

## RESEARCH REGARDING THE EFFECT OF SOME SYNTHETIC GLUCOCORTICOID HORMONES ON LEUKOCYTES, IN RATS

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### Abstract

*In this paper, the assessment is to investigate the effect of some synthetic glucocorticoid hormones (used frequently in therapy) on leukocytes (WBC and leukocyte formula), in rats. It is mentioned that the hormones used in this study (Prednisone, Dexamethasone and Betamethasone) have a large utilization in therapy due to the anti-inflammatory effect, overlooking in many cases the immunosuppressive effect. The determinations regarding the number of leukocytes and the leukocyte formula have been made using the IDEEX Analyzer, at the beginning and at the end of the experiment. As a result of the analysis it was revealed that the WBC%, at the end of the experiment, was higher, for all the experimental groups, comparing with the control group. After the interpretation of the obtained results regarding to the values of the leukocyte formula in the case of the experimental groups, it was shown increases of the percentages of neutrophils and basophils, decreases of percentages of eosinophils, lymphocytes and monocytes, comparing to the values obtained for the control group.*

**Key words:** leukocytes, glucocorticoid hormones, rat, leukocyte formula.

### INTRODUCTION

This research paper aims to elucidate the effects produced on leukocytes of some solutions that have synthetic glucocorticoid hormones as active substance. Currently, these preparations are used extensively in veterinary medicine, often without taking into account the adverse effects. In our study, we had in view the effect of these hormones on both the total leucocyte count and the different leukocyte categories (effect observed by changes in leukocyte formula).

By studying the literature in the field (Cotor G. et al., 2014; Codreanu I., 2018), we noticed that treatments with such products may increase the present chronic diseases, thus limiting the chances of success in some cases.

It is also known that the synthesis of glucocorticoid hormones intensifies in all stress states (Cotor G. et al., 2012), which is an aggravating factor in the subsequent evolution of the pathologic process. Thus, the emphasis on symptomatic medication has the immunosuppressive effect (Cotor G. et al,

2006), as main disadvantage and also the possible systemic effects of synthetic glucocorticoid hormones.

The present paper aims to demonstrate the effects of these hormones in blood in rats, one of the most used species regarding the researches (Codreanu I., 2011).

We consider our study to be important because the hormones we take into account have a wide use in the clinic (often their use is exaggerated) and the doctors do not give importance to the effect on the leukocytes. These effects may have serious repercussions on animal health, mainly due to the immunosuppressive effect (Cotor G. et al., 2014).

### MATERIALS AND METHODS

The materials used for this study were rats, blood analyzer, drugs, syringes, needles, blood harvesting tubes, etc.

The biological material consisted of laboratory rats from the Wistar line, which were divided into 4 lots (10 rats per lot), a control group and three experimental lots.

In our study, we used several synthetic glucocorticoid hormones contained in the following drugs: Prednisol (1 ml contains: Prednisolon acetat 25 mg, Benzyl alcohol 9 mg., Sodium chloride 8 mg., Carboxymethylcellulose sodium 3 mg., Polysorbate 80 mg.), Colvasone (1 ml contains: Dexamethasone disodium phosphate 2 mg and Benzyl Alcohol - preservative 20 mg) and Diprophos (1 ml contains: Betamethasone dipropionate, the equivalent of 5 mg of betamethasone and Betamethasone sodium phosphate, the equivalent of 2 mg of betamethasone).

The rats have been administered for 7 days in this way: physiological serum (control group), Prednisol (1st experimental group), Colvasone (2nd experimental group), Diprophos (3rd experimental group).

After two weeks (the period time that rats required to adapt to new living conditions), the rats were weighed (average weight was about 250 grams) in order to calculate the amount of substance (glucocorticoid hormone) that would be administered.

The rats have been administered for 7 days in this way: 0.3 ml physiological serum (control group), 1mg/kg Prednisolon - Prednisol (1st experimental group), 1 mg/kg Dexamethasone - Colvasone (2nd experimental group), 1 mg/kg Betamethasone - Diprophos (3rd experimental group). One day after the last administration, there were collected blood samples and were determined the leukocyte count and leukocyte formula.

## RESULTS AND DISCUSSIONS

The results obtained (number of leukocytes and leukocyte formula) will be presented in the following tables (average values for each lot) and figures accompanied by explanatory comments.

The comparisons regarding the statistical relevance of the differences between the experimental groups were made using the t test (Student).

The results obtained by the point of view of white blood cell count for the control group and the experimental groups at the end of the experiment are presented in Table 1 and Figure 1.

Table 1. The mean WBC values for the control and the 3 experimental lots

Lot category	WBC (thousands/mm <sup>3</sup> blood)
Control group	10.18
1 <sup>st</sup> group	10.62
2 <sup>nd</sup> group	12.18*
3 <sup>rd</sup> group	12.94*

\*P<0.05

Based on the statistical analysis it was found that between the WBC values recorded at the end of the experiment for the control group and for the 3 experimental groups there are the following significant differences (P<0.05):

- the discussed parameter is higher in the case of the Prednisolone group with 4.32% compared to the control group;
- the discussed parameter is higher for the Dexamethasone treated group with 19.64% compared to the control group;
- the discussed parameter is higher for the Betamethasone treated group with 27.11% compared to the control group.

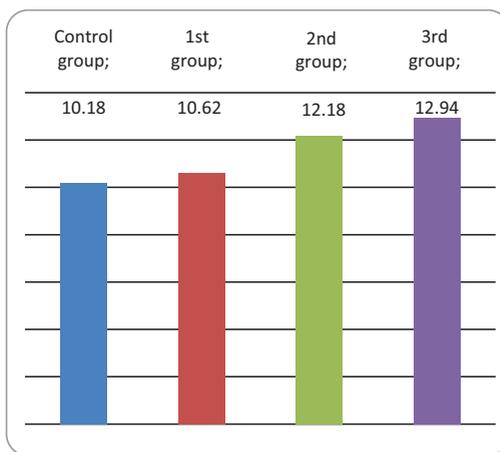


Figure 1. Comparative chart of the WBC for the control group and the 3 experimental groups

The data obtained by us falls within the values mentioned by the literature in the field (Cotor G. et al, 2014). Concerning the effect of synthetic glucocorticoid hormones on WBC, our results show an increase in this parameter, which is also highlighted by other researchers (Ghiță M. et al., 2015). This increase is attributed to neutrophils and basophils as follows. These results were reported by other

authors (Găjăilă G. et al., 2016; Medina-Martel M. et al., 2013).

The results obtained regarding the values of the leucocyte formula recorded in the rats in the control group and the rats in the 3 experimental groups at the end of the experiment are presented in Table 2 and Figure 2.

Table 2. Avarage weight of different leucocyte categories in the leucocyte formula in the control lot and experimental lot

Lot category	N (%)	E (%)	B (%)	L (%)	M (%)
Control group	33.4	2.8	0.4	59.8	3.6
1 <sup>st</sup> group	36.8*	2*	0.4	57.2*	3.6
2 <sup>nd</sup> group	38*	1.6*	0.6*	56.6*	3.2*
3 <sup>rd</sup> group	40.6*	1.6 *	0.6*	54*	3.2*

(N-neutrophils, E- eosinophils, B-basophils, L-lymphocytes, M-monocytes)

From the statistical analysis of the results obtained regarding the determination of the leucocyte formula for the groups treated with glucocorticoid hormones, compared to the control group, significant differences were observed which will be presented below for each category of leukocytes.

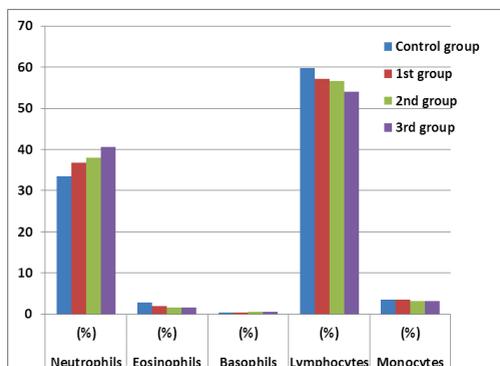


Figure 2. Comparative graph of each leukocyte group within the leucocyte formula for the control group and the 3 experimental groups

Concerning the leukocyte formula were found the following:

- the percentage of neutrophils was higher for all the experimental groups, the increases being of 10.17% (1st experimental group), 13.77% (2nd experimental group) and of 21.55% (3rd

experimental group), comparing to the control group.

- the percentage of the eosinophils was lower for all the experimental groups, the values being of 28.57% (1st experimental group) and of 42.85% (2nd experimental group \*P<0.05 and the 3rd experimental group), comparing to the control group.

- the percentage of the basophils was increased by 50% comparing to the control group, for all the experimental groups.

- the percentage of the lymphocytes was decreased by 4.34% (1st experimental group) 3.67% (2nd experimental grope) and of 9.69% (3rd experimental group), comparing to the control group.

- the percentage of the monocytes was lowered by 11.11%, comparing to the control group, for all experimental groups.

Our obtained data confirms the results of other authors (Gotor G. et al., 2014; Ghiță M. et al., 2015; Găjăilă G. et al., 2014). A particular aspect of our study is that besides the neutrophils, in the leukocyte formula the basophils are also high. We also found a relationship of direct proportionality between the amount of changed leukocyte in the leukocyte formula and the hormone product retention in the animal body.

## CONCLUSIONS

Following the statistical analysis of the results obtained, the following conclusions can be drawn:

1. Prednisolone caused a significant increase of WBC and neutrophil percentage and significant decrease of lymphocyte and eosinophil percentages compared to the control group.
2. Dexamethasone caused a significant increase of WBC, neutrophil count and basophil count and a significant decrease of eosinophil, lymphocyte and monocyte counts compared to the control group.
3. Betamethasone caused significant increase of WBC, neutrophil and basophil percentages and significant decrease of eosinophil, lymphocyte and monocyte counts compared to the control group.

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