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# Blood variations of male albino mice treated with the antibiotic (Cefoperazone)

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# Blood variations of male albino mice treated with the antibiotic (cefoperazone)

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**Abstract.** The effect of the antibiotic cefoperazone was examined. The experiment was conducted on white male mice. The experiment was divided into three groups, 11-12 mice were allocated to each group to conduct different experiments, It was injected Intraperitoneal into the groups for 7 consecutive days. The control group was given distilled water throughout the experimental days. The second group was given the antibiotic cefoperazone at a dose of 500 mg/kg/day. The third group was given the antibiotic cefoperazone 800 mg/kg/day. Cefoperazone affects blood components, causing a noticeable increase in the total number of blood cells, a slight increase in the number of red blood cells, and an increase in hemoglobin and hematocrit. It also caused an increase, but it did not constitute a significant difference in blood counts. As for white blood cells, there was a significant difference for lymphocytes and eosinophils, and there was an increase in neutrophils, while for Basophil cells there was no significant increase.

**Keywords:** cefoperazone. red blood cells. white blood cells, CBC, Lymphocyte, Neutrophile



## INTRODUCTION

During the past decade, studies conducted on antibiotics mainly targeted the activity of Gram-negative and Gram-positive bacteria, while maintaining international drug safety standards. Previous studies have shown that Cefime antibiotics do not lead to clear effects on the immune system, but it has been shown that repeated injections of Cefime antibiotics in large quantities lead to different immunological effects, whether in humans or animals [1]. Cefoperazone is a semi-synthetic third-generation cephalosporin used to treat respiratory tract infections, bacterial septicemia, skin infections, intra-abdominal infections and urinary tract infections. Cefoperazone acts on intestinal bacteria, Pseudomonas aeruginosa, bacilli, *Staphylococcus aureus*, and others. It has a broad spectrum against bacteria. [2, 3]. Bacterial flora is found in both humans and animals and are also known as opportunistic pathogens [4]**.** For example, human skin and mucous microbes contain Staphylococcus aureus, which is common but opportunistic and causes major diseases and infections [5]. Approximately 20-25% of healthy people are carriers of Staphylococcus aureus bacteria [6]. There is also Candida albicans, a yeast found as part of the normal microbial flora in most healthy humans. It is found on mucous surfaces as a usually harmless colonizer [7]. In cases of weak immunity, it can cause harm, there are more than 100 different species, but only a few can infect humans. For example, Candida spp is of continuing medical importance, and may also be life-threatening with systemic infections and chronic mucocutaneous infections. Patients with weak immunity [8]. While the study conducted on some bacterial isolates from burn wound infections showed their high sensitivity against some beta-lactam antibiotics [9]. On the other hand, many studies have found that strains of bacteria producing beta-lactamases are more common and that Staphylococcus aureus bacteria have increased their resistance to beta-lactam antibiotics. Many cephalosporins and penicillin's have become ineffective for treatment due to beta-lactamase enzymes [10]**.** Cefoperazone works by penetrating the outer layers of the bacterial cell wall, so it acts as a bactericide [11]. Matsubara, (1980) mentioned that Cefoperazone has antimicrobial activity because it has an affinity for penicillin-binding proteins. It has a high ability to bind to penicillin-binding proteins 1B, 1A, and 2A, which contribute to the initiation of cell wall elongation [12]. Third-generation cephalosporins selectively

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inhibit the synthesis of mucin polypeptides in the bacterial cell wall. It is unique in its ability and the presence of the beta-lactamase enzyme to remain stable [13]. All cephalosporins are expected to cross the placenta and are thought to be excreted into breast milk [14]. all non -steroidal anti -inflammatory drugs are offered from the body of the liver and kidneys [15].

## MATERIALS AND METHODS

### Experimental animals:

Thirty-five male mice were obtained from the Iraqi Ministry of Health/National Center for Pharmaceutical Control and Research. At 12 weeks of age, animals were fed a regular diet and water ad libitum in a controlled room at 23°C humidity .Whole blood was collected and placed in a tube containing clean ethylenediaminetetraacetic acid (EDTA) to measure the number of white blood cells (WBCs) and differential cell counts [16]

*Animal Breeding:*

This study was conducted on healthy, adult male white mice. The mice were cared for throughout the duration of the study in terms of cleanliness, ventilation, lighting, temperature, and continuous provision of feed and water to the mice [17].

*Cefoperazone Antibiotic*

Cefoperazone from Smart (New Delhi, India) is a commercial product.

*Treatments*

Initially, 11-12 mice were allocated to each group to conduct different experiments. The first and second groups were injected intraperitoneally with a dose of 500 mg/kg/day and 800 mg/kg/day, respectively, of the antibiotic, while the third group was injected with distilled water (control). The experiment lasted for seven days. At the end of the experiments, blood was drawn from the heart for examination.

*Statistical Analysis*

The Statistical Analysis System- SAS (2018) program was used to detect the effect of different groups in study parameters. The least significant difference- LSD was used to significantly compare between means. Estimate of the Correlation coefficient between different parameters in this study the complete blood count [18].

**3. Results and Discussion**

Intraperitoneal injection of cefoperazone was therapeutic at a dose of (500 and 800 mg/kg body weight) in male mice, where the antagonist cefoperazone was given at doses of 180 and 360 mg/kg body weight for 7 days.

Table 1: Comparison between different groups in WBC, RBC, HGB and HCT

| Group | Mean ±SE | | | | |
|---|---|---|---|---|---|
| | WBC ($x10^9$/L) | RBC ($x10^{12}$/L) | HGB (g/dl) | HCT (%) | PLT ($x10^9$/L) |
| Control | 5.64 ±0.07 b | 7.91 ±0.85 | 12.87 ±0.37 a | 38.27 ±0.61 a | 795.25 ±84.27 |
| Conc. 1 | 9.82 ±0.67 a | 7.20 ±0.60 | 10.67 ±0.90 b | 35.50 ±3.04 ab | 912.00 ±48.53 |
| Conc. 2 | 6.87 ±0.25 b | 7.97 ±0.23 | 11.00 ±0.57 ab | 32.52 ±1.60 b | 877.00 ±36.71 |
| LSD | 1.334 ** | 1.970 NS | 2.096 * | 5.452 * | 191.88 NS |
| P-value | 0.0002 | 0.631 | 0.0487 | 0.0491 | 0.406 |
| Means having with the different letters in same column differed significantly.<br>* ($P \leq 0.05$).<br>** $p < 0.01$ versus saline-treated controls. | | | | | |

Table 2 presents a comparison between the different groups in terms of white blood cell (WBC) differential. The table shows the mean ± standard error (SE) values for the number of neutrophils, lymphocytes, monocytes, eosinophils, and basophils in each group..

Table 2: Comparison between different groups in the differential of White blood cells.

| Group | Mean ±SE | | | |
|---|---|---|---|---|
| | Neutrophil | Lymphocyte | Eosinophil | Basophil |
| Control | 0.108 ±0.004 b | 5.39 ±0.11 c | 0.00 ±0.00 c | 0.055 ±0.006 |
| Conc. 1 | 0.195 ±0.034 a | 8.79 ±0.28 a | 0.165 ±0.01 a | 0.045 ±0.011 |
| Conc. 2 | 0.160 ±0.012 ab | 6.50 ±0.24 b | 0.050 ±0.009 b | 0.060 ±0.009 |
| LSD | 0.0691 * | 0.718 ** | 0.025 ** | 0.0282 NS |
| P-value | 0.0498 | 0.0001 | 0.0001 | 0.499 |
| Means having with the different letters in the same column differed significantly.<br>* (P≤0.05), ** (P≤0.01). | | | | |

The results that were conducted to analyze the average values of the white blood cell (WBC), red blood cell count (RBC), hemoglobin concentration (HGB), hematocrit level (HCT), and platelets (PLT) showed between three different groups: control, and the two concentrations. the first and the second. The results are presented in Table 1, where mean values with standard errors (SE) are shown..

1- white blood cell (WBC): In the control group, the average number of white blood cells was (5.64 x $10^9$)/liter, which is significantly lower compared to the two groups treated with the antibiotic, as the first concentration represented the highest value, as it rose to (9.82 x $10^9$)/liter, while the second concentration: It decreased from the first concentration, but remained higher than the control group, so that its value represented (6.87 x $10^9$)/litre, as shown by different letters in the same column as in Figure (1)

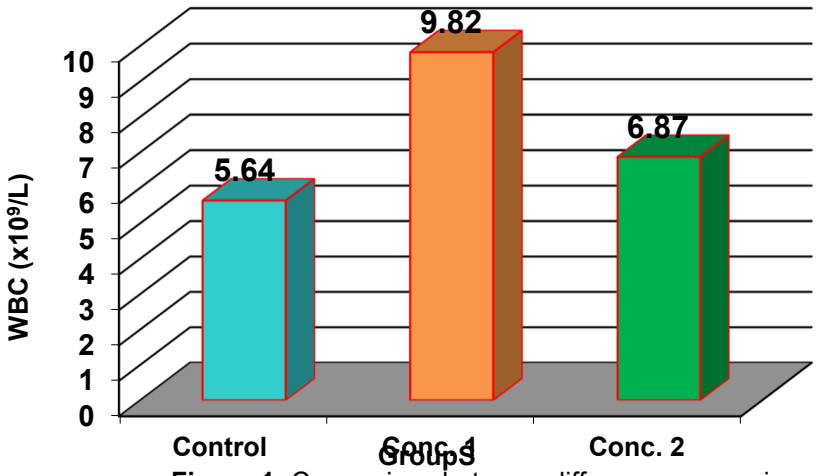


**Figure 1:** Comparison between difference groups in WBC

2- Red blood cell count (RBC): The average number in the control group was 7.91 x 1012/L. There was a slight increase, but there was no significant difference from the rest of the treated groups as shown in Figure (2).

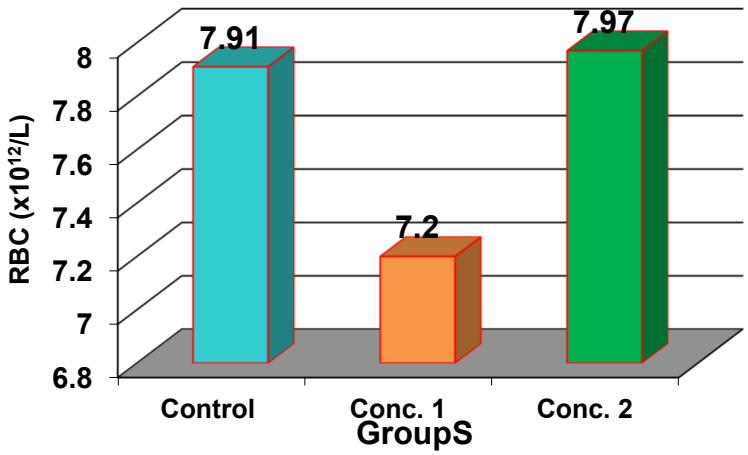


**Figure 2:** Comparison between difference groups in RBC

3- Hemoglobin) HGB) : the average hemoglobin concentration in the control group increased to 12.87 g/dL, which is much higher compared to the two concentrations for the control groups as shown in Figure(3).

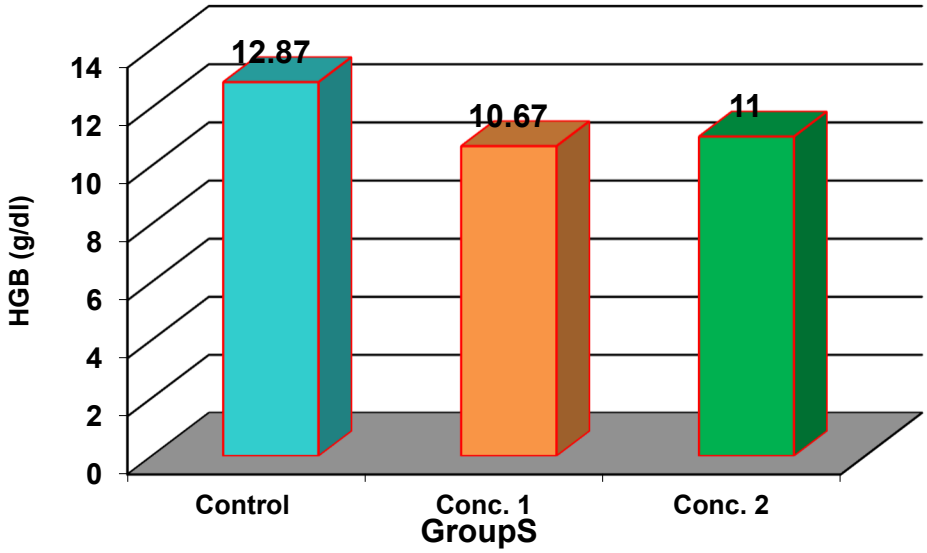


**Figure 3:** Comparison between difference groups in HGB

4- hematocrit level (HCT): the mean HCT level in the Control group was 38.27%, which was significantly higher compared to the other two groups as shown in Figure(4).

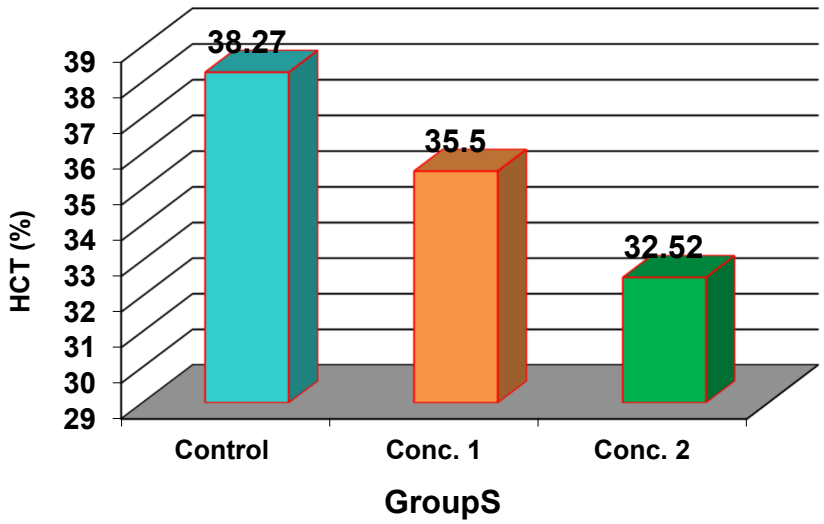


**Figure 4:** Comparison between difference groups in HCT

5- platelets (PLT) : In the control group, the average number was (795.25 x $10^9$)/liter, which is lower compared to the two groups treated with the antibiotic, as the first concentration represented the highest value, as it rose to (912.00) ×$10^9$)/L. As for the second concentration: it decreased from the first concentration, but remained higher than the control group, so that its value represents (877.00 ×$10^9$)/L, as in Figure 1) as in Figure (5)

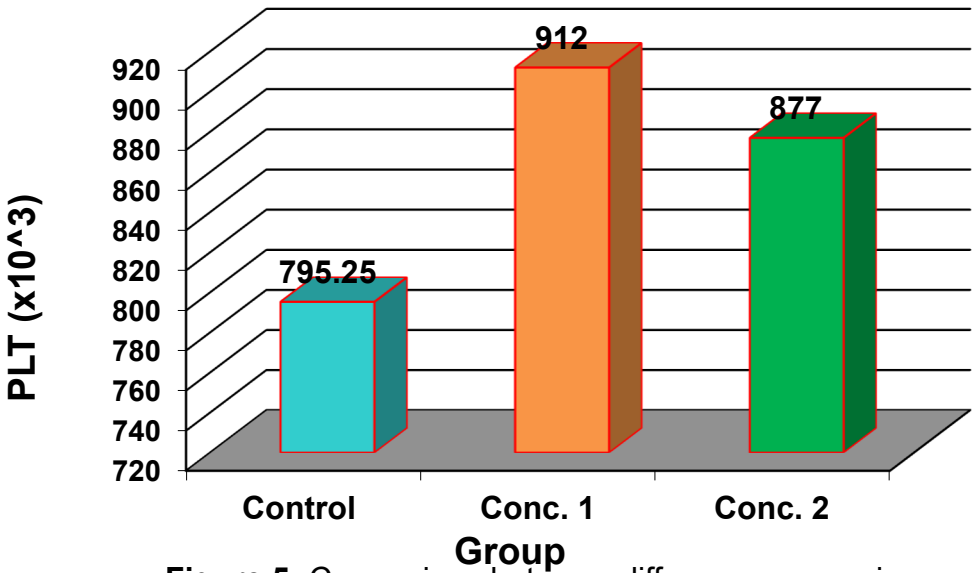


**Figure 5:** Comparison between difference groups in PLT

6- Table 2 presents a comparison between the different groups in terms of white blood cell (WBC) differential. The table shows the mean ± standard error (SE) values for the number of neutrophils, lymphocytes, monocytes, eosinophils, and basophils in each group..

The mean values in the control group were 0.108 ± 0.004 for neutrophils, 5.39 ± 0.11 for lymphocytes, 0.00 ± 0.00 for eosinophils, and 0.055 ± 0.006 for basophils. In comparison, in the group labeled "Conc. 1," the mean values were significantly higher for neutrophils (0.195 ± 0.034), lymphocytes (8.79 ± 0.28), and eosinophils (0.165 ± 0.01), but remained the same for basophils (0.045). ±0.011). The "Conc.2" group also showed that the mean values were found

to be lower than Conc. 1 for neutrophils (0.160 ± 0.012) and lymphocytes (6.50 ± 0.24) but similar for eosinophils (0.050 ± 0.009) and basophils (0.060 ± 0.009).
The p values for each comparison were. Neutrophils ($p = 0.0498$) and lymphocytes ($p = 0.0001$) showed significant differences between groups, while eosinophils ($p = 0.0001$) and basophils ($p = 0.499$) did not show significant differences. .
The results highlight differences in white blood cell differentiation between different groups Cefoperazone is a broad-spectrum cephalosporin antibiotic that was first developed in the 1970s. It is commonly used to treat a variety of bacterial infections, including pneumonia, sepsis, and urinary tract infections. The drug works by inhibiting cell wall synthesis in susceptible bacteria, leading to their death or inability to replicate.
The use of broad-spectrum antibiotics to treat infectious bacteria diseases to improve human health [19, 20]. However, heavy use of these antibiotics can lead to significant changes in the gut microbiome [21, 22] . Gu , (2020) mentioned that Exposure to β-Lactam Antibiotics on Mice for 4 days reduced the alpha and beta diversity of intestinal bacteria and produced an increase in the blood for inflammatory cytokines [23] . The changes persisted for a long time, but the bacterial community tended to return to its normal level after stopping treatment. The effect of four antibiotics on immunity and intestinal microbes was evaluated in mice, and it was concluded that short-term use on healthy mice could lead to disruption of the diversity and biomass of intestinal microorganisms, leading to long-term effects resulting in inflammatory response and the deficient resilience of microbiota[24].
After reviewing the literature and scientific research, I did not find research conducted on mice. On the other hand, a study was conducted on humans. Drug-induced hemolytic anemia (DIHA) is a rare drug complication most commonly caused by second- and third-generation cephalosporins. Cefoperazone/sulbactam is a combination preparation of cefoperazone, a third-generation cephalosporin, and sulbactam sodium, a beta-lactamase inhibitor) Iakovlev VP).
Another study indicated that it has been repeatedly reported that cefoperazone/sulbactam may lead to abnormal coagulation function resulting from it. However, only two reports of DIHA associated with cefoperazone/sulbactam were available. It indicated the presence of a case of abdominal infection in a patient who was prescribed treatment with intravenous cefoperazone/sulbactam (3 g, q. 12 hours) and resulted in an adverse reaction, namely hemolytic anemia. As treatment was discontinued, the condition began to improve and the negative effects decreased The research that took place about a 93 -year -old woman who had abdominal pain, nausea and vomiting for 3 days with fever at 38.5 ° C for two days. After accepting the hospital, its vital signs were normal. The full number of blood showed the number of white blood cells (WBC) 11.72 x 109/l (normal 3.5-9.5 x 109/l), the number of neutrophils (NE) 86.0 % (normal 40 % - 75 %), the number of red blood cells (RBC (3.24 x 1012/L) 3.8-5.1 x 1012/L), hemoglobin (HB) 110 g/l (normal 115-150 g/l), hematocrit (HCT) 32.8 % (normal 35-45 %) and platelet (PLT) 225 × $10^9$/L (normal 125–350 × $10^9$/L). It has been diagnosed with abdominal infection, abnormal liver function, jaundice and post -bile. On the day of acceptance, it was given by the doctors of Sevroberzon/Solbachamam as an anti -infection treatment (3.0 g, Q12H, IVGTT [Guttae intravenously]. On the third day, RBC, HB and HCT decreased a little bit and PLT showed a trend of fluctuation, which corresponds to our study on cefoperazone, and values were 3.10 x 1012/L, 108 g/l, and 31.8 %, respectively. After one month of anti -infection therapy, the infection improved, WBC and NE also fell. Meanwhile, RBC, HCT and HB continued to decline. The results of the laboratory test excluded the possibility of anemia caused by anemia, iron anemia or other types of anemia, and the tomography did not show any bleeding in the abdomen, so it was diagnosed with hemolytic anemia on the day 34. Medicines given before a decrease in RBC, HCT and HB (on the first day and day 2) are cefoperazone/sulbactam, omeprazole for injections, phosphatidil colin, glutamine, nadroprin calcium, aspolol, and potassium chloride. cefoperazone/sulbactam has been given only 34. At the same time, we searched for stickers and considered that it might be associated with cefoperazone/sulbactam. We stopped cefoperazone/sulbactam immediately on the day 34. Although anemia was 3 degrees as evaluated. Because we considered it a reaction to drugs, we stopped the medicine for monitoring. WBC was on the height while it is compatible with our studies, After pulling cefoperazone/sulbactam, RBC, HCt and HB levels recovered and returned to normal after 20 days[25].
The antibiotic caused changes in the shape and structure of red blood cells, including increased fragility and altered hemoglobin content. Side effects of cefoperazone/sulbactam include diarrhea, abdominal pain, bruising, congestion of the skin and mucous membranes, bleeding, blood clotting disorders, blood in the urine, and hemoptysis) [26] . Activated partial thrombin time (APTT), prolongation of prothrombin time (PT), and coagulation disorders have also been shown. Cefoperazone is characterized by the presence of the N-methylthiotetrazolium side chain, the molecular structure of which is similar to the chain found in glutamate. In liver microsomes, glutamate carboxylase competitively binds to vitamin K1, forming a complex. The defect in the clotting process within the body is the result of the effect of this structure on the synthesis of clotting factors, as clotting factors depend on the group of vitamins II, VII, IX, and X . Cephalosporins also contain a COOH group located on the 7C atom substituent, which causes bleeding because

it hinders platelet aggregation. [27]. In another study, it was shown that 25.8% of patients treated with cefoperazone/sulbactam for 3-12 days showed a blood clotting defect [28].
Due to the morphological changes, the red blood cells' ability to transport oxygen throughout the body was significantly reduced. Cefoperazone was associated with the development of hemolytic anemia. Hemolytic anemia results in a decreased ability to carry oxygen due to low levels of hemoglobin and the number of red blood cells. To ensure that adequate oxygen reaches the tissues during treatment, hemoglobin levels must be monitored [29][30] .
On the other hand, our study agreed with a study on mice that showed that cefoperazone increased IgM production in serum. In addition, cefoperazone has never been shown to activate germinal centers in the spleen. Thus, the discrepancy between the data on the immunostimulatory activity of cefoperazone reported by Gilesen and Pusztay-Marcus and our data may be due to differences in the assay methods between the reverse and direct PFC and in the rat strains used. The clinical relationship between the immunostimulatory and antibacterial activities of ceftisol should be investigated further, as the antibiotic is likely to require much higher doses than usual to demonstrate its immunostimulatory activity.

## CONCLUSION

It was clearly obvious that Cefoperazone caused morphological changes in the red blood cells' which led to affect their ability to transport oxygen throughout the body Cefoperazone was associated with the development of hemolytic anemia. Hemolytic anemia results in a decreased ability to carry oxygen due to low levels of hemoglobin and the number of red blood cells. To ensure that adequate oxygen reaches the tissues during treatment, hemoglobin levels must be monitored. Also, Cefoperazone found to elict and improved the immunity of the body by increased the numbers of WBC. The study found a significant difference in white blood cell count, Hgb concentration, and HCT level between the three groups. However, no statistically significant differences were observed for the number of red blood cells and the number of platelets. These results indicate that the concentration of the antibiotic cefoperazone affected blood cell counts and hemoglobin levels in the groups tested.

## REFERENCES


1. J. Descotes , " Immunotoxicology of Drugs and Chemicals, ed 2," ***Amsterdam, Elsevier,*** vol.1 ,no1, pp.31-48 , 1988.
2. S. Mitsuhashi , S . Minami, N. Matsubara , A . Yotsuji , S. Kurashige , I "Saikawa ,. In vitro and in vivo antibacterial activity of cefoperazone" ***Clinical Therapeutics***, vol.1 ,no3 ,1980 ,pp. 1-13,1980, PMID: 6446381.
3. A.E.Czeizel , M. Rockenbauer, H.T. Sorensen , and J. Olsen, "Use of cephalosporins during pregnancy and in the presence of congenital abnormalities: a population-based, case-control study" ***American journal of obstetrics and gynecology***, vol.*184*,no.6, pp.1289-1296 , 2001, https://doi.org/10.1016/S0002-9378(02)70079-3 .
4. A.W. Anwer, G.M. Saleh , M.E.Ahmed, "Antimicrobial Susceptibility of Enterococcus faecium Isolated from Bee-gut on PhzMGene of Pseudomonas aeruginosa Isolates from Infected Wounds**"** ***bn Al-Haitham Journal for Pure and Applied Sciences (IHJPAS)***, vol.37, no.2,pp.39-48, **2023, https://doi.org/10.30526/37.2.3379 .**
5. A.M. Abd Zaid, and N.J. Kandala, IRAQI *JOURNAL OF AGRICULTURAL SCIENCES,* Vol.52, No.6, pp. 1356-1364, 2021, 10.36103/ijas.v52i6.1475.
6. A.A.I. Al-Halaq, and N.M. Utba, "Prevalence of Methicillin-resistant Staphylococcus aureus Carrying lukS-lukF Gene in Iraqi Patients with Furunculosis" ***Iraqi Journal of Science***, pp.3323-3329, 2023, http://dx.doi.org/10.24996/ijs.2023.64.7.13 .
7. S.R.M. AL-Rubaie, and S. A. Al-Qaysi, " Detection of Some Virulence Factors in Candida albicans Obtained from Different Clinical Specimens of Iraqi Patients" Departmentof Biology, *Ibn AL-Haitham Journal For Pure and Applied Sciences,* vol.37,no.2,pp.1-11, 2024, doi.org/10.30526/37.2.3315 .
8. K.I.Othman, S.M. Abdullah , and A.M. Majid, " Isolation and Identification Candida spp. from Urine and Antifungal Susceptibility Test" *Iraqi Journal of Science*,vol.59, no.4B , pp.1981-1988 ,2018 , http://dx.doi.org/10.24996/ijs .
9. M. Al-Azzawi, & I. Alkhalifawi, "Detection of Bacteria Causing Burn Infection Isolated from Several Hospitals in Baghdad" *Ibn AL-Haitham Journal For Pure and Applied Sciences.* Vol.36, no.3, pp.1-8 , 2023,doi.org/10.30526/36.3.3090 .
10. E.D. Salman, A.J. AL-Saedi, A.G.M. AL-Kazzaz, and S. S. Yahya, "The Effect of Aqueous Extract and Essential Oils of the Leaves of Eucalyptus incrassate on β-Lactam Resistant Staphylococcus aureus" *Ibn AL-Haitham Journal For Pure and Applied Science*, vol.*27,*no.1, pp.76-82, 2014.

11. W. Zimmermann, 1979 "Penetration through the Gram- negative cell wall: a co – determinant of the efficacy of B- lactam antibiotics" International *journal of clinical pharmacology and biopharmacy*, vol.*17*,no.3,pp. 131–134, 1979, PMID: 107128
12. N.Matsubara, S. Minami, M. Matsuhashi, M. Takaoka, , & S.Mitsuhashi, "Affinity of cefoperazone for penicillin binding proteins*"* *Antimicrobial agents and chemotherapy*, vol.*18,*no.1, pp.195–199,1980, https://doi.org/10.1128/AAC.18.1.195
13. B. A. Cunha, & A. M. Ristuccia, "Third generation cephalosporins" *The Medical clinics of North America*, vol.*66*, no.1,pp. 283–291,1982, https://doi.org/10.1016/s0025-7125(16)31460-2
14. R.Elzoghby, "TERATOLOGICAL AND PATHOLOGICAL STUDIES OF CEFOPERAZONE IN FEMALE ALbino rats" *Lucr ST.USAMV Iasi Med Vet Series*, vol 53,no.12,pp.236-248, 2010.
15. I.M. Mnati, and B.H. Mutlak, "Histopathological Changes in Liver Induced by Piroxicam Administration in Adult Male Albino Mice Mus musculus". *Indian Journal of Forensic Medicine & Toxicology*, vol.14, no.3,pp.1535-1541, 2020.
16. R.A. Salih, , Z.H. Kadri, &, H.M. Al-Abassi "Study of the Immunomodulatory Properties of Pistacia khinjuk Extract on Rats" *INTERNATIONAL JOURNAL OF DRUG DELIVERY TECHNOLOGY,*vol.12,no.2 , pp.918-923, 2022, DOI: 10.25258/ijddt.12.2.79
17. B.Al-Musawiu, and N. Al-Bakri, " Effect of Hydroxychloroquine Drugs on Amino Acids in the Kidneys of White Mice (Mus musculas)" *INTERNATIONAL JOURNAL OF DRUG DELIVERY TECHNOLOGY*,vol. 12,no.4,pp.1590-1594, 2022,DOI: 10.25258/ijddt.12.4.18
18. SAS, "Statistical Analysis System, User's Guide. Statistical" *Version 9.6th ed. SAS. Inst. Inc. Cary. N.C. USA*. 2018
19. K. Nicolaou, S. A. Rigol, "brief history of antibiotics and select advances in their synthesis"*The Journal of antibiotics*, vol.71,no.2, pp.153–184. ,2018, https://doi.org/10.1038/ja.2017.62
20. R.Benkő, , M.Gajdács, M. Matuz, , G. Bodó, , A. Lázár, , E.Hajdú, , E.Papfalvi, , P.Hannauer, P. Erdélyi, & Z. Pető, "Prevalence and Antibiotic Resistance of ESKAPE Pathogens Isolated in the Emergency Department of a Tertiary Care Teaching Hospital in Hungary: A 5-Year Retrospective Survey" *Antibiotics (Basel, Switzerland)*, vol.*9*(9), 624 ,2020, https://doi.org/10.3390/antibiotics9090624 .
21. L. Dethlefsen, D.A. Relman ,"Incomplete recovery and individualized responses of the human distal gut microbiota to repeated antibiotic perturbation" Proceedings of the National Academy of Sciences of the United States of America, vol. 108 ,(Suppl 1), pp.4554–4561, 2011, https://doi.org/10.1073/pnas.1000087107 .
22. J. C. Clemente, L. K. Ursell, L. W. Parfrey, & R. Knight, (). "The impact of the gut microbiota on human health: an integrative view" *Cell,* vol.*148*,no.6, pp.1258–1270 , 2012, https://doi.org/10.1016/j.cell.2012.01.035
23. S. L. Gu, , Y.Gong, J. Zhang, Y. Chen, , Z. Wu, , Q.Xu, , Y.Fang, , J.Wang, , & L. L. Tang, "Effect of the Short-Term Use of Fluoroquinolone and β-Lactam Antibiotics on Mouse Gut Microbiota" *Infection and drug resistance*, vol.13, pp.4547–4558 ,2020, https://doi.org/10.2147/IDR.S281274 .
24. K. Korpela, A. Salonen, L. J.Virta, R. A.Kekkonen, K. Forslund, P. Bork, & W. M.de Vos, "Intestinal microbiome is related to lifetime antibiotic use in Finnish pre-school children" *Nat Communications*, vol.7,no.1,pp.1-8,10410, 2016, doi:10.1038/ncomms10410 .
25. X. M.Sun, L. H. Liu, Q.Wu, & H. G. Wang, "Cefoperazone/sulbactam-induced hemolytic anemia" *Journal of postgraduate medicine*, vol.*69*,no.1, pp.46–49, 2023, https://doi.org/10.4103/jpgm.JPGM_1335_20
26. L. J.Chen, F. Y.Hsiao, L. J. Shen, F. L. Wu, W. Tsay, C. C.Hung, & S. W. Lin, "Use of Hypoprothrombinemia-Inducing Cephalosporins and the Risk of Hemorrhagic Events" *A Nationwide Nested Case-Control Study*, *PloS one* , vol.*11*,no.7,pp,1-11, e0158407,2016, https://doi.org/10.1371/journal.pone.0158407
27. R. S.Wong, G. Cheng, , N. P.Chan, W. S. Wong, & , M. H.NG, "Use of cefoperazone still needs a caution for bleeding from induced vitamin K deficiency" *American journal of hematology*, vol.*81*,no.1,pp. 76-80 ,2006, https://doi.org/10.1002/ajh.20449 .
28. X.Shao, Y. Ren, N. Xie, K.Fan, H.Sun, J. Lu, J. Wei, & X. Tian, "Effect of Cefoperazone/Sulbactam on Blood Coagulation Function in Infected Emergency Department Patients and the Necessity of Vitamin K1 (VK1) Preventive Intervention: A Single-Center, Retrospective Analysis. *Medical science monitor" international medical journal of experimental and clinical research*, *29*, e939203. 2023, https://doi.org/10.12659/MSM.939203
29. E. J.Favaloro, G.McCaughan, &, L. Pasalic "Clinical and laboratory diagnosis of heparin induced thrombocytopenia: an update" *Pathology*, vol.*49*,no.4, pp.346–355,2017, https://doi.org/10.1016/j.pathol.2017.02.005 .

30. Zhou, L., Bao, J., & Ma, J. (2019). Hemolytic Anemia and Reactive Thrombocytosis Associated With Cefoperazone/Sulbactam. *Frontiers in pharmacology*, vol. *10*, pp.1-4, 1342, https://doi.org/10.3389/fphar.2019.01342 .
31. K.Akahane, K.Furuhama, M.Kato, T.Une, & T. Onodera, "Influences of cephem antibiotics on the immune response in mice" *Chemotherapy*, vol.*36*, no.4 , pp.300–307,1990, https://doi.org/10.1159/000238781 .