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Original article

Hematological Changes with Covid-19 and Post Covid-19 Syndrome and its Outcome in Hospitalized Children in Benha Children Hospital

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Abstract:

Background: Our goal is to study the hematological changes in Covid-19 and the relation between these changes and the progression of the disease. **Methods:** It combined a retrospective and prospective study at the isolation unit of Covid-19 department of Benha Children Hospital. The time frame for this research was from January of 2021 through December of the same year. All patients diagnosed with COVID-19 or Multisystem inflammatory syndrome in children (MIS-C) /Kawasaki, were included in the study. **Results:** This study included 300 patients who were divided into two groups based on their PCR results. Group I: negative PCR group (223 patients). Group II: positive PCR group (77 patients). Mean age was statistically significantly higher among negative COVID 19 group than positive group (p=0.03). Mean HB and HCT % were statistically significantly higher among negative COVID 19 group than positive group (p=0.02 and 0.03 respectively), Mean D-dimmer, S-ferritin and L D H were statistically significantly higher among positive COVID 19 group than negative group (p<0.001). Contact with a probable, confirmed or linked to cluster results, was statistically significantly higher among positive COVID 19 (58.4%) group than negative group (14.8%) (p<0.001). Less than 1 year old age was statistically significantly higher among positive COVID 19 (61%) group than negative group (45.7%) (p=0.02). **Conclusion:** Diagnosing COVID-19 and determining prognosis may be aided by testing blood parameters. Shifts in the lymphocyte count are not readily apparent in children. However, D-dimer levels are elevated in COVID-19-infected children. Abnormal hematological parameters are one way in which COVID-19 might manifest itself, and they can serve as early evidence for diagnosis and prognosis.

Keywords: COVID-19; Pediatrics; Post-COVID Syndrome

Introduction

In children less than 5 years old, pneumonia is one of the main causes of illness and death. These days, covid-19 is a major reason why kids get pneumonia. Since its first outbreak in Wuhan, China in December 2019, the new coronavirus disease 2019 (COVID-19) has spread throughout the globe. Severe acute respiratory syndrome coronavirus (SARS-cov-2) was identified as the causative agent, and Corona Virus Disease 2019 was officially designated (COVID-19). (1)

A wide range of risk factors for morbidity and mortality are known, including age, gender, ethnicity, and comorbid conditions. However, even in the absence of an underlying disease, young people can develop potentially fatal complications such as myocarditis and disseminated intravascular coagulopathy. (2) Children with severe COVID-19 frequently have abnormal laboratory parameters that reveal a systemic inflammatory response and prognostic biomarkers, which are usually accompanied by leukopenia, lymphopenia, thrombocytopenia, and coagulopathy, which frequently lead to disseminated intravascular coagulopathy. (3) However, data in the pediatric age group is still lacking; it was initially stated that children are always asymptomatic to COVID-19, but subsequent reports indicated that they can develop the pathology and symptomatology of the disease. (4)

According to the findings of several epidemiological studies, the disease is more severe and fatal in the elderly population, whereas infectivity rates and fatalities are lower in the child population. (5)

Venous thromboembolism (VTE), including pulmonary embolism (PE) and Deep venous thrombosis (DVT), is extremely common in critically ill COVID-19 patients. Pulmonary embolism causes an increase in dead-space ventilation, resulting in a sudden increase in arterial carbon dioxide partial pressure. Increased D-dimer (> 0.5 g/mL) on admission was identified as one of the risk factors for venous thromboembolism. Stroke (reported in 1.6%) and myocardial infarction (reported in 8.9%) were associated with an increase in mortality rate (HR 1.99; 95% CI 1.65-2.40). An Italian single-center study reported 20 patients with COVID-19 and acute limb ischemia. When compared to the previous year, the incidence rate of critical limb ischemia was higher (16.3% vs. 1.8%; p 0.001). (6)

COVID-19 coagulation testing results typically range from normal or slightly prolonged prothrombin time (PT) and

activated plasma thromboplastin time (aPTT) in mild disease to marked prolongation of PT and aPTT in severe disease. Platelet counts can be normal or slightly elevated in mild illness, but significantly lower in severe disease (7). A recent systematic review discovered an increase in D-dimer in patients with severe COVID-19 disease and acute respiratory distress syndrome (ARDS). ARDS patients who did not survive had higher D-dimer levels than survivors. (8)

Lymphopenia

SARS-CoV-2 infection is thought to be associated with CD4+ and CD8+ T cell lymphopenia, which occurs through different mechanisms. SARS-CoV-2 is known to attack human cells by binding to the angiotensin-converting enzyme-2 (ACE-2) receptor, which is primarily found in the lungs, heart, and gastrointestinal tract. These receptors can be found on the surface of lymphocytes as well. As a result, SARS-CoV-2 may bind to lymphocytes directly and cause cell lysis. As a result, lymphopenia may contribute to the inflammatory cascade. The disease progresses due to a decrease in cytotoxic T cells (CTLs) and natural Killer (NK) cells, which are essential for viral infection control. Lymphopenia has been reported in a high number of COVID-19 patients, particularly those with severe illness. In severe disease, CTLs, NK cells, memory T cells, and B cells are reduced. (9)

Neutrophilia

Immune dysregulation caused by COVID-19 results in neutrophil production and lymphocyte apoptosis. As previously stated, neutrophilia is associated with lymphopenia. Furthermore, neutrophilia can be caused by a secondary bacterial infection, which is more likely in patients with severe disease. A lesser-known neutrophil function has recently been proposed as a contributor to SARS-CoV-2 pathogenesis. Neutrophil extracellular traps (NETs) are web structures made of DNA and proteins by neutrophils that are designed to entrap pathogens. (10)

Neutrophilia is linked to the progression of COVID-19 disease, an increased risk of (ARDS), and death. The neutrophil-to-lymphocyte ratio (NLR) has been used as a prognostic indicator for conditions such as acute or chronic hepatitis B liver failure, as well as a risk factor for mortality in cancer, acute coronary syndrome, and cerebral hemorrhage. (11)

Biomarkers

C reactive protein (CRP)

As a sensitive biomarker for a variety of inflammatory diseases, such as infection and tissue injury, this acute-phase reactant

is activated by IL-6 and generated by the liver. Most individuals with life-threatening illnesses have elevated CRP levels. Multiple studies have demonstrated that elevated blood CRP levels are a reliable indication of SARS-CoV-2 infection presence and severity. (12)

Lactate dehydrogenase (LDH)

An enzyme found in almost all human cells, including those of the lungs, heart, liver, kidneys, and bone marrow that also catalysis the conversion of pyruvate to lactate inside cells. High blood LDH levels result from damage to any of the several types of cells that express LDH. Critically ill COVID-19 patients often have an increase in LDH.

D-dimer

It results from the breakdown of fibrin crosslinks and represents an uptick in coagulation and fibrinolysis. Furthermore, D-dimer was investigated as a possible predictive indicator of disease severity in COVID-19. Patients who died from COVID-19 had considerably higher D-dimer levels on admission, and these levels remained high throughout the late stages of illness in all fatalities, according to a preliminary, small research dealing with coagulation parameters in COVID-19 patients with pneumonia. (13)

Ferritin

It is not only useful for iron storage, but it is also a well-known acute phase reactant. It is made up of subunits H and L, and its synthesis is triggered by various inflammatory stimuli such as cytokines like interlukin6 (IL-6). Notably, it is proposed that inflammatory stimuli must drive the production of the H subunit.

Methods

It combined a retrospective and prospective study, carried out at the isolation CoVid department of Benha Children Hospital. The time frame for this research was from January of 2021 through December of the same year. All patients diagnosed with COVID-19 or post-COVID MIS-C/Kawasaki based on PCR results were included in the study. On the day of admission, full blood and differential counts were taken. This study included 300 patients who were divided into two groups based on their PCR results. Group I: negative PCR group (223 patients). Group II: positive PCR group (77 patients).

Children ranging in age from 29 days to 19 years old and of both sexes were chosen. All cases of covid-19 infection, whether confirmed or suspected, were included. All children who had direct or indirect contact with covid-19 cases were also included.

The following clinical manifestations may occur:

1. Respiratory manifestations (fever, nasal congestion, sore throat, cough, respiratory difficulty, rhinorry and poor feeding).
2. Symptoms of the gastrointestinal tract (Diarrhea, vomiting, and abdominal pain).
3. Skin manifestations (maculopapular, urticarial, and vesicular eruptions).
4. Neurologic manifestations (severe encephalopathy,stroke,infection/demyelination of the central nervous system, Guillain-Barre syndrome/variants, acute fulminant cerebral edema).
5. Cardiovascular problems (heart failure, arrhythmias, myocarditis, pericarditis, cardiogenic shock, pulmonary embolism, ST elevation myocardial infarction). (14, 15)

Before enrolling in the study, an Ethical Scientific Committee of Benha University approved the study protocol, and informed consent was obtained from the parents. (Coding: Ms.6.11.2021)

All cases were subjected to a complete clinical examination, including a general examination, cardiopulmonary examination, gastrointestinal examination, and laboratory investigations, including measurement of (CBC, CRP, ferritin, and d-dimmer), as well as nasopharyngeal and oropharyngeal swabs for PCR (polymerase chain reaction).

Statistical methods

SPSS version 25(IBM, Armonk, New York, United States) was used for data management and statistical analysis. The Kolmogorov-Smirnov test and direct visualization methods were used to determine the normality of numerical data. The numerical information was then summarized as means and standard deviations or medians and ranges. Numbers and percentages were used to summarize categorical data. For normally and non-normally distributed numerical data, the independent t-test or Mann-Whitney U test were used to compare the two groups. Categorical data were compared using the Chi-square test or, if appropriate, Fisher's exact test.

Results

Mean age was statistically significantly higher among negative COVID 19 group than positive group (p=0.03), however there was no statistically significant difference between groups regarding sex (Table 1).

Seventy three of cases presenting by acute attach of gastro enteritis (24.3%) while all other cases were presented by respiratory manifestations. Eight cases were diagnosed as post COVID MIS-C/ Kawasaki disease and associated diseases were congenital heart disease (6 cases), cerebral palsy (6 cases), failure to thrive (3

cases), Down syndrome (4 cases), familial Mediterranean fever (2 cases), immune deficiency (1 case) and asthmatic patient (1 case).

Mean HB and HCT % were statistically significant higher among negative COVID 19 group than positive group (p=0.02 and 0.03 respectively), however there was no statistically significant difference between groups regarding total leukocytic count (TLC), Lymphocyte, neutrophil, N/L ratio, red blood cells (RBCs) and platelets (PLT) (Table 2).

Mean D-dimmer, S-ferritin and L D H were statistically significantly higher among positive COVID 19 group than negative group (p<0.001), however there was no statistically significant difference between groups regarding serum level of CRP (Table 3).

Table (1): Comparison of study groups regarding age and sex

Characteristics		**Negative Group (n=223)**		**Positive Group (n=77)**		**Test of sig.**	**p-value**
Age/years (mean ± SD)		2.54	3.06	1.75	2.29	2.1	0.03
Sex No. (%)	**F**	90	40.4%	35	45.5%	0.6	0.4
	M	133	59.6%	42	54.5%		

Table (2): Comparison of study groups regarding CBC findings

Characteristics	**Negative Group (n=223)**		**Positive Group (n=77)**		**Test of sig.**	**p-value**
	mean	**SD**	**mean**	**SD**		
TLC (10^3/mm)	9.57	4.98	9.60	6.01	0.8	0.4
Lymphocyte(10^3/mm)	2.54	2.07	2.44	2.08	0.8	0.4
neutrophil(10^3/mm)	6.59	4.58	6.91	5.24	0.2	0.8
N/L ratio	3.85	3.84	4.29	4.30	0.9	0.3
RBCs(mg/dl)	4.15	0.56	4.09	0.83	0.8	0.5
PLT (10^3/mm)	314.18	140.78	299.99	181.57	1.2	0.2
Hb (g/dl)	10.83	1.53	10.35	2.02	2.4	0.02
HCT (%)	33.66	24.93	29.96	6.89	2.2	0.03

Table (3): Comparison of study groups regarding inflammatory markers

Characteristics	**Negative Group (n=223)**		**Positive Group (n=77)**		**Test of sig.**	**p-value**
	mean	**SD**	**mean**	**SD**		
CRP (mg/l)	29.49	32.62	30.80	31.63	0.3	0.8
D-dimmer(mg/L)	0.61	1.03	2.23	1.46	9.5	<0.001
S-ferritin(ng/ml)	226.80	214.23	588.62	342.15	8.6	<0.001
L D H(IU/L)	180.77	116.87	304.47	198.96	4.8	<0.001

Contact with a probable, confirmed or linked to cluster results, was statistically significantly higher among positive COVID 19 (58.4%) group than negative group (14.8%) (p<0.001) and there was no statistically significant difference between groups regarding CT findings (Table 4).

Less than 1 year old age was statistically significantly higher among positive COVID 19 (61%) group than negative group (45.7%) (p=0.02). However, there was no statistically significant difference between groups regarding Co-morbidities (Table 5).

There was no statistically significant difference between groups regarding both complications and outcome (Table 6).

Table (4): Comparison of study groups regarding Contact results and CT chest

Characteristics		Negative Group (n=223)		Positive Group (n=77)		X^2	p-value
Contact* No. (%)	Negative	190	85.2%	32	41.6%	56.7	<0.001
	Positive	33	14.8%	45	58.4%		
CT No. (%)	Free	59	26.5%	14	18.2%	2.1	0.1
	*GGO	164	73.5%	63	81.8%		

*GGO: Ground glass opacity.
*Contact with a probable, confirmed, or linked to cluster results.

Table (5): Comparison of study groups regarding risk factors

Characteristics		Negative Group (n=223)		Positive Group (n=77)		X^2	p-value
Age No. (%)	<1 y	102	45.7%	47	61.0%	5.3	0.02
	≥ 1 Y	121	54.3%	30	39.0%		
Co-morbidities No. (%)	No	205	91.9%	70	90.9%	0.1	0.8
	Yes	18	8.1%	7	9.1%		

Table (6): Comparison of study groups regarding outcome and complications

Characteristics		Negative Group (n=223)		Positive Group (n=77)		X^2	p-value
outcome No. (%)	Died	6	2.7%	1	1.3%	0.5	0.5
	improved	217	97.3%	76	98.7%		
Severity No. (%)	No	140	62.8%	58	75.3%	4.01	0.04
	Yes	83	37.2%	19	24.7%		

Discussion

Since the discovery of coronavirus, numerous studies have been published emphasizing the importance of hematological abnormalities in predicting disease severity and prognosis. Many studies have found an increased prevalence of lymphopenia in patients with critical COVID-19 infection. (16) COVID-19 is a multisystem infection that wreaks havoc on the hematopoietic system and hemostasis. According to the severity of the infection, the most common hematological findings were lymphopenia, leukocytosis, neutrophilia, and hypercoagulability. Peripheral blood differences to assess disease severity have been widely reported in adults, but not so much in children (21). According to recent studies, the majority of children with COVID-19 had a normal WBC count. Leukopenia was the most commonly reported hematological abnormality. Lymphopenia was said to be more common in children than in adults.

Pediatric patients account for approximately 5% of all COVID-19 patients diagnosed. COVID-19 infection in children is milder than in adults, with less than 1% of hospital admissions. (19) As a result, this could be a plausible explanation for the absence of leukopenia in significant pediatric patients.

Our study sought to ascertain the effect of COVID-19 on complete blood count and hematological changes, as well as the impact of these changes on the child's progress and outcome.

All confirmed and suspected cases of COVID-19 infection and post-COVID MIS-C/Kawasaki. Disease diagnosed on the basis of PCR will be included. Complete blood and differential counts is performed on the day of admission.

Ethical permission for this study was obtained from the parents, who were fully informed about all procedures and consented before their children enrolled in this study. The ethical committee of the faculty of Medicine at Benha University Hospitals approved our study.

In our study, 77 (25.6%) of 300 patients were diagnosed with COVID-19, 223 (74.3%) were negative, and 8 were diagnosed with post COVID MIS-C/ Kawasaki disease. Positive contact results were statistically significantly higher in the positive COVID 19 (58.4%) group than in the negative group (14.8%) (p0.001). the positive group was discovered in clusters of cases surrounding patients. Furthermore, there was no statistically significant difference in CT findings between the two groups.

In our study, age was a risk factor that increased the rate of admission and severity of the disease, so age less than one

year old was statistically significantly higher in the positive COVID 19 (61%) group than the negative group (45.7%) (p=0.02). In terms of co-morbidities, however, there was no statistically significant difference between the two groups. Congenital heart disease (6 cases), cerebral palsy (6 cases), failure to thrive (3 cases), Down syndrome (4 cases), familial Mediterranean fever (2 cases), immune deficiency (1 case), and asthmatic patient were the most common co-morbidities (1 case).

In our study, there was a statistically significant difference between groups in terms of complications, with 17 cases out of 77 (22%) being complicated in the positive group. MISC (7 cases, 4 of which died), Kawasaki disease (4 cases), Diabetic-keto acidosis (2 cases), and appendicitis were the most common complications (4 cases).

In our study, the clinical presentation of COVID-19 in children and the most commonly reported symptoms were fever, cough and respiratory manifestations that agree with the study in the United States (25) and a cohort study involving 651 pediatric cases in the UK as the main presenting symptoms were fever and a runny nose that were more common in younger children, while vomiting, abdominal pain, headache and a sore throat showed an increasing trend with age. (26)

The mean age of children in the negative COVID-19 group was statistically significantly higher (3.06 Age/years) than in the positive group (2.29 Age/years, p=0.03).

Our findings are consistent with the findings of a study in Kuwait (20) and Saudi Arabia (23) (the mean age was 2.8 years). While the mean age of children in the study in Pakistan (21) was 7.04.3 years, the mean age of children in the study in Turkey was 10.75 years. (22)

In our study, there was no statistically significant difference in sex between positive and negative groups, agreeing with search in Turkey (22) and disagreeing with search in Pakistan (21) where male predominance was found and with a study in Saudi Arabia (23) where female predominance was found. In our study, we discovered that the mean HB and HCT% were statistically significantly higher in the negative COVID 19 group than in the positive group (p=0.02 and 0.03 respectively). The mean Hg was 10.35 g/dl, which agrees with a search in Kuwait (20) and a search in Pakistan. (21).

However, there was no statistically significant difference between the positive and negative groups in terms of TLC (mean 9.60x109/L), Lymphocyte (mean2.44x109/L), neutrophil (mean 6.91x109/L), N/L ratio (mean4.29), RBCs (mean 4.09x109/L), and PLT

(mean299.99x109/L) in the positive group, agreeing with the studies by the study in Pakistan (21). Our study found that mean D-dimmer (2.23), S-ferritin (588.62), and LDH (304.47) levels were statistically significantly higher in the positive COVID 19 group than in the negative group (p0.001), which agrees with studies by the study in Turkey (22) and a study in Saudi Arabia (23)

However, there was no statistically significant difference between the two groups in terms of CRP, as reported by the study in Pakistan (21) and the study in the United States. (24).

In the end, there was no statistically significant difference in outcome between groups. All seven of the dyed cases were complicated by MISC. Other cases improved and were able to return home safely after a few days of precautionary measures.

Conclusion

Diagnosing COVID-19 and determining prognosis may be aided by testing blood parameters. Shifts in the lymphocyte count are not readily apparent in children. However, D-dimer levels are elevated in COVID-19-infected children. Abnormal hematological parameters are one way in which COVID-19 might manifest itself, and they can serve as early evidence for diagnosis and prognosis.

Recommendations

Dynamic blood parameters may have crucial reference value for dynamically monitoring patients' condition and assessing the therapy impact, thus clinicians should pay greater attention to them.

Research on the effects of COVID 19 on children is needed.

It is recommended that further research be done to determine the effects of COVID-19 in children.

Abbreviations

MIS-C Multisystem inflammatory syndrome in children
VTE Venous thromboembolism
PE pulmonary embolism
DVT Deep venous thrombosis
PT prothrombin time
aPTT activated plasma thromboplastin time.
NLR neutrophil-to-lymphocyte ratio
ARDS Acute respiratory distress syndrome
NETs Neutrophil extracellular traps
GGO Ground glass opacity

Statements

Before enrolling in the study, an Ethical Scientific Committee of Benha University approved the study protocol (Coding: Ms.6.11.2021), and informed consent was obtained from the parents.

Permission to publish.

Potential conflict of interest

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