Available online at www.scholarsresearchlibrary.com

Scholars Research Library

Archives of Applied Science Research, 2017, 9 (3):11-15
(http://scholarsresearchlibrary.com/archive.html)

ISSN 0975-508X
CODEN (USA) AASRC9

Effects of Anti-Inflammatory and Immunosuppressive Doses of Dexamethasone on Blood Parameters in Ouled Djellal Sheep

Kellali Narimane[1]*, Kerrour Mustapha[2], Lakhdara Nedjoua[1]
[1]*Laboratory of Health Management and Animal Production, Constantine, Algeria*
[2]*Institute of Veterinary Science of Constantine, Constantine, Algeria*

ABSTRACT

This study aimed to determinate the effects of Dexamethasone (DEX) administered parentally (Intramuscularly: IM) at therapeutic doses conventionally used in the anti-inflammatory and immunosuppressive treatments on blood parameters values in sheep.

Fifteen sheep of Ouled Djellal breeds were randomly divided into three groups of five rams. The animals received an injection of Dexamethasone (DEXALONEND solution) at a rate of 1.52 mg *DEX/d during 6 days for group1 and 3.4 mg DEX/d during 6 days for group 2, the third group has served as a control group and did not receive any treatment. Blood samples taken over a period of 3 weeks were analyzed to define blood parameters (WBC: White Blood Cells, RBC: Red Blood Cells, HCT: Hematocrit, HGB: Hemoglobin, and PLT: Platelets). It has been shown that Dexamethasone induced in treated animals, an increase of WBC number and platelets with a diminution of RBC number and HGB rate, at the same time, variations in hematocrit were more fluctuating. From these results, we can deduce that Dexamethasone effects are transitory and therefore do not have a significant impact on the health of the animals.*

Keywords: Dexamethasone, Sheep, WBC, RBC, HGB, HCT, PLT

INTRODUCTION

The haematological characterization of animals is of particular interest especially for the diagnosis of many diseases. Moreover, studies on the management and valorization of animal resources clearly indicate the need for reliable haematological standards [1]. Iatrogenic factors may modify the values of haematological parameters such as the use of synthetic glucocorticoids. It's well known that glucocorticoids (GCs) are widely used in veterinary medicine for their anti-inflammatory and immunosuppressive effects [2]. In Algeria, sheep herds are subjects for excessive use of synthetic glucocorticoids by veterinary practitioners. The disturbance of haematological parameters can have an impact on animal's health. It is within this framework that our trial aims to elucidate the possible effects of different doses of DEX on hematological parameters. The importance of this work is to know precisely the possible changes provoked by the GCs and thus to be able to assist veterinary practitioners in their work.

MATERIALS AND METHODS

Animals

Fifteen Ouled Djellal sheep males, aged from 12 to 14 months, weighing 45-53 kg was included in this study. All sheep's were clinically healthy and did not receive any medication prior to the experiment. The animals were randomly divided into three groups of five sheep each.

Experimental protocol

Two groups of sheep were injected intramuscularly with Dexamethasone (DEXALONEND solution, 2 mg disodium phosphate of Dexamethasone, COOPHAVET, France) at a dose of 1.52 mg/day (anti-inflammatory dose) for group I and 3.4 mg/day (immunosuppressive dose) for group II. Dexalone concentration is about 152 mg of DEX/100 ml of

Dexalone (1.52 mg of DEX/1 ml of Dexalone). The dosage is from 1 to 4 ml/100 kg of body weight (0.5 to 2 ml/50 kg of body Wright). 1 ml of Dexalone (1.52 mg of DEX) was used for the anti-inflammatory dose and 2 ml Dexalone (3.4 mg of DEX) for the immunosuppressive dose. The third group, which is the control group, received no treatment. Treatment with Dexamethasone lasted 6 days (usual duration of anti-inflammatory treatment).

Samples collection

The blood samples were obtained from the jugular vein and collected in vacuum heparinized tubes. The first sample was taken 48 h before Dexamethasone administration.

The second sample (1st one upon the treatment) was performed 6 h after the first injection of Dexamethasone. Between the 3rd and the 8th, the samples were collected at 24, 48, 72, 96 and 120 h after the beginning of the treatment. From the 9th to the 11th intake, the samples were taken at 1, 6 days and 14 days after the end of the treatment. The same sampling protocol was adopted for the three groups.

Treatment of samples

Hematological parameters (GB, GR, HGB, HCT and PLT) were determined using a HEMASCREEN 18 (Hospacex Diagnostics, Italy).

Statistical analysis

Data were analyzed using Turkey's test and ANOVA test (Minitab® 15). Differences were considered as significant when $P<0.05$.

RESULTS

Leukocytes

The number of leukocytes increased 24 h after treatment initiation in all sheeps who received DEX compared to the control group with a significant difference ($p<0.05$). The leukocytes number obtained in sheeps treated with DEX returned to its initial values 72 h after the beginning of the treatment, but remained superior to that of the control group, no significant difference was observed as shown in Table 1.

Table 1: Temporal evolution of white blood cells number ($\times 10^3/\mu L$)

	48 h before	6 h	24 h	48 h	72h	96 h	120 h	24 h after	6 days after	14 days after
Control	10.67 ± 2.37	10.75 ± 1.13	10.47 ± 1.38	10.27 ± 2.07	9.64 ± 2.03	10.04 ± 1.80	9.72 ± 1.78	10.59 ± 1.37	11.64 ± 2.74	10.28 ± 2.47
Group I	11.25 ± 1.49	11.88 ± 0.89	12.35 ± 0.85	11.87 ± 1.83	11.18 ± 1.55	11.47 ± 1.35	10.97 ± 1.83	11.62 ± 1.70	12.46 ± 1.63	11.03 ± 1.46
Group II	11.93 ± 1.31	12.14 ± 0.90	12.66 ± 0.75	12.64 ± 1.65	12.03 ± 1.25	11.98 ± 1.13	12.00 ± 1.43	11.77 ± 1.82	13.18 ± 0.96	11.15 ± 0.85

Erythrocytes

Fluctuations in the RBCs number were similar for the three groups. As shown in Table 2 a significant decrease ($P<0.05$) was observed in 24 h after the beginning of the treatment followed by a significant increase ($P<0.05$) up to 48 h after the start of treatment, bringing buck the number of RBCs to its baseline values.

Table 2: Temporal evolution of red blood cells number ($\times 10^6/\mu L$)

	48 h before	6 h	24 h	48 h	72 h	96 h	120 h	24 h after	6 days after	14 days after
Control	9.18 ± 0.57	8.58 ± 0.63	8.43 ± 0.36	8.33 ± 0.39	8.98 ± 0.42	9.66 ± 0.60	9.15 ± 0.46	9.12 ± 0.49	9.00 ± 0.62	8.68 ± 0.56
Group I	8.43 ± 0.70	8.60 ± 0.45	7.87 ± 0.50	8.10 ± 0.81	8.49 ± 0.66	8.79 ± 0.88	8.92 ± 0.84	8.74 ± 0.95	8.93 ± 0.79	8.97 ± 0.83
Group II	9.35 ± 0.27	8.81 ± 0.11	7.98 ± 1.22	8.86 ± 0.41	9.46 ± 0.42	9.44 ± 0.62	9.80 ± 0.53	9.30 ± 0.32	9.33 ± 0.67	9.19 ± 0.82

Hemoglobin

Hemoglobin level obtained for all three groups decreased gradually throughout the experiment (P<0.03) and passed from 10.30 ± 0.48 to 9.26 ± 0.62, from 9.52 ± 0.43 to 8.34 ± 0.42 and from 10.10 ± 0.33 to 8.90 ± 0.77 for the control, group I and group II respectively as shown in Table 3. It should be noted that no significant difference was detected between the values obtained for the three groups and that they were within the range of the usual values reported by Ndoutamia [1], Etim [3] and Oramari [4].

Table 3: Temporal evolution of hemoglobin HGB (g/dL)

	48 h before	6 h	24 h	48 h	72 h	96 h	120 h	24 h après	6 days after	14 days after
Control	10.30 ± 0.48	9.90 ± 0.72	10.06 ± 0.49	9.14 ± 0.57	9.36 ± 0.59	9.48 ± 0.58	9.70 ± 0.61	9.38 ± 0.50	9.08 ± 0.33	9.26 ± 0.62
Group I	9.52 ± 0.43	9.56 ± 0.36	9.26 ± 0.19	8.74 ± 0.46	8.64 ± 0.53	8.36 ± 0.52	9.08 ± 0.55	8.46 ± 0.40	8.72 ± 0.41	8.34 ± 0.42
Group II	10.10 ± 0.33	10.12 ± 0.33	10.22 ± 0.58	9.36 ± 0.58	9.64 ± 0.61	8.94 ± 0.61	9.88 ± 0.61	9.22 ± 0.60	9.18 ± 0.90	8.90 ± 0.77

Platelets

Except a transient and significant decrease (P<0.03) of thrombocytes number observed 24 h after the end of treatment in the control group, no other significant changes were observed over time, neither in control group nor in group II.

On the other hand, the scattering of platelets number (DEX at 1.52 mg/day) was very important in group I, with two peaks: a peak at 24 h after the start of treatment where the number of thrombocytes was significantly greater than that obtained for the two other groups (P<0.03) and another peak at the 120th hour after the injection of DEX. An episodic and significant decrease (P<0.001) was observed following the second peak as shown in Table 4.

Table 4: Temporal evolution of platelets number PLT (× 10^3/µL)

	48 h before	6 h	24 h	48 h	72 h	96 h	120 h	24 h après	6 days after	14 days after
Control	319 ± 34.98	336 ± 56.24	361.4 ± 92.11	321.5 ± 35.52	292.2 ± 48.76	381.6 ± 23.81	379.2 ± 73.66	212 ± 36.41	426.2 ± 90.82	374 ± 70.51
Group I	336.6 ± 38.25	372.6 ± 36.03	537.2 ± 81.40	320 ± 38.37	309.4 ± 32.68	421 ± 65.44	452.4 ± 30.94	266 ± 32.52	441.8 ± 47.18	441 ± 49.81
Group II	295.8 ± 90.03	333 ± 74.39	387 ± 92.28	271.4 ± 72.83	274.4 ± 70.62	320.2 ± 20.23	369.4 ± 87.51	286.6 ± 118.84	362.8 ± 84.88	398.8 ± 45.33

Hematocrit

For this parameter, the values observed in the control sheep showed no significant variation over the duration of the experiment. For the first group, hematocrit rate increased slightly 6 h after DEX injection before decreasing significantly (P<0.05) 48 h after the initiation of the treatment. The values obtained at 48 h approach the baseline values, then the hematocrit levels have increased, without exceeding baseline values as shown in Table 5.

In group II, changes in hematocrit followed the same rate as group I with a non-significant increase followed by a significant decrease (P<0.05) 24 h after starting the treatment, followed by a gradual increase until 120 h (P<0.05).

However, it should be noted that despite these fluctuations, the values obtained were close to the baseline values.

Table 5: Temporal evolution of hematocrit HCT (%)

	48 h before	6 h	24 h	48 h	72 h	96 h	120 h	24 h after	6 days after	14 days after
Control	30.16 ± 2.05	29.7 ± 2.33	27.02 ± 4.19	26.96 ± 1.96	28.96 ± 1.87	30.86 ± 2.34	29.30 ± 1.69	29.92 ± 1.50	28.98 ± 1.72	27.92 ± 2.24
Group I	27.64 ± 1.93	30.16 ± 1.48	26.92 ± 2.01	26.1 ± 1.76	27.18 ± 1.33	27.94 ± 1.81	28.22 ± 1.86	27.55 ± 1.14	28.80 ± 1.41	27.06 ± 0.82

Group II	30.18 ± 1.10	30.78 ± 0.80	27.48 ± 2.80	28.22 ± 1.36	30.02 ± 1.67	29.72 ± 1.95	30.84 ± 1.61	29.84 ± 1.07	29.70 ± 2.60	27.36 ± 1.69

DISCUSSION

In our experiments, we evaluated the effect of the administration of DEX in IM at an anti-inflammatory dose (1.52 mg/day for 6 days) and immunosuppressive (3.4 mg/day for 6 days) on hematological parameters.

The main finding in our trial is that administration of DEX in IM induced an increase in the number of leukocytes and platelets with a decrease in red blood cells and hemoglobin in the treated animals. Hematocrit values were more fluctuating. The mean values obtained for these parameters were included in the reference limits throughout the duration of the study.

The significant increase in WBC number caused by DEX was noted 24 h after the beginning of the treatment. The number of leukocytes returned to baseline after 72 h post-treatment. This increase was confirmed by Thanasak [5] who reported that 48 h after a single injection of DEX (0.02 mg/kg IM) to dairy cows at about two weeks postpartum, the number of Leucocytes increased. Similar results were observed in dogs receiving oral methylprednisolone (2 mg/kg/day for 10 days, then 2 mg/kg on alternating days for 50 days) where a significant increase of WBC number was observed between 11th and 41st days after the start of treatment. WBC number has returned to its initial values at 51st day [2].

However, our results contradicted those obtained by Yasuhiko [6] who found that single administration of DEX in rats significantly reduced the number of leukocytes with a nadir 8 h post-injection.

This contradiction could be explained by the different Dexamethasone injection protocol and the important dose used by Ysuhiko [6]. WBC regained baseline levels after 24 h. It has been shown that oral administration of Dexamethasone at 60 µg/Kg/day for 30 days induced a decrease in leucocytes number in rats.

The leukocytosis observed in our experimentation could be explained by the increase in the number of polymorphonuclear neutrophils (PNN). The increase in the number of PNN is the result of several phenomena: increased release of immature neutrophils from the bone marrow into the circulation decreased passage of PNN into tissues and increased degeneration of neutrophils in the vascular endothelium [2,5].

DEX provoked a significant increase of platelets number compared to baseline values 24 h after initiation of treatment in group I (animals receiving 1.52 mg/day for 6 days). The number of thrombocytes returned to baseline 48 h after the start of treatment.

These results are in agreement with those obtained by Amar [7] who have noted, after 30 days of treatment, a significant increase in the number of thrombocytes in Wistars rats receiving DEX at 180 µg/Kg/J. The administration of DEX to premature infants induced a significant increase of thrombocytes number [8]. Furthermore it has been reported that both prednisolone and Dexamethasone are capable of increasing the platelet count in patients with autoimmune thrombocytopenia [9]. DEX, at a rate of 10 mg every 6 h (oral or intravenous) for 4 days, induced a significant increase of platelets number at the end of the 5th day of treatment. The reason of this increase remains unexplained in the literature. Notwithstanding the reduction in RBC number observed 24 h after the beginning of the treatment, the values obtained remained within the reference limits and no significant differences were detected between the values obtained for the control group and the treated groups. Therefore DEX had no effect on RBC number. Our results agree with those of Marin [10] who reported that the daily administration of low doses of DEX (0.75 mg/day) to bull calves did not influence the number of erythrocytes. However, Dexamethasone administered subcutaneously once a day for 7 days at decreasing doses (from 2 mg/kg to 0.1 mg) in beagle dogs induced a decrease of RBC number [11].

In our experiment, the hemoglobin level was gradually reduced throughout the experiment and no significant difference was detected between the values obtained for the three groups. This suggests that DEX had no effect on the HGB rate.

These results are consistent with those obtained by Marin [10] who reported that DEX administered to bull calves at (0.75 mg/day) had no effect on HGB. In contrast, Deniz [11] reported a decrease in HGB following administration of Dexamethasone in beagle dogs subcutaneously once a day for 7 days at decreasing doses (ranging from 2 mg/Kg to 0.1 mg).

Finally, for the hematocrit (which is the percentage of the volume of RBC relative to the total volume of the blood), the decrease observed 24 h after the start of treatment could be explained by the decrease in the number of RBC. However, this decrease was noted in the three groups and the number of GRs remained within the usual values throughout the experiment. This suggests that DEX had no effect on HCT and that fluctuations in HCT may be justified by possible changes in hydration status of the animals.

CONCLUSION

The changes in haematological parameters observed following short-term intramuscular administration of DEX at therapeutic doses were reversible; WBC, RBC, HCT, HGB and PLT returned to their initial values 24 h after the treatment withdrawal. Regarding our results, the effects of DEX were only transient and therefore they may not have a significant effect on animal's health within a short period of treatment.

REFERENCES

[1] Ndoutamia, G. and Ganda, K., *Revue Méd Vét*, **2005**. 156(4): p. 202-206.

[2] Altuğ, N. and Ağaoğlu, Z.T., *Bull Vet Inst Pulawy*, **2007**. 51: p. 109-115.

[3] Etim, N.N., et al., *Agric Sci*, **2014**. 2(1): p. 37-47.

[4] Oramari, R.A.S., Bamerny, A.O. and Zebarih, M.H., *Adv Life Sci Technol*, **2014**. 2: p. 56-62.

[5] Thanasak J., et al., *Vet Res*, **2004**. 35: p.103-112.

[6] Yasuhiko O., et al., *J Health Sci*, **2010**. 56(2): p. 215-220.

[7] Amar, M.I., et al., *J Med Sci*, **2013**.13(6): p. 432-438.

[8] Bourchier, D. and Weston, P.J., *J Paediatr Child Health*, **1991**. 27(2): p. 101-104.

[9] Praituan, W. and Rojnuckarin, P., *J Thromb Haemost*, **2009**. 7: p. 1036-1038.

[10] Marin, A., et al., *Animal*, **2008**. 2(7): p. 1080-1086.

[11] Deniz, A., Spiecker-Hauser, U. and Rehagen, M., *Intern J Appl ResVet Med*, **2009**. 7(3): p. 116-129.