Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Clinics and Research in Hepatology and Gastroenterology 45 (2021) 101610

Available online at
ScienceDirect
www.sciencedirect.com

Elsevier Masson France
EM|consulte
www.em-consulte.com/en

''Health status of children with chronic liver disease during the SARS-CoV-2 outbreak: results from a multicentre study''

KEYWORDS
SARS-CoV-2;
COVID-19;
Chronic liver disease;
Children

The ongoing novel Coronavirus SARS-CoV-2 disease (COVID-19) may cause a systemic disease with possible involvement of other organs, including the liver, because of ubiquitous distribution of the main viral entry receptor, namely angiotensin converting enzyme 2 (ACE2) [1]. Indeed, ACE2 receptor is expressed in the liver cells, particularly in the bile duct epithelial cells, and this might favor viral entry and cause liver damage [1]. Previous studies are reassuring as far as susceptibility of immunosuppressed pediatric patients to the infection and the disease showing that, among children in the follow-up for liver transplantation and autoimmune liver disease, none developed a severe clinical pulmonary disease, despite some tested positive for SARS-CoV-2 [2–5]. However, due to the absence of published studies, it is still unclear whether children with different etiologies of chronic liver disease (CLD) are more susceptible to SARS-CoV-2 infection.

Northern Italy has been the earliest and most extensively affected European area during the earliest COVID-19 epidemic in the first semester of 2020. In this area, Lombardy, Piedmont and Veneto have represented the three regions counting the highest incidence of COVID-19 cases, leading to dramatic hospital occupation surge and high mortality [6,7].

Therefore, this dramatic scenario represents a reliable opportunity to explore the health status and possible challenges presented by children with CLD who lived in northern Italy during the outbreak.

In this multicentre study we collected data on children with CLD followed up at three pediatric centres for liver disease and transplantation in northern Italy: 1 centre in Bergamo (Lombardy region), 1 in Turin (Piedmont region), and 1 in Padua (Veneto region). Patients aged 0–18 years with CLD were included in the study; those on immunosuppressive treatment (IS) were excluded.

A questionnaire composed of 29 questions was used by physicians to collect data on health status and risk exposure during the outbreak. The questions related to the study period from January 2020 to June 2020. According to the standard definitions we defined suspected COVID-19 the presence of at least one of the following two conditions: (1) An episode of acute respiratory tract infection (RTI) (sudden onset of at least one of the following: cough, fever, shortness of breath); (2) A close contact with a confirmed or highly probable case of SARS-CoV-2 infection [9]. We defined confirmed COVID-19 case a subject having a positive nasalpharyngeal swab (NPS) for SARS-CoV-2 nucleic acid using real-time reverse-transcriptase polymerase-chain-reaction (RT- PCR) assay [8]. With regard to the estimated COVID-19 cases, considering the estimated attack rate in Italian regions, and assuming that 1.8% of the cases occurred in the age bracket 0–18 years, 24188 [19496–30354], 6126 [4899–7681] and 3312 [2597–4231] children are estimated to have been infected as of May 2020 in Lombardy, Piedmont and Veneto regions, respectively [9,10].

Of 377 eligible patients, 369 (98%, mean age: 11.1 years, ±7.7; M = 60%) filled the questionnaire. Etiologies of CLD and laboratory features are reported in Table 1; 97 patients (26%) had cirrhosis based on histology (n = 12) or radiological images (n = 85) showing: (a) surface nodularity, (b) heterogeneous echotexture, and (c) segmental hypertrophy/atrophy [11]. Thirteen patients (4%) were listed for liver transplantation (LT) (Table 1).

Fifty-six of 369 patients (15%) were classified as suspected COVID-19 according to the given definition. Forty-three of 56 patients (77%, mean age 11.3 years, ±8.1, M = 49%) developed respiratory symptoms, including fever in 28 patients (65%), cough in 23 (53%), shortness of breath in 4 (10%); 17 (40%) had a close contact with a suspected, case of COVID-19. Diagnoses were: biliary atresia (BA) in 9 patients (out of 91 with BA, 10%), chronic hepatitis B or C in 6 (out

https://doi.org/10.1016/j.clinre.2020.101610

Table 1 Etiologies, COVID-19 cases and outcome in children with chronic liver disease.

Number of patients	369
Survey response rate	98%
Male (%)	220 (60%)
Age at survey, years (SD)	11.1 (±7.7)
Etiologies of chronic liver disease	
Biliary atresia	91 (25%)
Chronic viral hepatitis B or C	79 (22%)
Vascular disorders	42 (11%)
Portal vein thrombosis, n	21
Obliterative portal-venopathy	11
Congenital porto-systemic shunting	5
Budd-Chiari syndrome	5
Alagille syndrome	29 (8%)
Inborn errors of metabolism	20 (5%)
Hereditary fructose intolerance	11
Glycogen storage disease type IX	3
LAL deficiency	3
Tyrosinemia type I	2
methyl-malonic acidemia	1
Wilson disease	16 (5%)
Fontan associated liver disease	15 (4%)
Ciliopathies	12 (3%)
Alpha1 antitrypsin deficiency	12(3%)
Primary sclerosing cholangitis	12(3%)
Progressive familial intrahepatic cholestasis*	10 (3%)
Cystic fibrosis related liver disease	7 (2%)
Miscellaneous ^	24 (6%)
Laboratory features	
AST (nv <45 UI/L)	60 (±64)
ALT (nv <45 UI/L)	68 (±69)
GGT (nv <35 UI/L)	85 (±112)
Total bilirubin mg/dl	1.3 (±2.23)
Serum bile acids (nv <6 micromol/L)	70.5 (±106)
International normalized ratio (INR)	0.7 (±0.5)
Serum albumin gr/dl (nv 32−55 gr/dl)	4.3 (±0.4)
Haemoglobin gr/dl	13.1 (±1.7)
WBC (mm/3)	6013 (±2900)
Platelet count (10^9/L)	219 (±116)
Patients listed for LT, n (%)	13 (4%)
Suspected cases of COVID-19, n (%)	56/369 (15%)
Patients with acute respiratory tract infection (RTI)**	43/56 (77%)
fever	28 (65%)
cough	23 (53%)
shortness of breath	4 (10%)
Close contact with highly suspected COVID-19 case, n^^	8/56 (14%)
Close contact with confirmed COVID-19 cases, n^^	5/56 (9%)
Confirmed cases of COVID-19, n (%)°	2/369 (0.5%)
Outcome	
Survived	369 (100%)
Patients requiring hospitalization	none

Laboratory values are expressed a mean and standard deviation.
*Type 1 in 2 patients, type 2 in 6, type 3 in 3, type 4 (TJP2) in 1.
^Cryptogenic cirrhosis in 6, non-alcoholic steatohepatitis in 5, bile acids synthetic defects in 4, non-syndromic bile duct paucity in 2, and neonatal hemochromatosis, giant hepatic hemangioendotelioma, intrahepatic artero-venous fistula, focal nodular hyperplasia, Glycerol-3-phosphate dehydrogenase (GPD 1) deficiency, Sodium taurocholate cotransporting polypeptide (SLC10A1) deficiency, arthrogryposis-renal dysfunction-cholestasis (ARC) syndrome in the other 7 patients. WBC: white blood cells; LT: liver transplantation.
**Diagnoses were: biliary atresia in 9 patients, chronic hepatitis B or C in 6, Alagille syndrome in 5, Wilson disease in 3, primary sclerosing cholangitis in 3, Fontan associated liver disease in 3, portal vein thrombosis in 3, hereditary fructose intolerance in 2, congenital hepatic fibrosis in 2, PFIC in 2, and alpha 1 antitrypsin deficiency, non-alcoholic steatohepatitis, congenital portosystemic-shunt (in a child with Down syndrome), non-syndromic bile duct paucity, and cryptogenic cirrhosis in the other 5 patients. ^^all patients remained asymptomatic. °: n = 1 patient with biliary atresia was on waiting list for liver transplantation.

of 79, 8%), Alagille syndrome (AS) in 5 (out of 29, 17%), Wilson disease (WD) in 3 (out of 16, 19%), others in 20 patients (Table 1). Nine children had cirrhosis. Overall, all presented with mild/moderate symptoms, none required admission to hospital nor oxygen therapy. No patients received a NPS for SARS-CoV-2, and therefore they were named as suspected cases of COVID-19.

Thirteen out of 56 patients (23%, mean age 11.2, ±7.8, M = 70%) had a close contact with a suspected (n = 8) or confirmed (n = 5) COVID-19 case. Diagnoses were: WD in 3 patients (1 patient with cirrhosis), BA in 2 (1 with Cat-Eye syndrome and cirrhosis), and hereditary fructose intolerance (HFI), obliterative portal venopathy (OPV), congenital hepatic fibrosis (CHF), AS, alpha 1 anti-trypsin deficiency (AAT-D), chronic HCV, focal nodular hyperplasia (plus overweight and raised transaminases), and Fontan associated liver disease (FALD) in the other 8 patients. All 13 patients remained asymptomatic and none received NPS for SARS-CoV-2 infection (Table 1).

Overall, of 30 patients who had a close contact with a highly suspected (n = 25) or confirmed COVID-19 case (n = 5), 17 (57%) developed respiratory symptoms and 13 (43%) remained asymptomatic.

Two of 369 patients (0.5%) had a positive NPS for SARS-CoV-2 and were classified as confirmed COVID-19 cases:

- Patient n. 1. Female (aged 6 years, with Adams-Oliver syndrome), from Lombardy region, no comorbidities. She had type-2 Abernethy malformation, with histological features

of OPV, complications of portal hypertension (PH) and normal liver synthetic function (serum albumin 3.8 gr/dl, INR 1.1). In June 2020, the patient (and her mother) received a NPS for SARS-CoV-2 before being admitted to our ward to undergo endoscopic surveillance of oesophageal varices; NPS was positive in both subjects. Thus, the endoscopic procedure was cancelled, and the patient was discharged home. She remained asymptomatic and 2 weeks later received NPS which turned up negative.

- Patient n. 2. Male (with BA and situs viscerum inversus), aged 18 years, from Lombardy, no comorbidities. He was recently listed for liver transplantation due to complications of PH. In June 2020 the patient received a call for LT but the NPS performed at the time of admission was positive for SARS-CoV-2. The patient was asymptomatic but, according to the Italian Organ Sharing Network policy, the transplant was not performed, and the patient was sent home.

Both cases reported above were classified as asymptomatic COVID-19 cases.

Of interest, with the over mentioned assumptions, at the time of the survey the estimated incidence in Lombardy, Piedmont, and Veneto would be 1392 (95%CrI: 1122—1747], 896 (95%CrI: 717—1124), and 401 (95%CrI: 314—512) COVID-19 cases per 100,000 inhabitants respectively.

In this cohort, putting together the suspected (n = 56) and the confirmed cases (n = 2), the observed incidence of SARS-CoV-2 infection was 20547 per 100,000 inhabitants with 29 observed cases of COVID-19 in Lombardy; 15238 per 100,000 inhabitants with 17 cases in Piedmont; 10169 per 100,000 inhabitants with 12 observed cases in Veneto.

Overall, at the end of the study period 369 patients (100%) survived: n = 311 healthy, n = 56 suspected COVID-19 (n = 43 were symptomatic), n = 2 confirmed asymptomatic COVID-19.

No patients were admitted due to severe RTI nor severe course of COVID-19. The percentage of symptomatic children who were classified as suspected COVID-19 (n = 43) among cirrhotic patients (9/97, 9%) was similar to that reported among non-cirrhotic patients (34/272, 12%, $p > 0.05$).

In this study we enrolled children who had different causes of CLD, 25% of patients (n = 97) had histological and/or radiological features of cirrhosis, and 13 (4%) were listed for LT.

Previous studies demonstrated that SARS-CoV-2 basic reproductive number during unmitigated circulation ranges from 2.2 to 2.6, supporting the impression of higher contagiousness of this virus compared to the previous ones (SARS-CoV and MERS-CoV) [12]. The results from this study show that, of 30 children (8%) who had a close contact with a suspected (n = 25) or confirmed (n = 5) case of COVID-19, half (n = 17, 57%) became symptomatic, confirming the high contagiousness of this virus also in children with CLD. This percentage (57%) is similar to that reported in our previous studies on pediatric liver transplant recipients (44%) and patients with AILD (50%) suggesting that the susceptibility to SARS-CoV-2 infection in children with CLD is similar to that reported in those with liver diseases requiring IS treatment [2,3].

In adults with COVID-19, cirrhosis is a risk factor associated with a worse outcome [1]. In Singh's study, patients with pre-existing liver diseases were found to have an increased risk of mortality compared to patients without liver disease; the relative risk was higher in patients with cirrhosis [13]. Different results are reported in our study. Of 43 symptomatic patients classified as suspected COVID-19, only 9 (21%) had histological and/or radiological features of cirrhosis and these patients had a favorable outcome, similarly to patients without cirrhosis, suggesting that cirrhosis likely does not represent an increased risk factor to SARS-CoV-2 infection or more severe course of COVID-19 in children.

The observed incidence of cases in our cohort of patients was higher than the estimated incidence in the general population and this might suggest that children with CLD are more susceptible to COVID-19 than the general population. On the other hand, we support the hypothesis that the calculated estimated incidence in the general population may be underestimated because, as reported in Buonsenso's study, the real incidence of COVID-19 cases in the pediatric population is likely higher, as SARS-CoV-2 is not actively searched in children since they are usually asymptomatic or have mild respiratory symptoms [14].

Overall, health status of our patients was satisfactory since the majority of them (n = 326, 88%) remained asymptomatic during the study period.

Among symptomatic patients (n = 43) classified as suspected COVID-19 we found children with different causes of liver disease; furthermore, we did not identify subgroups of patients with significantly higher percentages of symptomatic cases, suggesting a similar susceptibility to SARS-CoV-2 infection regardless of the underlying CLD. These results are different from what reported in adult studies where some subgroups of patients (with cirrhosis, NASH or liver cancer) have been found to be at increased risk for severe COVID-19, and have poorer prognosis [13,15]. Of interest, in our cohort, all symptomatic patients developed only mild/moderate respiratory symptoms not requiring hospitalization nor oxygen therapy. Furthermore, 2 patients with confirmed COVID-19 remained asymptomatic despite they had a CLD complicated by PH (in both patients) and cirrhosis (in 1). These results show that children with CLD and COVID-19 may be asymptomatic or have a mild course of disease, similarly to what reported in the general pediatric population [1—4].

In conclusion, during the SARS-CoV-2 outbreak, the majority of patients maintained a good health status. Susceptibility to SARS-CoV-2 infection was similar in the different groups of patients, regardless of the underlying CLD. Despite a high incidence of observed suspected cases, the absence of major clinical events and a favorable outcome, even in confirmed COVID-19 cases, suggests that in pediatric patients an underlying liver disease does not represent an additional risk factor for severe COVID-19.

Contribution of authors

All authors contributed to the conception, design, acquisition, analysis and interpretation of data, drafting, revising and final approval of the manuscript.

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Conflict of interest

The Authors declare no conflict of interest.

References

[1] Zhang Chao, Shi Lei, Wang Fu-Sheng. Liver injury in COVID-19: management and challenges. Lancet Gastroenterol Hepatol 2020;5:428—30, http://dx.doi.org/10.1016/S2468-1253(20)30057-1.
[2] Di Giorgio A, Nicastro E, Speziani C, De Giorgio M, Pasulo L, Magro B, et al. Health status of patients with autoimmune liver disease during SARS-CoV-2 outbreak in northern Italy. J Hepatol 2020;73:702—5, http://dx.doi.org/10.1016/j.jhep.2020.05.008.
[3] Nicastro E, Di Giorgio A, Zambelli M, Ginammi M, Bravi M, Stroppa P, et al. Impact of the severe acute respiratory syndrome coronavirus 2 outbreak on pediatric liver transplant recipients in Lombardy, Northern Italy. Liver Transpl 2020;26(10):1359—62, http://dx.doi.org/10.1111/apt.15813.
[4] D'Antiga Lorenzo. Coronaviruses and immunosuppressed patients: the facts during the third epidemic. Liver Transpl 2020;26:832—4, http://dx.doi.org/10.1002/lt.25756.
[5] Doná D, Canizales JT, Benetti E, Cananzi M, De Corti F, Calore E, et al. Pediatric transplantation in Europe during the COVID-19 pandemic: early impact on activity and healthcare. Clin Transplant 2020;12:e14063, http://dx.doi.org/10.1111/ctr.14063.
[6] Flaxman S, Mishra S, Gandy A, Unwin HJT, Coupland H, Mellan TA, et al [cited 2020 Apr 6]. Available from: http://spiral.imperial.ac.uk/handle/10044/1/77731, 2020.
[7] Istituto Nazionale di Statistica, ISTAT. Popolazione residente, 2019. http://dati.istat.it/Index.aspx?QueryId=18462.
[8] Huang C, Wang Y, Xingwang Li X, Ren L, Zhao J, Hu Y, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet 2020;395:497—506.
[9] Vollmer Michaela AC, Mishra S, Unwin H, Juliette T, Gandy A, Mellan TA, et al. Using mobility to estimate the transmission intensity of COVID-19 in Italy: a subnational analysis with future scenarios. Imperial College London; 2020, http://dx.doi.org/10.25561/78677.
[10] https://www.epicentro.iss.it/coronavirus/bollettino/Bollettino-sorveglianza-integrata-COVID-19_20-maggio-2020.pdf.
[11] Sharma S, Khalili K, Nguyen GC. Non-invasive diagnosis of advanced fibrosis and cirrhosis. World J Gastroenterol 2014;7(20):16820—30, http://dx.doi.org/10.3748/wjg.v20.i45.16820.
[12] Wu JT, Leung K, Leung GM. Nowcasting and forecasting the potential domestic and international spread of the 2019-nCoV outbreak originating in Wuhan, China: a modelling study. Lancet 2020;395:689—97, http://dx.doi.org/10.1016/S0140-6736(20)30260-9.
[13] Singh S, Khan A. Clinical characteristics and outcomes of coronavirus disease 2019 among patients with preexisting liver disease in the United States: a multicenter research network study. Gastroenterology 2020;159(2):768—71, http://dx.doi.org/10.1053/j.gastro.2020.04.064, e3.
[14] Buonsenso D, Zampino G, Valentini P. Novel coronavirus disease 2019 infection in children: the dark side of a worldwide outbreak. Front Pediatr 2020;8:215, http://dx.doi.org/10.3389/fped.2020.00215.
[15] Ji D, Qin E, Xu J, Zhang D, Cheng G, Wang Y, et al. Non-alcoholic fatty liver diseases in patients with COVID-19: a retrospective study. J Hepatol 2020;73:451—3, http://dx.doi.org/10.1016/j.jhep.2020.03.044.

A. Di Giorgio[a,*]
E. Nicastro[a]
S. Arnaboldi[a]
O. Montini[a]
F. Di Stasio[a]
L. D'Antiga[a]
[a] *Paediatric Hepatology, Gastroenterology and Transplantation, Hospital Papa Giovanni XXIII Bergamo, Italy*
P. Gaio[b]
L.N. Fovino[b]
M. Cananzi[b]
[b] *Unit of Gastroenterology, Digestive Endoscopy, Hepatology and Care of the Child With Liver Transplantation, University Hospital of Padova, Italy*
M. Pinon[c]
PL. Calvo[c]
[c] *Paediatric Gastroenterology Unit, Regina Margherita Children's Hospital, AOU Città Della Salute e Della Scienza Di Torino, University of Turin, Turin, Italy*
V. Camelli[d]
[d] *Postgraduation School of Paediatrics, Regina Margherita Children's Hospital, AOU Città Della Salute e Della Scienza Di Torino, University of Turin, Turin, Italy*

[*] Corresponding author.
E-mail address: adigiorgio@asst-pg23.it (A. Di Giorgio)

Available online 1 January 2021