Analysis of hematological parameters of COVID-19 / SARS-CoV-2 positive patients treated at a university hospital in northeastern Brazil

Análise dos parâmetros hematológicos de pacientes positivos COVID-19 / SARS-CoV-2 tratados em um hospital universitário no nordeste do Brasil

Análisis de parámetros hematológicos de pacientes COVID-19 / SARS-CoV-2 positivos atendidos en un hospital universitario del noreste de Brasil

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Abstract
The objective of this work was to analyze the hematological parameters of patients positive for COVID-19, admitted to the COVID intensive care unit (ICU-COVID) of the University Hospital Lauro Wanderley (HULW), to correlate these data with the clinical conditions of critically ill patients, compared to patients in the ward of the COVID Infectious Parasitic Diseases Unit (IDP-COVID). The study population consisted of patients in (ICU-COVID) and (IDP-COVID) of both genders, considering children, adolescents, adults, and elderly people hospitalized in this unit with positive serology or molecular tests (RT-PCR). This was a retrospective observational cross-sectional study involving patients in the (ICU-COVID) and (IDP-COVID) of the HULW. The study was carried out in accordance with the requirements of the Research Ethics Committee (CAAE 46718921.4.0000.5183) of the HULW. 152 exams were analyzed. Of these, 88 (58%) were male and the average in relation to the age group was 61.5 years. By correlating the data obtained from inpatients in the COVID-ICU and IDP-COVID, it can be observed in the ICU-COVID patients there was the predominance of leukocytosis (50.4%), as well as lymphopenia (90.9%), neutrophilia (80%) and anemia (70%). Thrombocytopenia is present in 23% of patients in the COVID-ICU and alterations in Prothrombin Time (PT) and Activated Partial Thromboplastin Time (APTT) were identified. These data corroborate the literature, in which the total leukocyte count varies between patients, which may reflect the dominance of lymphopenia or neutrophilia. Taken together, decreased lymphocytes accompanied by mild thrombocytopenia are among the most common abnormal findings that attract attention in the blood count of COVID-19 patients.

Keywords: SARS-CoV-2 Infection; Hematological disease; Intensive care unit; Severe acute respiratory syndrome; Hemoglobin A.
Resumo
O objetivo desse trabalho foi analisar os parâmetros hematológicos de pacientes positivos para COVID-19, admitidos na unidade de terapia intensiva COVID (UTI-COVID) do Hospital Universitário Lauro Wanderley (HULW), com a finalidade de correlacionar esses dados com as condições clínicas dos pacientes em estado grave, comparando com os pacientes da enfermaria da Unidade de Doenças Infecciosas Parasitárias COVID (DIP-COVID). A população do estudo foi composta por pacientes na (UTI-COVID) e (DIP-COVID) de ambos os gêneros, considerando crianças, adolescentes, adultos e idosos hospitalizados nessa unidade com sorologia ou testes moleculares (RT-PCR) positivos. Tratou-se de um estudo transversal retrospectivo observacional envolvendo pacientes na (UTI-COVID) e (DIP-COVID) do HULW. O estudo foi realizado de acordo com as exigências do Comitê de Ética em Pesquisa (CAAE 46718921.4.0000.5183) do HULW. Foram analisados 152 exames. Destes, 88 (58%) eram do gênero masculino e a média em relação a faixa etária ficou 61,5 anos. Ao correlacionar os dados obtidos de pacientes internos na UTI-COVID e DIP-COVID, pode observar nos pacientes UTI-COVID houve o predomínio de leucocitose (50,4%), como também linfopenia (90,9%), neutrofilia (80%) e anemia (70%). A trombocitopenia está presente em 23% dos pacientes na UTI-COVID e identificou-se alteração no Tempo de Protrombina (TP) e no Tempo de Tromboplastina Parcial Ativada (TTPA). Esses dados corroboraram com a literatura, em que a contagem total de leucócitos varia entre os pacientes, o que pode refletir a dominância de linfopenia ou neutrofilia. Em conjunto, a diminuição de linfócitos acompanhada de trombocitopenia leve está entre os achados anormais mais comuns que atramen atenção no hemograma de pacientes com COVID-19.
Palavras-chave: Infecção por SARS-CoV-2; Doença hematológica; Unidade de tratamento intensivo; Síndrome respiratória aguda grave; Hemoglobina A.

Resumen
El objetivo de este trabajo fue analizar los parámetros hematológicos de pacientes positivos para COVID-19, ingresados en la unidad de cuidados intensivos COVID (UCI-COVID) del Hospital Universitario Lauro Wanderley (HULW), con el fin de correlacionar estos datos con la clínica, condiciones de los pacientes críticos, en comparación con los pacientes en la sala de la Unidad de Enfermedades Infecciosas Parasitarias COVID (DIP-COVID). La población de estudio está compuesta por pacientes en (UCI-COVID) y (DIP-COVID) de ambos los sexos, considerando niños, adolescentes, adultos y ancianos hospitalizados en esta unidad con sorología o pruebas moleculares (RT-PCR) positivas. Se trata de un estudio transversal observacional retrospectivo en el que participaron pacientes de (UCI-COVID) y (DIP-COVID) del HULW. El estudio se realizó de acuerdo con los requisitos del Comité de Ética en Investigación (CAAE 46718921.4.0000.5183) del HULW. Se analizaron 152 exámenes. De estos, 88 (58%) eran del sexo masculino y el promedio en relación al grupo de edad fue 61,5 años. Al correlacionar los datos obtenidos de los pacientes ingresados en la UCI-COVID y DIP-COVID, se puede observar que en los pacientes de la UCI-COVID predominó la leucocitosis (50,4%), así como la linfopenia (90,9%), la neutrofilia (80%). (%) %) y anemia (70%). La trombocitopenia está presente en el 23% de los pacientes en la UCI-COVID y se identificaron alteraciones en el Tiempo de Protrombina (PT) y el Tiempo de Tromboplastina Parcial Activada (TTPA). Estos datos corroboran la literatura, en la que el recuento total de leucocitos varía entre pacientes, lo que puede reflejar el predominio de la linfopenia o la neutrofilia. En conjunto, los hallazgos de disminución de linfocitos acompañados de trombocitopenia leve se encuentran entre los anormales más comunes que llaman la atención en el hemograma de los pacientes con COVID-19.
Palabras clave: Infección por SARS-CoV-2; Enfermedad hematológica; Unidad de cuidados intensivos; Síndrome respiratorio agudo severo; Hemoglobina A.

1. Introduction
According to the World Health Organization (WHO) (2020), the disease caused by the infection of the new coronavirus (COVID-19), refers to a severe acute respiratory syndrome caused by coronavirus 2 (SARS-CoV-2), a betacoronavirus that it is mainly transmitted by direct or indirect contact with respiratory droplets from infected people. (Johns Hopkins University & Medicine, 2020; Zhu et al., 2020; Dong et al., 2020; Gorbalenya et al., 2020).

The first suspicions of the disease emerged from reports of cases of pneumonia of unknown etiology in the city of Wuhan and China (Lu et al., 2020). Just over 6 months after these reports, the COVID-19 epidemic became a serious pandemic leading to more than three million cases that overwhelmed health systems and led to an alarming number of deaths worldwide (Worldometer, 2020).

Coronaviruses are positive-sense, non-segmented enveloped RNA viruses belonging to the Coronaviridae family and the order Nidovirales and widely distributed in humans and other mammals. In the last two decades, two other serious
respiratory diseases in humans, termed as severe acute respiratory syndrome (SARS) and Middle East Respiratory Syndrome (MERS), have co-etiological agents betacoronavirus, SARS-CoV and MERS-CoV, respectively. SARS-CoV-2 shares 79% genetic sequence identity with SARS-CoV, the virus responsible for the 2002-2004 SARS outbreak (Lu et al., 2020; Zhou et al., 2020).

While much of the pathogenesis of Covid-19 remains to be unraveled, it is known that SARS-CoV-2, like SARS-CoV, binds to host cells through its receptor, angiotensin-converting enzyme 2 (ACE 2), which is expressed in a wide range of human cells from different tissues, including type II lung pneumocytes and the endothelium of blood vessels (Ziegler et al., 2020 & Hamming et al., 2004).

There is still limited knowledge about the risks of transmission of the SARS-CoV-2 virus and its associated disease (Covid-19) from routine clinical samples. The first published study of 41 early cases of Covid-19 infections admitted to Wuhan detected SARS-CoV-2 RNA in the blood of six patients, corresponding to 15% of the total (Huang et al., 2020). However, in another study conducted on 1,070 clinical samples collected from 205 confirmed, infected Covid-19 patients residing in China had the highest positive rates of SARS-CoV-2 reverse transcriptase in the polymerase chain reaction (rRT-PCR) test. of respiratory samples such as bronchoalveolar lavage, sputum, and nasopharyngeal swabs (32%-93%). In contrast, only 1% of the blood samples and none of the urine samples tested positive (Wang et al., 2020).

Understanding of the COVID-19 process in patients with solid malignancies is increasing, while reports of patients with hematologic malignancies remain scarce. Patients with hematologic malignancies are at increased risk of COVID-19 due to disease biology and associated therapy. In addition, immunocompromised patients with COVID-19 may also be at higher risk of bacterial or fungal pneumonia overlap (Paul et al., 2020).

In a study of hematology parameters in COVID-19 patients hospitalized in Singapore, mean absolute lymphocyte count (ALC) was significantly lower in patients requiring intensive care unit (ICU) admission (0.4 × 109/L vs. 1.2 × 109 / L), as well as neutrophilia (11.6 × 109 / L vs 3.5 × 109 / L) (Fan, Chong, & Chan, 2020). Mild thrombocytopenia (100-150 × 109/L) has been reported in up to 20%-36% of COVID-19 cases, however, thrombocytopenia (<50 × 109/L) is uncommon. In an inpatient case series in an intensive care unit in Wuhan, a platelet count <100 × 109/L was observed in only 5% of patients (Huang et al., 2020).

Based on the presentation of eleven clinical cases of Covid-19, (Dong et. al., 2020), reported the complexity of the clinical presentation of patients affected with the disease, which can present itself through mild to severe symptoms, with or without pneumonia, to asymptomatic cases. In this work, the authors also cite the main clinical manifestations of patients in clinical studies, which were fever, fatigue, dry cough, nausea, vomiting and diarrhea, in addition to the presentation of images on computed tomography of the chest, characteristics of a viral pneumonia, with bilateral ground-glass opacity foci. In laboratory findings, lymphopenia and leukopenia are usually found.

These laboratory findings are related to many cytokines in the body, caused by an immune response that culminates in changes in the levels of leukocytes and lymphocytes in peripheral blood (Gao et al., 2020). Nevertheless, lymphopenia may be associated with a reduction in the immune response to the virus. Associated with this, neutrophilia is related to bacterial super infections in the course of the disease. Thrombocytopenia and D-dimer, markers present in severe disease states, denote widespread changes in blood clotting (Lippi & Plebani, 2020).

In this perspective, this study aims to evaluate hematological parameters of patients positive for Covid-19, admitted to the intensive care unit of Hospital Universitário Lauro Wanderley (HULW), to correlate these data with the clinical conditions of patients in serious.
2. Methodology

Data source

This is an observational retrospective cross-sectional study involving the population of João Pessoa and neighboring cities attended at the COVID Intensive Care Unit (ICU-COVID) and COVID Infectious Parasitic Diseases Unit (IDP-COVID) at the University Hospital Lauro Wanderley (HULW). The study was conducted as previously described by Aragão Neto et al. (2021). Data collection took place from March 2020 to August 2020, when the pandemic began in Brazil. Data collection was carried out through the i9LIS information system, which is a laboratory management software. i9LIS can control, in an automated way, the flow of information about the exam. Thus, such information was used to obtain the hematological parameters of interest to be analyzed, such as: leukocytes, neutrophils, lymphocytes, monocytes, platelets, hemoglobin, prothrombin time, activated partial thromboplastin time. The information collected referred to the period from March 2020 to August 2020, marked by the beginning of the pandemic in Brazil. The data obtained were formatted in tables and organized in a database using the program Microsoft Office Excel® version 2010. The study was carried out in accordance with the requirements of the Research Ethics Committee (CEP) of the HULW (CAAE 46718921.4.0000.5183).

Laboratory tests

**Table 1.** List of hematology and coagulation tests used in this data analysis.

| Test                        | Sample type                        |
|-----------------------------|------------------------------------|
| Whole blood count           | Whole blood in EDTA                |
| Peripheral blood film       |                                    |
| Coagulation assays          |                                    |
| Prothrombin time (PT)       | Whole blood with 3.2% sodium citrate|
| Activated partial thromboplastin time (APTT) | |
| International normalized ratio (INR) | |

Source: Authors.

Statistical analysis

Continuous variables were expressed as median (IQR) and compared with the t-test or Mann-Whitney test; categorical variables were expressed as number (%) and compared by the $\chi^2$ test or Fisher's exact test among patients who received care in the ICU and IDI (infectious diseases infirmary). Results were considered significant when the p value was less than or equal to 0.05. Statistical analyzes were performed using GraphPad Prism Software, version 6.01.

3. Results and Discussion

We retrospectively analyzed 152 hematological exams that were requested to the Clinical Analysis Laboratory of the Hospital Universitário Lauro Wanderley from March 2020 to August 2020. First, the epidemiological data were collected
Based on the requests for the aforementioned exams, above where it was observed that of the 152 requests, 88 (58%) were male and 64 (42%) were female. The average in relation to age group was 61.5, observing a higher incidence for patients aged 60 years (Table 2).

**Table 2.** Demographic data of patients infected with SARS-CoV2.

| Characteristics | All patients (n=152) | ICU (n=121) | Infirmary (n=31) | P value  |
|-----------------|----------------------|------------|-----------------|----------|
| Age, in years   | 61.5 (45-71)         | 67 (46-73) | 47 (40-57)      | 0.0002   |
| Gender          |                      |            |                 |          |
| Male            | 88 (58%)             | 67 (55%)   | 21 (68%)        | 0.2294   |
| Female          | 64 (42%)             | 54 (45%)   | 10 (32%)        |          |

Data are median (IQR), n (%) or n/N (%), where N is the total number of patients with available data. P values are expressed by comparing patients who depend on intensive care unit (ICU) and ward care (no ICU care), using the χ² test, Fisher's exact test, t test, or Mann-Whitney test. Source: Authors.

By correlating the data obtained from inpatients in the ICU-COVID and IDP-COVID, it can be seen that males were the most affected by COVID-19 (Table 2), with older patients developing the most severe clinical form. of the virus requiring greater care (Figure 1).

**Figure 1.** Analysis of demographic data in relation to incidences between the COVID Intensive Care Unit (ICU-COVID) and the COVID Infectious Parasitic Disease Unit (IDP-COVID).

Through Table 2, it is possible to identify those children and adolescent with COVID-19 infections have less severe symptoms compared to adults, which may justify the lower number of hospitalizations in the COVID-ICU, reported in this data collection of only 2 cases (age group 0 to 18 years). It corroborates the literature, which recognizes that children with
COVID-19 infections have less severe symptoms compared to adults and that some confirmed cases were asymptomatic (Lam et al., 2020).

In the analysis of the hematological parameters, what can be observed is that 50.4% of the patients in the COVID-ICU had leukocytosis and the greatest variations in relation to the total leukocyte count were from the patients of the ward, presenting respectively 25.8%, leukocytosis and 9.7% leukopenia (Table 3) (Figure 2). According to the literature, leukocyte elevations are related to the prediction of poor outcomes in patients with covid-19, that is, leukocytosis associated with severe infection by this virus (Yamada et al., 2020).

Table 3. Laboratory findings of patients infected with SARS-CoV2 during hospital stay.

| Characteristics                  | All patients (n=152) | ICU (n=121) | Infirmary (n=31) | P value |
|----------------------------------|---------------------|------------|------------------|---------|
| **WBC count, × 10⁹ per L**       |                     |            |                  |         |
| <4                               | 3/152 (2%)          | 0/121 (0%) | 3/31 (9.7%)      | 0.0003  |
| 4–11                             | 69/152 (45%)        | 61/121 (50.4%) | 8/31 (25.8%)   |         |
| **Band count, %**                | 2 (0.0-5.0)         | 2 (0.0-6.0) | 0 (0.0-2.0)      | 0.0010  |
| **Segmented neutrophil count, %**| 79.0 (70.0-85.0)    | 80.0 (76.0-86.0) | 69.0 (59.0-77.0) | < 0.0001|
| **Metamyelocyte count, %**       | 0.0 (0.0-1.0)       | 2.0 (0.0-6.0) | 0.0 (0.0-2.0)   | 0.0010  |
| **Lymphocyte count, %**          | 10.0 (7.0-16.0)     | 9.0 (6.0-14.0) | 18.0 (14.0-27.0) | < 0.0001|
| <20                              | 126/152 (84.2%)     | 110/121 (90.9%) | 18/31 (58.1%) | < 0.0001|
| 20–50                            | 23/152 (15.1%)      | 10/121 (8.3%) | 13/31 (41.9%)   |        |
| >50                              | 1/152 (0.7%)        | 1/121 (0.8%) | 0/31 (0%)       |        |
| **Atypical lymphocyte count, %** | 0.0 (0.0-0.5)       | 0.0 (0.0-2.0) | 0.0 (0.0-0.0)   | 0.064  |
| **Monocyte count, %**            | 6.0 (4.0-8.0)       | 5.0 (4.0-7.0) | 8.0 (6.0-9.0)   | 0.0005  |
| **Red blood cell count, × 10⁹ per L** | 3.78 (2.96-4.38) | 3.65 (2.89-4.26) | 4.37 (3.56-4.77) | 0.0039  |
| **Hemoglobin, g/L**              | 11.0 (9.0-13.0)     | 10.9 (6.7-12.6) | 13.0 (10.3-13.5) | 0.0043  |
| **RDW, %**                       | 13.9 (12.8-15.3)    | 14.6 (13.5-15.8) | 13.2 (12.5-14.6) | 0.0020  |
| **Hematocrit, %**                | 33.2 (26.7-38.1)    | 32.6 (25.5-37.5) | 36.7 (29.4-40.3) | 0.0127  |
| **Platelet count, × 10⁹ per L**   | 216.5 (146.6-316.6) | 208.0 (144.5-265.5) | 314.0 (181.0-472.0) | 0.0043  |
| <140                             | 33/152 (22%)        | 28/121 (23%) | 5/31 (16%)      | 0.4724  |
| ≥140                             | 119/152 (78%)       | 93/121 (77%) | 26/31 (84%)     |        |
| **Prothrombin time, s**          | 13.9 (13.0-15.9)    | 16.2 (15.2-17.8) | 13.5 (13.0-14.6) | < 0.0001|
| **Activated partial thromboplastin time, s** | 33.4 (29.5-39.4) | 39.2 (32.4-49.8) | 13.5 (13.0-14.6) | < 0.0001|
| **INR**                          | 1.09 (1.0-1.29)     | 1.11 (1.0-1.3) | 1.05 (1.0-1.15) |        |

Data are median (IQR) or n/N (%), where N is the total number of patients with available data. P values are expressed by comparing patients who depend on intensive care unit (ICU) and ward care (no ICU care), using the χ² test, Fisher's exact test, t test, or Mann-Whitney test. Source: Authors.
Figure 2. Analysis of the total leukocyte count compared between the COVID Intensive Care Unit (ICU-COVID) and the COVID Infectious Parasitic Disease Unit (IDP-COVID).

![Leukocytes](Image)

Source: Authors.

As shown in Table 3, a prominent finding in most critically ill patients admitted to the COVID-ICU was lymphopenia 90.9%, whereas in the IDP-COVID patients 58.1% had lymphopenia, which may reflect the dominance of neutrophilia 80% ICU-COVID and 69% IDP-COVID (Figure 3). This abnormality agrees with published studies, as lymphopenia occurs in 35%–83%, as does neutrophilia. Another study revealed a similar pattern with more severe cases with higher neutrophils (4.3 × 10^9/L vs 3.2 × 10^9 /L; P < 0.001), lower lymphocyte counts (0.8 vs 1.0 × 10⁹/L; P < 0.001), higher ratio of neutrophils to lymphocytes (5.5 vs 3.2; P < 0.001), as well as lower percentages of monocytes, eosinophils, and basophils (Agbuduwe e Basu, 2020).

Figure 3. Analysis of lymphocyte counts in comparison between the COVID Intensive Care Unit (ICU-COVID) and the COVID Infectious Parasitic Diseases Unit (IDP-COVID).

![Lymphocytes](Image)

Source: Authors.
When analyzing the coagulation screening tests Prothrombin Time (PT) and Activated Partial Thromboplastin Time (APTT), an increase in these values can be observed in patients from the COVID Intensive Care Unit (ICU-COVID), with an average of 16.2%, which is not observed in less severe patients in the ward, average 13.5% (table 3). Studies already report that some patients with COVID-19 have increased prothrombin time (PT) along with prolonged activated partial thromboplastin time (APTT). Adding to these abnormalities, elevated D-dimers further support the occurrence of coagulopathy and is an important indicator of disease progression (Pourbagheri-Sigaroodi et al., 2020).

In the data presented in Table 1, it is noted that thrombocytopenia is present in 23% of patients hospitalized in the COVID-ICU and 16% in the IDP-COVID, corroborating findings in the literature, in which a small part of the reported cases with COVID-19 had mild to severe thrombocytopenia (Agbuduwe & Basu, 2020). Several comorbidities may be associated, mainly with thromboembolic phenomena, contributing to thrombocytopenia (Monteiro et al., 2021).

When analyzing the hematimetric indices, it was observed that 77.8% of the women and 70.1% of the men of the ICU-COVID presented anemia, when correlating these findings with the patients of the IDP-COVID, the predominance of anemia was only in women (70%) (Table 4). Recent data show that patients with COVID-19 tend to have decreased levels of hemoglobin, indicating the presence of anemia, and pathologically increased levels of ferritin. Anemia may be the result of iron-restricted erythropoiesis due to changes in iron metabolism. Increased ferritin levels may be indicative of a strong inflammatory reaction in COVID-19 or related to viral entry into the human body and its impact on iron metabolism (Taneri et al., 2020).

| Hemoglobin, g/L | ICU (n=121) | Infirmary (n=31) | P value |
|-----------------|-------------|-----------------|---------|
| Men             |             |                 |         |
| <13             | 47/67 (70.1%) | 7/21 (33.3%)    | 0.0042  |
| ≥13             | 20/67 (29.9%) | 14/21 (66.7%)   |         |
| Women           |             |                 |         |
| <11.5           | 42/54 (77.8%) | 7/10 (70%)      | 0.6874  |
| ≥11.5           | 12/54 (22.2%) | 3/10 (30%)      |         |

Data are median (IQR) or n/N (%), where N is the total number of patients with available data. P values are expressed by comparing patients who depend on intensive care unit (ICU) and ward care (no ICU care), using the χ² test or Fisher's exact test. Source: Authors.

4. Conclusion

Given the exposure, it was concluded that two of the 152 patients analyzed in this study, 58% were of the male gender and the average age in relation to their physical age was 61.5 years, being possible to identify that the children and adolescents present less severe symptoms, evidenced by minor admissions number. Not a patient with COVID-19, or leukogram is highly variable, leukopenia, leukocytes, as well as results within two reference values may occur, depending, therefore, on the phase of the pathology. This variation may be associated with the adaptation of the bone marrow to infection and normalization of cell production, as well as with the inflammatory stimulus of pain. Neutrophilia and lymphopenia observed in no study, is a striking feature in COVID-19, both associated with prognosis and severity of infection.

In the coagulopathies observed, two parameters of the coagulogram were elongated and events of a thrombolytic nature were more frequent. This clinical condition is associated with inflammation because inflammatory cytokines generate
abnormal responses to clot formation and platelet hyperactivation.

Likewise, future studies can be carried out, using other parameters such as the hemoglobin content of two reticulocytes, fraction of immature platelets, or D-dimer and fibrinogen, with the objective of evaluating the clinical evolution of the patient in relation to the pathology gravity.

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