Sex differences in an Italian pediatric population COVID-19 positive

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Abstract

Background: Since December 2019 coronavirus disease (COVID-19) emerged in Wuhan and spread rapidly worldwide. Despite the high number of people affected, data on clinical features and prognostic factors in children and adolescents are limited. We propose a retrospective study aimed to evaluate clinical characteristics of children infected with SARS-CoV-2 in Italy, taking into account gender differences.

Methods: A pediatric population admitted with COVID-19 to Bambino Gesù Children's Hospital of Rome (Italy) in the period from the end of February to May 2020 has been studied taking into account sex differences. Medical history, comorbidities, symptoms and laboratory findings were obtained from patients' electronic medical records.

Results: In 41 patients (21 males and 20 females) we found that: i) fever and cough were the dominant symptoms, while gastrointestinal symptoms were rare; and ii) all ages of childhood were susceptible to COVID-19. Moreover, we found that females with COVID-19, although not significantly, were older than males and required more days of hospitalization ($p = 0.01$). Conversely, males had, although not significantly, higher values of C reactive protein and erythrocyte sedimentation rate than females.

Conclusions: Based on the data listed above, sex differences were detected in an Italian pediatric COVID-19 positive population. Compared to the adults we found that COVID-19 infection in children is a non-severe inflammatory disease in both males and females. In any case, many detailed studies should be conducted.

Background

Since mid-December 2019, an infection caused by a new type of coronavirus (SARS-COV-2) emerged in Wuhan (Hubei Province, China) and spread rapidly worldwide. The emerging SARS-COV-2 is a beta coronavirus that can cause COVID-19, officially named by the World Health Organization (WHO) on February 11, 2020. This virus is highly contagious and can be transmitted by an infected person or an asymptomatic carrier through respiratory droplets, tear fluid and close contacts. The incubation period is variable. It has been estimated that the median incubation period is 5.1 days and that 97.5% of infected patients will develop symptoms within 11.5 days of infection.

Despite the high number of people affected, data on clinical features and prognostic factors in children and adolescents are limited. Children are part of a very special group. Similarly to the SARS-COV 2002-2003 epidemic [1,2], pediatric COVID-19 appears to be mild or asymptomatic [3,4]. Children become less ill than adults and most of them contract the infection mainly through close contact with their parents or other family members with COVID-19. Many children infected with SARS-COV-2 manifest a mild disease that often does not require hospitalization. Compared to adults, children have a lower chance of developing interstitial pneumonia, one of the most serious complications of the infection, which in the advanced form requires hospitalization in intensive care. As for the adults, the presence of congenital

heart disease, lung and airway disease, malnutrition and cancer makes children more susceptible to COVID-19.

There are several hypotheses on the mechanisms underlying the lower susceptibility of children to COVID-19 infection than adults: i) a more efficient immune response due to the stimulation given by typical age vaccinations; ii) a lower expression of the angiotensin-converting enzyme 2 (ACE2) receptor to which the virus would bind to enter cells [5]; iii) an "immaturity" of the ACE2 receptors, which makes it difficult for the virus to enter the body [6]; and iv) external factors (before the lockdown, children were less likely than adults to visit places that could have facilitated the spread of the virus, such as railway stations and airports) [7].

In children with COVID-19, fever and cough are the most common clinical manifestations, sometimes accompanied by fatigue, myalgia, nasal congestion, sneezing, sore throat, headache, dizziness, vomit and abdominal pain. Moreover, some children do not manifest fever, but only cough or diarrhea, or they may be asymptomatic.

Italy was one of the European countries most affected by the COVID-19 pandemic. By 16 April 2020, 1,123 children, up to nine years of age, and 1,804 adolescents, aged between 10 and 19 years old were tested positives for COVID-19 [8].

In Italy most of the data on COVID-19 pediatric patients derive from a multicenter study promoted by the Italian Society of Paediatric Infectious Diseases (SITIP), within the Italian Society of Paediatrics (SIP). In this study 168 children aged 1 day to 17 years, 94 (55.9**%)** males and 74 (40.1%) females, with confirmed COVID-19 were analyzed [9]. 65.1% of these children were hospitalized: of these, only 17 (15.5%) were sent to the hospital after seeing a paediatrician or family doctor. Moreover, 5.9% of children documented co-infections with other viruses such as respiratory syncytial virus, rhinovirus, Epstein-Barr virus, influenza A virus and a non-SARS coronavirus. Bacterial co-infection with *Streptococcus pneumoniae* has also been documented. Pre-existing chronic pathologies, such as chronic lung diseases (n = 7), congenital malformations or complex genetic syndromes (n = 14), cancer (n = 4), epilepsy were found in 33 children. Moreover, gastrointestinal (n = 2) or metabolic (n = 1) disorders were found. Among these patients 4 were immunosuppressed and 3 immunocompromised. The hospitalization rate was similar between children with and those without co-morbidity.

Studies have reported a higher incidence of COVID-19 in males than in females in the adult population [10]. This study is aimed to evaluate clinical characteristics of children infected with SARS-CoV-2 in Italy, taking into account gender differences.

For this purpose, 41 patients admitted to Bambino Gesù Children's Hospital of Rome (Italy) in the period from the end of February to May 2020 were analysed.

Methods

Study design and participants

Fourty one patients (21 males and 20 females), admitted with COVID-19 to Bambino Gesù Children's Hospital of Rome (Italy) in the period from the end of February to to May 2020, were enrolled in this retrospective cohort study. Mean age of patients was 9 years (range ≤ 1-18 years).

All patients analyzed had contracted the infection from their parents and they were hospitalized because showed signs and symptoms such as fever, cough, vomit, diarrhea, convulsions, headache or pneumonie. Only 3 patients were asymptomatic (2 males and 1 female) (**Table 1**).

The study was performed in accordance with Good Clinical Practice and the Declaration of Helsinki principles for ethical research. Ethics approval and written informed consent were waived due to the rapid emergence of this infectious disease. Three researchers and a physician collected and reviewed the data. Medical history, underlying comorbidities, symptoms and laboratory findings both at admission and during hospitalization, were obtained from patients' electronic medical records.

The date of disease onset was defined as the day when the symptoms were noticed.

Laboratory measurements

All patients underwent nasopharyngeal, eye, urine and stool swab. The presence of SARS-CoV-2 in respiratory specimens was detected by real-time reverse transcription (RT-PCR) methods. Analyses by gene amplification reaction and Real Time RT-PCR were also carried out to exclude evidence of other viral infections, including influenza, respiratory syncytial virus, avian influenza, para-influenza, adenovirus and rhinovirus. Routine bacterial and fungal examinations were also performed. Moreover, EBV infection was routinely screened and detected by using a test that identifies antibodies to EBV in the blood.

Data include all paediatric patients in whom COVID-19 was documented by at least one nasal/ pharyngeal swab specimen positive for SARS-CoV-2 nucleic acid using RT-PCR assay.

Statistical analysis

To compare average values of a continuous variable between two groups we used the Student' T test[2] . The level of significance was determined at $p \leq 0.05$.

Results

Features of patients at admission

At admission patients presented: fever (25 patients), cough (13 patients), headache (7 patients), vomit (3 patients), diarrhea (5 patients) and pneumonia (2 patients). Moreover, 3 patients were asymptomatic and 4 patients had co-infections: 2 with Rhinovirus and 2 with Epstein-Barr virus (EBV) (**Table 1**). Some analyzed patients had a history of pneumonia, bronchiolitis, asthmatic bronchitis, gastroenteritis and convulsions. Furthermore, at admission only 23 patients (15 males and 8 females) presented

bronchospasm and underwent chest x-ray. In 3 males and 5 females, a modest thickening of the peri-broncho-vascular interstitium was found.

Interestingly, from the analysis of medical records we found that females: i) were older than males (not significant data) and ii) required more days of hospitalization ($p = 0.01$).

Moreover, as shown in the **table 2**, laboratory tests showed that in the pediatric population CRP, ESR, procalcitonin values and PLTs number were age and sex independent. Conversely, fibrinogen and LDH values, as well as RBCs and WBCs number were different according to the age of the patients and some of them were also significantly different in both sexes.

In particular, we found that: i) although not significantly, in all patients CRP and ESR values were higher than the normal range only in males; ii) procalcitonin values and PLTs numbers in all patients were within the normal range; iii) for all age groups, the fibrinogen values were within the normal range and were significantly ($p = 0.05$) different in the two sexes in patients from 11 to 18 years (the values are higher in males); iv) LDH values were higher than the normal range in both males and females aged $\leq$ 1 to 3 years and were significantly higher in males less than 1 year old ($p = 026$); v) RBCs number was within the normal range for all age groups, but was significantly ($p < 0.05$) higher in males aged 7 to 13 years; vi) WBCs number was higher than the normal range only in patients aged 3 to 7 years.

Patient characteristics during hospitalization

During hospitalization 5 males developed mild thrombocytosis (average number of PLTs: 607 x 10^3/uL; range of values: 496 – 663 x 10^3/uL) and an increase in inflammation measured in terms of high CRP levels (average of values: 2.546 mg/dL; range of values: 1.07 -4.61 mg/dL). Three of these patients were less than 1 year; 1 patient was 2 years old and 1 patient was 12 years old. During hospitalization, patients less than 1-year-old manifested gastrointestinal symptoms and diarrhea; the 2-year-old patient had inflammation of the airways and pharyngitis and previously hospitalized for bronchiolitis; the 12-year-old patient developed salmonella gastroenteritis. On the basis of these data we can speculate that reactive thrombocytosis occurring in these patients may be due to an inflammatory process related to gastrointestinal disorders or inflammation of the airways.

Patient treatments

There are no specific protocols to guide treatment of children with COVID-19. Analyzing the medical records we found that 3 patients (2 males and 1 female) did not receive any therapy (they were asymptomatic and apyretic); 13 patients (9 males and 4 females) were treated only with paracetamol as needed; 11 patients were treated with paracetamol and antibiotics (4 males and 7 females); 3 males were treated only with antibiotics e 3 males only needed oxygen. Moreover, in addition to paracetamol and antibiotics, 2 females were treated with corticosteroid (they manifested bronchiolitis or cough); 1 female with anti-rheumatic drugs; 3 females with anti-inflammatory drugs and 2 females with heparin (they

showed high values of inflammatory parameters and one of them was suffering from rheumatoid arthritis).

Discussion

This study describes the characteristics of a sample of children admitted with COVID-19 to Bambino Gesù Children's Hospital of Rome (Italy) in the period from the end of February to May 2020, taking into account possible gender differences. In this retrospective study based on medical records data, we found that, compared to adults, the pediatric population gets less COVID-19 and had less severe clinical manifestations. Fever and cough were the dominant symptoms, while gastrointestinal symptoms were rare. Moreover, we found that all ages of childhood were susceptible to COVID-19: from a few days of life to 18 years. In particular, we found that females with COVID-19, although not significantly, were older than males and required more days of hospitalization ($p < 0.01$). Interestingly, from medical records some significant gender differences emerged. In particular, we found higher fibrinogen values in males ($p = 0.05$) and higher LDH values in females ($p = 0.026$). Lactate dehydrogenase (LDH) is a cytoplasmic enzyme present in all major organ systems and is released into the peripheral blood after cell death. Increased serum LDH levels are associated with pulmonary disease such as obstructive diseases, microbial pulmonary diseases and interstitial lung diseases such as acute respiratory distress syndrome [11].

Furthermore, we found values of CRP and ESR higher than the normal range values and, although not significantly, were higher in the males patients.

During hospitalization some male patients developed mild thrombocytosis and exhibited increased inflammation evaluated in term of high CRP values. CRP is an inflammatory marker that plays an important role in host defense against invading pathogens [12]. Sun et al., [13] shown that CRP was elevated in severe and critically adult patients with COVID-19.

It has been found that in virus mRNA positive patients a decline of LDH in the serum correlated with viral mRNA elimination, suggesting that constitutive decrease of LDH levels probably predict a favourable response. LDH can thus be used as indicators of disease progression [2].

Platelets have been increasingly recognized as an important component of the immune response to infections, an increase in their number above the normal range (thrombocytosis) has often been considered a sign of normal inflammatory reaction. Compared to primary thrombocytosis, the reactive thrombocytosis is not associated with higher risk of cardiovascular or thrombotic events [14].

Conclusions

Compared to other pediatric studies on COVID-19, this retrospective study, in addition to evaluate clinical characteristics of children infected with SARS-CoV-2 in Italy, takes into account the differences related to sex during infection. Although the small number of cases this study highlights some sex differences. The

mechanisms underlying these differences are not yet known. Further prospective studies should be conducted

Abbreviations

ACE2: Angiotensin-converting enzyme 2; CRP: C-reactive protein; EBV: Epstein-Barr virus; ESR: Erythrocyte sedimentation rate; LDH: Lactate dehydrogenase; PLTs: Platelets; RBCs: Red blood cells; RT-PCR: Real-time reverse transcription; SIP: Italian Society of Paediatrics; SITIP: Italian Society of paediatric Infectious Diseases; WBCs: white blood cells.

Declarations

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Author’contributions

All authors have made substantial contributions to this work. I.T., R.V., A.V. and E.S. contributed equally to this work. Acquisition, analysis, or interpretation of data C.C., L.C. and E.S. Statistical analysis: C.C. and L.G. Concept and design: E.S., A.V., I.T., A. M. and R.V.

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Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Ethics approval and consent to participate

Ethics approval and written informed consent were waived due to the rapid emergence of this infectious disease.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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Tables

Table 1. Features of patients at admission

Characteristics	Males (n = 21)	Females (n = 20)	p value
Age, average (range)-years	8.3 (range ≤ 1 - 18)	10.4 (range ≤ 1- 16)	p < 0.209
Hospitalization, median (range)-days	8.21 (range 4 – 15)	*12.35 (range 5 – 22)	*p = 0.01
Signs and symptoms			
Asymtomatic	2 (9.5%)	1 (5%)	
Fever	13 (61.9%)	12 (60%)	
Cough	6 (28.6%)	7 (35%)	
Vomit	1 (4.8%)	2 (10%)	
Diarrhea	3 (14.3%)	2 (10%)	
Convulsions	1 (4.8%)	4 (20%)	
Headache	4 (19%)	3 (15%)	
Coinfection	2 Rhinovirus (9.5%)	2 EBV (10%)	
Pneumonie	2 (9.5%)	0	

For age and hospitalization days both average of values and range of values (from the lowest to the highest value) detected in male and female patients are shown.
For signs and symptoms both patients number and percentage of patients are shown.

Table 2. Laboratory findings of patients with COVID-19

Age independent markers	Normal range	Males (n = 21)	Females (n = 20)	p values
CRP (mg/dL)	< 0.5	0.64 (range 0.03 - 4.16)	0.19 (range 0.03 - 1.57)	p = 0.138
ESR (mm/h)	0 - 15	15.7 (range 10 – 25)	13.7 (range 4 – 49)	p = 0.825
Procalcitonin (ng/ml)	< 0.5	0.17 (range 0.02 – 0.44)	0.08 (range 0.02 – 0.3)	p = 0.282
PLT (10^3/mL)	150 - 450	285 (range 135 - 426)	253 (range 170 - 531)	p = 0.231

Age dependent markers	Normal range according to the age			
Fibrinogen (mg/dL)	212 – 433 (11 -18 years)	*418 (range 390 – 500)	335 (range 219 - 540)	*p = 0.05
LDH (U/L)	120 – 300 (< 1 year)	*363 (range 345 – 380)	412 (range 393 – 425)	*p = 0.026
	160 – 370 (1- 3 years)	382 (range 295 - 557)	379 (range 346 - 453)	p = 0.970
RBC (10^6/mL)	3.8 - 4.8 (7-13 years)	*4.8 (range 4.4 - 5.27)	4.5 (range 4.16 - 4.91)	*p < 0.05
WBC (10^3/mL)	5.5 – 15 (3-7 years)	4.66 (range 3.03 – 5.6)	4.99 (range 3.8– 7.06)	p = 0.740

For age dependent markers only data above the normal range or significantly different in the two sexes were shown. In this table for each marker both average of values and range of values (from the lowest value to the highest value) detected in male and female patients are shown.