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Relationship of caspase-3 and calpain-2 proteins and some hematological parameters in adult acute myelogenous leukemia patients in Baghdad City

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Abstract:

The study was conducted to find out the effect of caspase-3 and calpain-2 protein and some blood parameters on acute myelogenous leukemia patients for adults in the city of Baghdad. The number of samples in the study was 30 samples for the acute myelogenous leukemia group. Patients before treatment, a group of patients during treatment, a group of patients after the end of treatment, and 10 outwardly healthy control samples. From Baghdad Teaching Hospital in the Medical City and some medical laboratories for outpatient clinics for the period between September 1, 2022 and February 30, 2023. Physiological and biochemical tests were conducted at the International Center for Research and Development in Baghdad. The study summarized that there is a significant increase in the concentration of caspase-3 protein and the level of calpain protein. 2 in all consecutive patient groups compared to the control group at a significant level $p \leq 0.05$, While there is a significant decrease in the level of the number of red blood cells, the level of hemoglobin concentration, and the percentage of blood platelets in all groups of patients, respectively, compared to the control group at a significant level $p \leq 0.05$, and there was no statistically significant increase or decrease in the number of white blood cells in the serum of the patient group Before treatment, knowing that there was a slight increase in the number of white blood cells when compared with the healthy control group, while there was a significant decrease in the number of white blood cells in the patient group during treatment and after treatment when compared with the healthy control group and the group of patients before treatment. At a significant level $p \leq 0.05$.

Keywords: Leukemia , Myeloid , Caspase-3 , Calpain-2 .

INTRODUCTION

Wilhelm Ebstein 1889 used the description of acute leukemia to describe a specific disease that was characterized by rapid and severe as the clinical description was attributed to this scientist, as this was the first time that leukemia was divided in to acute and chronic clinically, Nenmann also proposed the term myeloid, because white blood cells are produced from the bone marrow. In 1800, Mosler in vented a technique for formally examining the bone marrow to diagnose leukemia. As for myeloblasts which are malignant cells in AML, they were identified by Naegeli, who divided leukemia in to two types: myeloid leukemia and lymphocytic leukemia (Bukša,2021). Acute myelogenous leukemia (AML) accounts for about 80% of acute leukemia cases and is the most common in adults, as the average age for best diagnosis is 67 , in addition to the incidence rate per 100,000 for men or women is 3.7, according to surveillance, epidemiology , and the end result (SEER) (SEER,2013) In the United States, about 14,590 new cases are diagnosed annually (American Cancer Society, 2013). About 58% of cases were diagnosed at age 65 or older, which in dicates that AML is more common in adults (Howlader *et al* ., 2019). Acute myelocytic leukemia is a disease that arises from immature myeloid cells called blast cells so AML is a hematological malignancy, age and disease biology act as good factors for prognosis, AML is fatal if left untreated (Short *et al* ., 2018).

Materials & Method:

Experiment Designs:

Forty samples were collected divided into 30 samples for a group of patients with acute myelogenous leukemia, whose ages ranged between (15-75) for males and females. Baghdad Teaching Hospital in the Medical City and some medical laboratories for outpatient clinics for the period between September 1, 2022 and February 30, 2023. Physiological and biochemical tests were conducted at the International Center for Research and Development in Baghdad.

Sample Collection :

A questionnaire form was designed for the study and included all the required information. (5) milliliters of venous blood were drawn from patients with acute myelogenous leukemia and healthy people after sterilizing the withdrawal area with 70% ethyl alcohol using a sterile medical syringe and placing it in test tubes free of anticoagulant for the purpose of obtaining The serum was collected in a centrifuge at a speed of (3000) revolutions / 10 minutes, and then the serum was withdrawn by a fine pipette and placed in clean and sterile laboratory tubes with tight lids and then kept in the freezer at a temperature of (-20) Cº until physiological and biochemical tests are performed.

Physiological and biochemical variables

Serum caspase-3 and calpain-2 were measured using a colorimetric enzymatic method using a kit from Fine Test , China by device Huma Reader Hs . Complete blood picture analysis (CBC) to evaluate some components of the blood, including the number of white blood cells (WBCs), red blood cells (RBCs), hemoglobin (Hb), and platelets (PLTs).

Statistical Analysis

The well- known statistical system (Graph Pad prism ver. 5) was adopted, and the analysis of variance table one – way anova (by Tukey´s multiple comparisons test) was used for the comparison among subdivided groups in the measured parameters. The results were expressed as (Mean ± Stander Error) .by using SPSS program (Version v 25).

The Results and Discussion:

Concentration level of caspase-3 proteins in AML patients

The results of the study showed in Table (1) that there was a significant increase at a significant level $p \leq 0.05$ in the level of concentration of caspase-3 proteins in the blood serum of all groups of patients (before treatment, during treatment, after treatment) when compared with the healthy control group. While a significant decrease was observed in the concentration of caspase-3 proteins in two groups during treatment and after treatment compared with the group before treatment, and a significant decrease was also observed in the concentration of caspase-3 proteins in group after treatment compared with the group during treatment .

Table (1) Concentration level of caspase-3 proteins in AML patients

Groups Standards	control N=10	before treatment N=10	during treatment N=10	after treatment N=10
Caspase−3	1.776±0.025 d	8.798±0.061 a	6.377±0.422 b	3.615±0.264 c

- Values represent mean ± standard error = Mean ± S.E

- The different letters (horizontally) indicate that there are significant differences at a significant level $P \leq 0.05$.

The results of the current study came close to a number of studies that focused on the role of caspase-3 proteins in the process of programmed cell death. As a good factor in diagnosing newly infected patients with chronic myeloid leukemia, and this is consistent with the current study, Jamal (2019) indicated that there is a significant increase in caspase-3 proteins in breast cancer patients compared to healthy subjects. He also indicated that the activation of caspase-3 proteins without appropriate control leads to To treatment resistance and apoptosis does not occur Li Y et al. (2015) found that caspase-3 was not only used as an indicator to predict treatment response, but as a therapeutic target directly or through other apoptosis-related proteins.

Concentration level of Calpain-2 proteins in AML patients

The results of the study showed in Table (2) that there was a significant increase at a significant level $P \leq 0.05$ in the level of concentration of calpain-2 proteins in the blood serum of all groups of patients (before treatment, during treatment, after treatment) compared with the healthy control group, while a decrease was observed Statistically significant in the concentration of calpain-2 proteins in the group during treatment compared to the group before treatment and there was also a significant decrease in the concentration of calpain-2 proteins in the group after treatment compared to two groups before treatment, during treatment .

Table (2) Concentration level of Calpain-2 proteins in AML patients.

Groups Standards	control N=10	before treatment N=10	during treatment N=10	after treatment N=10
Calpain−2	1.755±0.024 d	3.798±0.074 a	3.249±0.141 b	2.874±0.076 c

- Values represent mean ± standard error = Mean ± S.E
- The different letters (horizontally) indicate that there are significant differences at a significant level P ≤ 0.05 .

Effect of acute leukemia on some hematological parameters

Table (3) Effect of AML on some hematological parameters .

Groups Standards	control N=10	before treatment N=10	during treatment N=10	after treatment N=10
WBCs	8.547±0.143 a	9.646±1.069 a	3.245±0.398 b	5.205±0.557 b
RBCs	5.315±0.083 a	3.096±0.151 b	2.969±0.145 b	2.746±0.141 b
Hb	14.150±0.264 a	9.778±0.454 b	8.482±0.341 b	7.564±0.341 bc
PLTs	253.75±11.35 a	142.50±5.62 b	151.80±7.83 b	66.40±2.61 c

- Values represent mean ± standard error = Mean ± S.E
- The different letters (horizontally) indicate that there are significant differences at a significant level P ≤ 0.05 .

Effect of AML on white blood cells

The results of the study showed in Table (3) that there was no statistically significant increase or decrease at a significant level P ≤ 0.05, knowing that there was a slight increase in the percentage of white blood cells (WBCs) in the blood serum of the patients group before treatment when compared with the control group While there is a significant decrease in the group of patients during treatment and after treatment when compared with the healthy control group and the group of patients before treatment, we did not find a significant decrease or increase in the number of white blood cells statistically, noting that there was a slight increase in the group after treatment compared to group during treatment .

The results of the study agreed with what Maryam (2022) reached, where the results indicated that there was no statistically significant difference in the number of white blood cells for patients with acute myeloid leukemia when compared with the control group, Rasha (2020), where she indicated that there was a significant decrease in The total white blood cell count (WBCs) in the group of acute leukemia patients during chemotherapy treatment in all patients when compared with the healthy control group, as between Humam (2018) that there is a significant decrease in the total white blood cell count in the group of acute lymphoblastic leukemia patients during chemotherapy in all patients when compared with a healthy control group.

While the results of the study did not agree with the findings of Taisir (2021), which indicated that there was a significant increase in the total white blood cell count in the group of acute leukemia patients before chemotherapy when compared with the healthy control group, and the results of the study did not agree with what was indicated by Thamer (2019) that there was a significant increase in the total white blood cell count in the group of patients with myeloid leukemia before chemotherapy when compared with the healthy control group.

Effect of AML on red blood cells

The results of the study showed in Table (3) that there was a statistically significant decrease at a significant level P ≤ 0.05 in the number of red blood cells (RBCs) in the blood serum of all groups of patients (before treatment, during treatment, after treatment) when compared with the control group While there was no statistically significant increase or decrease, knowing that there was a slight decrease among the patients' groups in the number of red blood cells.

The results of the study agreed with the findings of Taisir (2021) and Taiba (2020), as each of them indicated that there was a significant decrease in the number of red blood cells compared with the healthy control group at the early diagnosis stage before treatment and during chemotherapy when compared with the healthy control group. This decrease in the ability of myeloid cells to differentiate leads to the accumulation of blood cell blasts of abnormal shapes in the bone marrow and peripheral blood, causing multiple systemic phenomena such as anemia (Alter, 2014) .

Effect of AML on blood hemoglobin

The results of the study showed in Table (3) that there was a statistically significant decrease at a significant level P ≤ 0.05 in the percentage of hemoglobin (Hb) concentration in the blood serum of all groups of patients (before treatment, during treatment, after treatment) when compared with the control group While there is no statistically significant increase or decrease, knowing that there was a slight difference between before treatment and during treatment in the level of hemoglobin in the blood, but there is a significant decrease in the group of patients after treatment when compared with the group of patients before treatment and during treatment .

The results of the study agreed with a number of studies whose results indicated that there was a significant decrease in the level of hemoglobin concentration in patients with myeloid leukemia, compared to the healthy control group (Thamer, 2019; Razij *et al*., 2018), and these results were also consistent with the results of Al-Mashhadani *et al* (2011), which It showed a significant decrease in the level of hemoglobin concentration in the blood in 40% of patients with myeloid leukemia .

Effect of AML on Platelets

The results of the study showed in Table (3) that there was a statistically significant decrease at a significant level P ≤ 0.05 in the percentage of platelets (PLTs) in the blood serum of all patient groups (before treatment, during treatment, after treatment) when compared with the healthy control group While there is no statistically significant increase or decrease, knowing that there was a slight difference between the group of patients before treatment and during treatment in blood

platelets, but there is a significant decrease in the group of patients after treatment when compared with the group of patients before treatment during treatment .

The results of the study were consistent with what Rasha (2020) and Taiba (2020) reached about a clear significant decrease in the platelet count of acute leukemia patients before and during chemotherapy for acute leukemia patients, because this type of disease grows and worsens very quickly and may threaten life to the point Large, as this type begins with the bone marrow to produce large numbers of immature white blood cells called (blasts), which enter the bloodstream, and these immature cells work quickly to crowd out normal cells in the bloodstream and do not do their job in fighting infection or stopping bleeding, what It makes the body very weak and vulnerable (Greaves, 2018) .

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