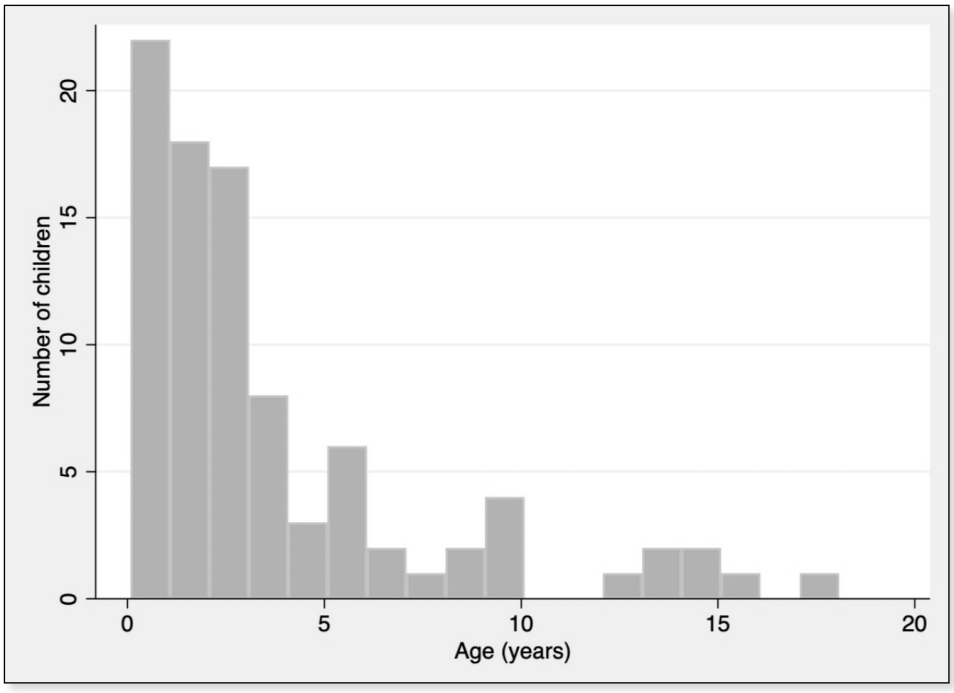

Figure 1. Age distribution of influenza infection cases in the 2018-19 epidemic.

The majority of the admissions occurred in January (n=47; 52,2%), followed by February (n=29; 32,2%) and December (n=14; 15,5%), with no admissions in March.

Regarding the patients' origin, 71 (78,9%) were brought to the Emergency Service (ES) by the parents from home, 13 (14,4%) were from the outpatient clinic or day care hospital, 4 were already hospitalized and 2 were send to the ES by a primary care unit.

Healthy and at-risk children

There were 55 (61%) previously healthy children, while 35 (39%) had at least one risk factor. Risk factors were divided into the following categories: asthma with underlying treatment, oncologic disease, nephrological disorders, cardiopathy, metabolic disease, others.

Clinical manifestations

Overall, fever was the most common sign at presentation, followed by cough, rhinorrhea, diarrhea, altered level of consciousness and myalgia (Figure 2).

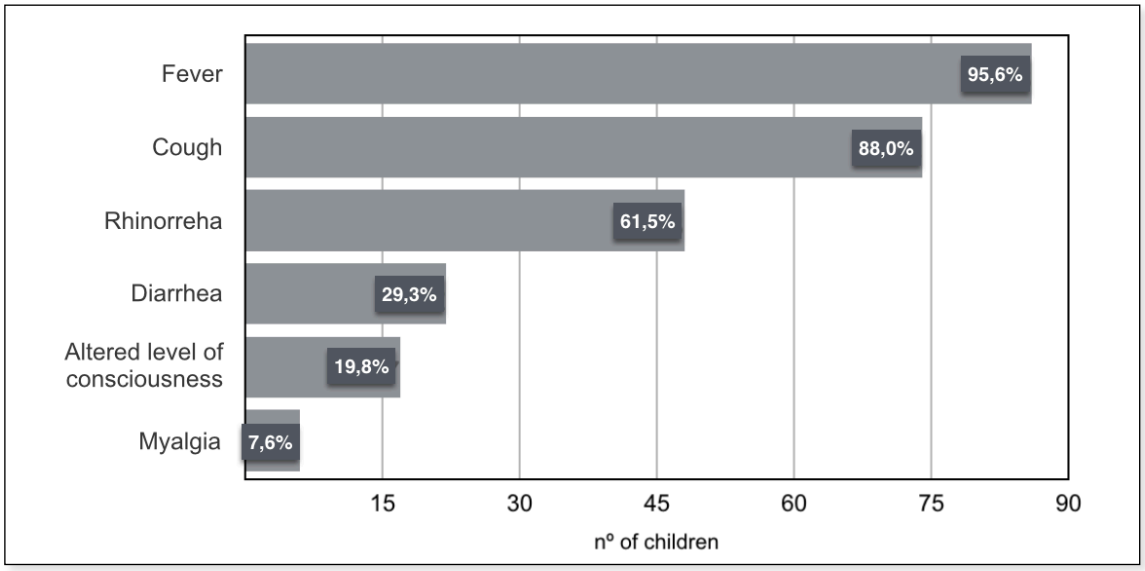

Figure 2. Clinical manifestations at presentation of influenza infection cases in the 2018-19 epidemic.

Chest X-ray

It was performed in half of the patients (n=46), showing abnormalities in all of them. Lobar or segmental consolidations were found in 12 children, while an interstitial pattern and/or hilar prominence was shown in 34.

Laboratory investigations

From the 59 (66%) children submitted to laboratory tests, 10 presented a leukocyte count >15.000/μL or <3000/μL; the CRP mean value was 3,74 ± 5,28 mg/dL, with a maximum CRP value of 23,54 mg/dL.

Influenza types and subtypes

Influenza A was the predominant type, detected in 89 (98,9%) children, from which the majority belonged to subtype H1 pdm09 (n=31; 34,4%). One child was coinfected by H1 and H3 and two were not subtyped. Only one child presented with influenza B virus infection (Figure 3).

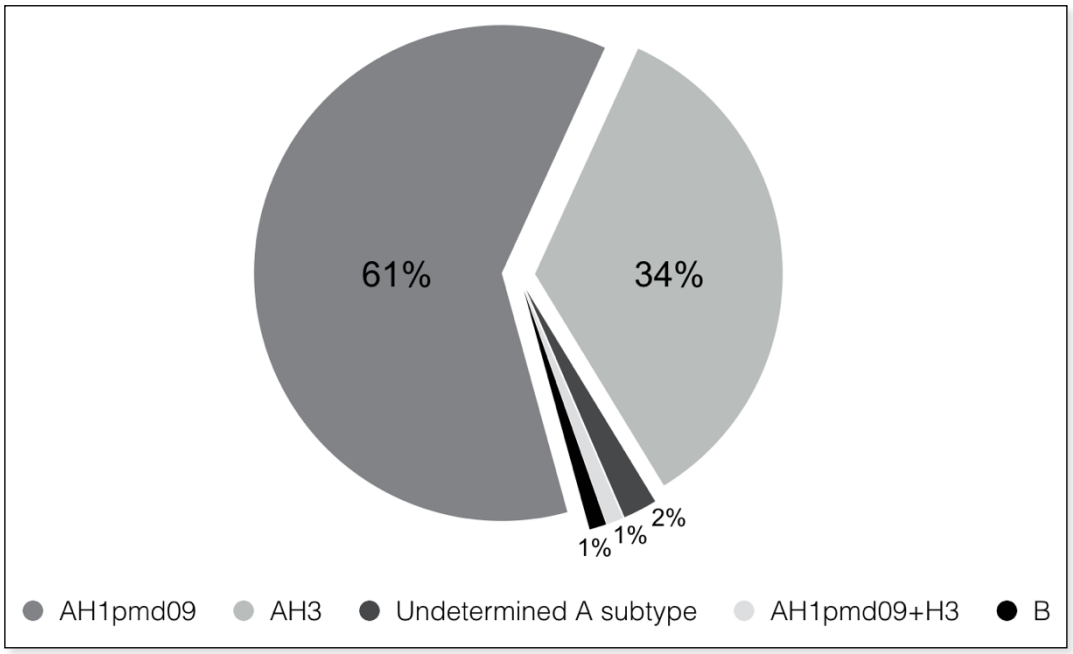

Figure 3. Distribution of influenza types and subtypes in children in the 2018-19 epidemic.

Co-detection of pathogens in respiratory secretions

Co-detection of other pathogens occurred in 41 (45,6%) cases. Respiratory syncytial virus (RSV) was the most common (n=19; 46,3%), followed by Rhino/Enterovirus and Coronavirus types HKU1, 229E, OC43, NL63 (n=14; 34,1% and n=12; 29,3% respectively). Adenovirus and Parainfluenza were founded in 7 (17,1%) children each. There were 17 (41,5%) children with bacterial co-detection, including 7 (17,1%) *Mycoplasma pneumoniae*, 10 (24,4%) *Chlamydia pneumoniae* and one child had both species.

Influenza virus vaccination

Seven (7,8%) children were vaccinated against influenza virus, all with risk factors.

Antiviral and antibiotic treatment

Oseltamivir was prescribed to 51 (57%) children. 10 out of 61 (16%) children with indication for antiviral treatment did not receive it. Of the 18 immunocompromised patients, 15 (83%) were treated with oseltamivir.

Antibiotics were used in one third of the cases (n=31). Amoxicillin-clavulanic acid was prescribed to 10 children, including six diagnosed with pneumonia and one with *Staphylococcus aureus* bacteremia. Eight children (including 6 with pneumonia, one with AOM and one with both diagnoses) received amoxicillin; macrolides were prescribed to nine children (8 with pneumonia and 1 with pneumonia and AOM).

Complications and hospitalization

Complications were observed in 46 (51%) children, being pneumonia the most common (n=27). AOM was diagnosed in four individuals, whereas encephalitis and toxic shock by *Streptococcus pyogenes* A group in one child each. There was one bacteremia by *Staphylococcus aureus*. Other complications were decompensation of an underlying disease (n=9), hypoxemia (n=4), febrile seizures (n=4), hypoglycemia (n=1) and myositis (n=2).

The most severe influenza-related complication was the toxic shock syndrome, that occurred in a one-year old boy and ended in finger's amputation.

Hospitalization was required in 29 patients (32%).

Mortality

There were no deaths.

Subgroups comparisons

Healthy and at-risk children

The mean age (5,6) of children with risk factors was higher than the mean age (2,6) of the healthy ones, showing a significant p value (p=0.001).

Chest X-ray was done in 29 (63%) healthy children and in 17 (37%) at risk children (p=0.701).

There were seven healthy children and two at risk children with leucocytes count >15.000/μL or <3000/μL (p=0.112).

As for the intake of oseltamivir, this medication was given to 22 (43%) healthy children and to 29 (57%) children with comorbidities (p<0.001). Antibiotic was given to 10 (32%) children at risk and to 21 (68%) with no risk (p=0.350).

Of the 46 patients that had complications, 32 were healthy children, and 14 belonged to risk groups, detailed in Table 1. No statistically significant association was found (p=0.093).

Table 1: Complications of healthy children and children with risk factors in the 2018-19 influenza epidemic.

Complications	Healthy Children	Children with risk factors
Pneumonia	n=22	n=5
Decompensated underlying disease	n=1	n=8
OMA	n=4	-
Hypoxemia	n=2	n=2
Myositis	n=2	-
Febrile seizure	n=3	n=1
Toxic shock syndrome	n=1	-
Bacteremia	-	n=1
Encephalitis	n=1	-
Hypoglycemia	n=1	-

Hospitalization was required in 18 (62%) previously healthy children and in 11 (38%) high risk patients. The presence of risk factors did not show an association with hospital admissions. However, a significant p value was found for the immunocompromised patients (p=0,047), showing that the majority of hospitalized children (n=27) were not immunocompromised. As for the asthmatic patients, no association was observed.

Of the 35 children that had risk factors, 7 (20%) were vaccinated; their age and comorbidities are shown in Table 2. In the non-vaccinated group, there were 18 immunocompromised children: 12 with chronic respiratory diseases and 5 with other chronic conditions. No healthy children had received the vaccine.

Table 2: Age and risk factors of vaccinated children in the 2018-19 influenza epidemic.

Vaccinated Children	Age, years	Risk factor
Child A	1	Acute lymphocytic leukemia
Child B	3	Autoimmune lymphoprolipherative syndrome (ALPS)
Child C	4	Acute lymphocytic leukemia
Child D	4	Acute lymphocytic leukemia
Child E	6	Acute myeloid leukemia
Child F	9	Splenectomy due to pyruvate kinase deficiency
Child G	13	Splenectomy due to pyruvate kinase deficiency

Table 3: Children characteristics by the presence of risk factors; p calculated using the Chi-square or Fischer's exact test, in case there is one table cell smaller than 5.

Characteristics	Total	Healthy children	Children with comorbidities	P value
Oseltamivir intake	51	22 (43%)	29 (57%)	**-0,000**
Antibiotic intake	31	21 (68%)	10 (32%)	0,350
Leukocytes count >15.000/μL or <3000/μL	9	7 (78%)	2 (22%)	0,112
X-ray consolidation	29	12 (41%)	17 (59%)	0,701
Complications	46	32 (70%)	14 (30%)	0,093
Hospitalization	29	18 (62%)	11 (38%)	0,898

Complications

There were no statistically significant differences found in the different age groups, between children with/without complications (p=0.111).

One third of the immunocompromised patients developed complications, being the majority of complications seen in immunocompetent children (p=0,027).

Of the seven vaccinated children, three had no complications and four had pneumonia (n=1), decompensated underlying disease (n=2) and bacteremia (n=1).

Hospitalizations

The majority of hospitalized children were less than 5 years old (p=0,222), 17 children were previously healthy and eight had risk factors. The immunocompromised children were less hospitalized than the immunocompetent (p=0,047).

Regarding the intake of oseltamivir or antibiotic, both showed a statistically significant association with hospital admissions (p=0,000 and p=0,001, respectively). A great majority (86%) of all hospitalized children were taking oseltamivir.

From 17 patients with bacterial co-detection, just four were hospitalized; no association was observed between these variables (p=0.566).

Of the 7 vaccinated children, one was hospitalized (p=0,421).

Almost all hospitalized children had influenza virus infection complications (89,7%), such as pneumonia (34,5%) and decompensated underlying disease (17,2%), which were the most commonly observed. There were three hospitalized children that did not develop any complications; however, they were high risk patients, with oncologic and cardiac risk factors.

When we consider a univariate logistic regression model to assess the possibility of using the patient's age as a predictor for admission, we obtain a statistically significant model in which for every 1-year increase in age, the odds ratio for admission decreases 16% (p value=0.041). However, when we adjust the previously mentioned model for potential confounders, this previously statistically significant association does not apply, which suggests that other factors might play a role in influencing our patients' outcomes in terms of hospital admissions.

Length of stay

From the 29 hospitalized children, the median length of stay (LOS) was 6 (2-10) days (Figure 5), being the shortest stay of 1 day and the longest of 86 days. The child with the longest stay was a previously healthy child with group A streptococcal shock syndrome. Of the four children that were hospitalized for 20-30 days, one was a previously healthy child and the others had associated risk factors, detailed in Table 4.

We noticed that children with bacterial co-detection, taking oseltamivir and antibiotic, with associated risk factors and asthma had a higher length of stay.

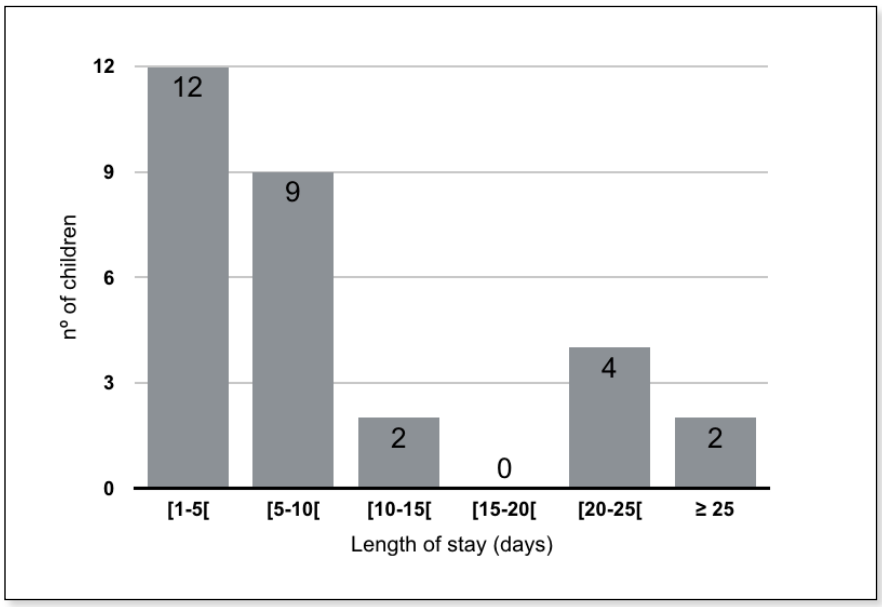

Figure 4: Distribution of children by the length of hospital stay.

Table 4: Characteristics of children hospitalized for a period of time ≥ to 20 days.

	Age, years	Risk factors / comorbidities	Complications	Length of stay, days
Child 1	0.5	Epileptic encephalopathy	Decompensated underlying disease + Pneumonia	20
Child 2	0.83	Asthma	Pneumonia with hypoxemia	20
Child 3	0.3	None	*Staphylococcus aureus* bacteremia	21
Child 4	0.64	Recurrent respiratory infections	Pneumonia with hypoxemia	23
Child 5	1.1	None	Toxic shock by *Streptococcus pyogenes* (presence of multiorgan failure, hand and feet fingers' necrosis and necrotizing pneumonia)	86
Child 6	1.66	Hypoxic-ischemic encephalopathy (already hospitalized)	Decompensated underlying disease with the need of non-invasive ventilation	30

For further information, the results on children characteristics by the need of hospitalization are presented in Appendix 1 and on children characteristics by the length of hospital stay in Appendix 2.

Discussion

This study provides details on the characteristics of laboratory-confirmed influenza in children in a pediatric hospital in one influenza season, showing that flu can be presented as a severe infection, including in healthy children.

Most cases occurred in children under five years of age and a large majority of the hospitalized children (25/29) are included in this age group. Studies consistently report higher rates and likelihood of hospitalization in younger children[5,4]. Although the National Influenza Surveillance Program (NSIP) reports that there was a higher incidence of influenza cases in individuals aged between 14-65 years old, it also states that children under 14 years old are underrepresented notified cases.
The mean age of children without risk factors was found lower (2,6 years old) than the mean age from children with comorbidities (5,6 years old), showing a significant association ($p < 0.001$). In a retrospective review[4] on influenza hospitalizations in children aged <16 years, the percentage of previously healthy hospitalized children was much higher in the early ages, while with the increase in age the percentage of children with and without risk factors started to be equivalent. This can be due to the fact that young children have a less developed immune system, even if healthy, than the older ones.

According to the NISP report of influenza season 2018/2019, there was a higher incidence of cases in December 2018 and February 2019, however in our study the majority of admissions occurred in January 2019.

The main clinical manifestations were fever, cough and rhinorrhea. These results are in agreement with the study of Silvennoinen *et al.*[12] who demonstrated in a prospective cohort study that fever was the most frequent sign of influenza infection, occurring in 95% of all children, followed by cough and rhinitis. As for the presence of gastrointestinal manifestations, they were found more frequent in our population (29,3%) than in the study mentioned above (9%). The low number of children with myalgia in our series could be explained by the fact that influenza B virus, which is more frequently associated with this complain, has been detected only in one child. Additionally, this is a subjective symptom, that might be difficult to evaluate in young children.

Influenza A was found in almost all cases in this season, from which 61% were H1pdm09 subtype and 34% had the H3 subtype. According to the NISP report, the 2018/19 season

was dominated by the A type (99%), subtype H3 (65,2%), which differs from our results. This difference can be due to the underrepresentation of children in the NISP report.

Almost half of the children had co-detection of another respiratory virus or bacteria. This proportion is in accordance with the NISP, that reported 66,4% cases of influenza illness associated with other virus, more frequently in children under 4 years of age. The report also shows that the RSV was the most common virus (66%), as it was in our study. The present of other viruses might represent a true co-infection, or their presence might represent colonization or a past infection.

A systematic review by Klein *et al*. showed that the most frequent bacterial coinfection among children with laboratory confirmed influenza was by *Streptococcus pneumoniae*. We did not identify pneumococcus in any of our blood cultures. In children it is difficult to identify this pathogen in respiratory infections because the proportion of positive blood cultures is low, and detection cannot be done in respiratory secretions in which pneumococcus is frequently found as a colonizer.

More than half of the children with bacterial co-detection in the respiratory secretions developed complications and 23,3% were hospitalized. This is in accordance with a recent publication[13] showing that coinfection is related to a more severe influenza illness, representing up to 22% of influenza-confirmed hospitalized cases. The presence of bacterial coinfection can be due to the fragility of the respiratory airways induced by the virus[14].

Some studies show that the use of antibiotics in hospitalized influenza children is usually seen in more than half of the patients, concluding that there is an inappropriate and unnecessary use of this medication[4,15,16]. In our study, antibiotics were used in 34% of children, a lower rate than in the studies mentioned before.

Regarding the intake of oseltamivir, we found that more than half of the children receiving it had comorbidities. The majority of the hospitalized children (89%) received oseltamivir, as recommended, however some that could benefit did not. In a retrospective cohort study[17] that included 35 909 children hospitalized with influenza during 2007-2015 influenza seasons, only 69% were treated with antiviral. A systematic review[4] on influenza hospitalizations in children aged <16 years over a 3 year period (2011-2013) showed an even lower percentage (20,5%) of antivirals' prescription. The NISP report says that in the past influenza season of 2018/19 oseltamivir was prescribed to 23% of

confirmed influenza patients, showing a higher frequency comparing to the previous season but still a very low number according to DGS recommendations. In our population we found higher percentages comparing to the Portuguese population data.
Since clinical deterioration and severity were the main reasons for the observed admissions, and being this a criteria for oseltamivir intake according to the DGS recommendations[7], oseltamivir was prescribed to almost all hospitalized children.

Regarding vaccination, only 20% of the children to whom the vaccine was recommended and funded[11] have received it and there was an absence of vaccination across the healthy cohort.
An Australian hospital-based cohort study by Iskander *et al.*[16] conducted in 275 hospitalized children under five years old, showed a vaccination rate of 8% for high risk children and 3,5% from the healthy group. A study with laboratory confirmed influenza in children younger than 15 years old in six tertiary/quaternary hospitals in Australia[15] reported that only 18,3% of children with formal indication for vaccination had received the vaccine.
The national report[6] shows that influenza vaccine was administrated to 15,9% of the notified cases. Furthermore, a report from the European Centre for Disease Prevention and Control (ECDC)[18] shows that, in three influenza seasons, the vaccination coverage rate (VCR) in the high risk population group (from seven Member States, in which Portugal was included) was between 15,7% and 57,1%. None of the countries achieved the UE target of 75%. There are several reasons for this low covarage[19], such as the parents' and doctors' perception of influenza as a benign disease, the lack of dissemination of correct information in the population and the presence of wrong beliefs that the vaccine can cause the illness itself.
Of our vaccinated children, four had hematologic cancer, two a genetic hematologic disorder and one an autoimmune hematologic syndrome, making us think that hematologists are probably more alert to the importance of influenza virus vaccination among high risk patients than other specialties or subspecialties.
Protection of children under six months of age, can be achieved with the vaccination of pregnant women, showing a clinical effectiveness of 63% in reducing laboratory-confirmed influenza illness in infants up to 6 months old[20] and of 91,5% in reducing hospitalization in children from the same group[21]. Information about maternal influenza vaccination was not available for analysis.
There was laboratory-confirmed influenza illness in the 7 immunized children. An Interim report across six European studies for the influenza season of 2018/19[22] showed a

vaccine effectiveness between 32% and 43%, among individuals of all ages with laboratory confirmed influenza A, in primary care and hospital settings.

More than half of our children developed complications. The most commonly found was pneumonia, followed by AOM. The most severe complications were an encephalitis and a toxic shock syndrome.
A retrospective review[4] of hospitalized children with laboratory-confirmed influenza during 2011-2013 influenza seasons, found 19,1% with pneumonia, 1,8% with bacteremia, 5,9% with febrile seizures and 1,5% with encephalitis/encephalopathy. Our pneumonia rate was higher and for the other complications rates were similar.

As reported in many seasonal influenza studies[1,5,15,23], younger children are more likely to develop complications and to be hospitalized. In this study we noticed that our younger children had higher number of complications and of hospitalizations than the ones over five years old. However, no significant correlation was found, which may be due to a small sample size of older children.
Considering the univariate logistic regression model, we obtain a statistically significant model in which for every 1-year increase in age, the odds ratio for admission decreases 16% (p value=0.041). This data is in accordance with the systematic review by Principi *et al.*[24] showing that children in the first two years of life had higher rates of hospitalization, which declined with increasing age. However, when adjusting our data to potential confounders, this association does not apply, meaning that other factors are influencing our patients' admissions. Increasing our sample size population, by studying other influenza seasons, could give us a better idea about the importance of each of these variables.

An association between vaccinated children and complications or hospitalization is difficult to determine, since our sample population of vaccinated children is very small.

We found that a large number of hospitalized patients were previously healthy children, as for the children with complications. It was also notable that only one third of the immunocompromised patients developed complications (p=0,027), and the majority of the hospitalized children were immunocompetent (p=0,047).
These findings are in accordance with several studies[4,16,19,25], showing that healthy children are at risk for influenza hospitalization and that the prevention of this infection should also focus on this group. It is also important to emphasize that, although influenza deaths are fortunately rare in children, they might happen in healthy children, with no high risk factors[3,19]. In fact, the child with the most severe influenza-related complication

from our study was a previously healthy child, that developed a toxic shock syndrome caused by *Streptococcus pyogenes*. Robert *et al*.[26] reported a case of a previously healthy 8-year-old boy that developed toxic shock complicating influenza A, caused by *Staphylococcus aureus*. The same was described in another case report by Sharkey *et al*.[27] on a 16-year-old child. This author concluded that although toxic shock represents a rare complication of influenza infection, it should be considered in any patient with multiorgan involvement following an influenza infection.

A review by Herrera *et al.*[28] highlighted the fact that influenza virus superinfections by group A *Streptococcus* are the cause of many deaths through decades. The mortality of superinfections (influenza-*Streptococcus*) in mice seemed to be related to the persistence of the bacteria in the lungs, that led to subsequent bacteremia, multi-organ infection and necrotizing fasciitis. Influenza virus vaccination of mice showed protection against mortality from *Streptococcus* infection.

In a study by Lee *et al.*[29] asthma was found to be a risk factor for hospitalization. In our study, we observed a low number of hospitalized asthmatic children (n=3). As our sample population for this risk factor is very small, no associations could be found.

The length of hospital stay reflects a direct measure of hospitalization burden. Four studies[4,16,15,30] of hospitalized children showed a median length of stay (LOS) of 2 days, shorter than observed in our study in which the median LOS was of 6 (2-10) days.

There were six children with a prolonged LOS, four had risk factors, while two of them were previously healthy, being all younger than two years of age.

According to a study by Khandaker *et al.*[15] on clinical epidemiology and predictors for adverse outcomes in children admitted with laboratory-confirmed influenza, children with nosocomial influenza infection showed longer LOS (median 10 days) than the ones who acquired influenza infection in the community (median 2 days). This can justify the longer LOS of two previously hospitalized children.

We observed that children with bacterial co-detection and with complications presented a slightly higher LOS median, however the presence of a small group representativity does not allow us to take conclusions. As for the intake of antibiotics, it showed a higher median LOS comparing with children to whom this medication was not prescribed.

As shown in some studies[8,5], prompt initiation of oseltamivir intake can shorten hospital stay in children with severe influenza virus infection. In our study we observed a considerably higher LOS median in children taking oseltamivir, probably because this antiviral medication was prescribed to patients with a more severe clinical condition.

High risk patients presented a slightly higher LOS.

Strengths and weaknesses of the study

Our study is focused on a relevant topic, since it is a very common infection in children, whose prevalence and burden worldwide are a fact. To our knowledge, there are not many national studies addressing the impact of influenza virus infection in children. Among the strengths of our study, we can point the relevance in an underrepresented population in Portuguese data. All cases were laboratory confirmed.

A limitation of our study is the fact that we are facing a selection bias while studying only children admitted to the emergency service, representing the ones with a more severe infection. The fact that we conducted this study in a single center, the study population may not have been completely representative of the all country. Potential limitations for the generalization of our results include the sample size, with a low number of vaccinated children and children with certain high-risk factors, making hard to compare different groups and their different outcomes. The presence of three previously hospitalized children in our study can influence the length of hospital stay results. Due to the retrospective nature of the study there was some missing data in the notes regarding signs and symptoms. Another limitation is that about half of the children had co-detection of other virus or bacteria in the respiratory secretions, which makes it hard to differentiate if the clinical signs are only due to the influenza infection. Additionally, there were very few influenza type B infections.

Unanswered questions and future research

During our study we identified some gaps that can be addressed in future research. For this to be possible, further studies with bigger cohorts and through more influenza seasons should be considered. This would allow more reliable comparisons between age groups, healthy and high-risk groups, vaccination status and outcomes (complications, hospitalization and length of stay). Another important aspect is the need of increasing vaccination rates among risk groups and the potential benefit of extending it to healthy children and during pregnancy to protect infants in the first months of life

Conclusion

In this retrospective study, we concluded that younger children are particularly vulnerable to influenza infection, showing higher rates of complications and hospitalization than older children. An important burden of the disease was seen not only in children with comorbidities, as expected, but also in previously healthy children. Vaccination coverage should be improved, aiming a reduction on influenza morbidity. The importance of vaccination should also be considered among healthy young children and pregnant women.

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Appendices

Appendix 1: Children characteristics by the need of hospitalization; p calculated using the Chi-square or Fischer's exact test, in case there is one table cell smaller than 5.

Characteristics	Total	Hospitalized	P value
Age, years • < 5 years old, n • ≥ 5 years old, n	 68 22	 25 (36.8%) 4 (18.2%)	 0,222
Bacterial co-detection	17	4 (23.5%)	0,566
Oseltamivir intake	51	25 (49.0%)	**-0,000**
Antibiotic intake	31	17 (54.8%)	**0,001**
Vaccinated	7	1 (14.3%)	0,421
Immunocompromised	18	2 (11.1%)	**0,047**
Asthmatic patient	12	3 (25%)	0,745

Appendix 2: Length of hospital stay and related characteristics; p calculated using Wilcoxon rank sum test; Q1 - quartile 1 (25%), Q2 - quartile 3 (75%).

	Total, n	Median LOS	Range, days	p value

		(Q1, Q3), days		
Bacterial co-detection • Not present • Present	 n=25 n=4	 6 (2,10) 7 (4.5, 16)	 1 - 86 4 - 23	0,425
Complications • Not present • Present	 n=3 n=26	 6 (3, 9) 5.5 (2, 14)	 3 - 9 1 - 86	0,942
Oseltamivir intake • No • Yes	 n=4 n=25	 2 (1.5, 4.5) 6 (3, 14)	 1 - 7 1 - 86	0,064
Antibiotic intake • No • Yes	 n=12 n=17	 3 (2, 8) 7 (4, 20)	 1 - 30 1 - 86	0,173
Risk factors • Not present • Present	 n=18 n=11	 5.5 (2, 10) 6 (3, 14)	 1 - 86 2 - 30	0,402
Asthmatic patient • No • Yes	 n=26 n=3	 5.5 (2, 9) 20 (2, 23)	 1 - 86 2 - 23	0,367