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EFFECT OF LONG ACTING OXYTETRACYCLINE ON TOTAL LEUCOCYTE COUNT HEMATOLOGICAL PARAMETERS IN WISTAR ALBINO RATS

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ABSTRACT

The immune system expresses an adaptive response in all the vertebrates against invading microorganisms. The role of immune system is to sustain host defense mechanisms and maintain homeostasis. The Long acting oxytetracycline administered at 20 and 40 mg/kg body weight mg/kg body weight through intramuscular route in wistar albino rats. Sheep RBC used as antigen. The present study was conducted to study the hematological parameters like Total Leucocyte count (TLC) in rats. There was no significant (P>0.05) difference in TLC values in antigen plus long acting oxytetracycline low dose (Group VII) and high dose (Group VIII). There was decrease in the TLC values in treated groups (Group VII and VIII) when compared to antigen (Group V) and antigen plus pyrrolidone (Group VI) group. This is suggestive that long acting oxytetracycline at low and high dose does not affect immnune response in rats.

KEYWORDS: Long acting oxytetracycline, Total Leucocyte Count. Immune response.

INTRODUCTION

Immunomodulation may involve either an increase in the magnitude of immune response i.e. immunostimulation or a decrease in the magnitude of the immune response i.e. immunosuppression. Immunomodulation can further be divided as specific and nonspecific. Specific immunomodulation implies a change in the response of the system to a particular antigenic stimulus as brought about by process of vaccination (specific immunostimulation) and desensitization (specific immunosuppression). Nonspecific immunomodulation implies a more fundamental change, where by the "State of Alertness" of the immune system is altered, this infusion affects the nature of its responses to the multiplicity of antigenic stimuli (Goodman and Gillman, 2001).

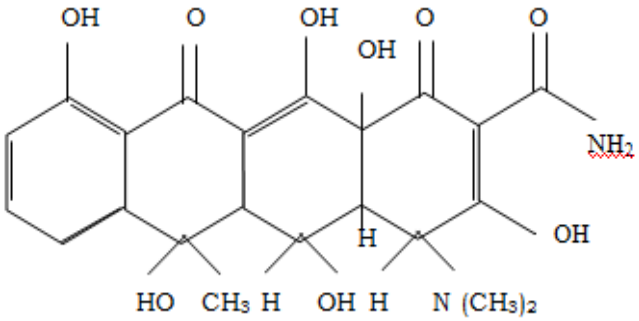
Long acting oxytetracycline belongs to tetracycline group of antibiotics. It was isolated from *Actinoomycete streptomyces rimosus.* Oxytetracycline is a broad spectrum antibiotic with bacteriostatic activity widely used in veterinary medicine for the treatment of respiratory and gastrointestinal infectious diseases. It is active against aerobic gram positive and gram negative bacteria, rickettsia, mycoplasma, chlamydial infections anaplasmosis, babesiosis, theilariosis, pasteurellosis, bovine kerato conjunctivitis, ovine foot rot etc (Swift and Thomas, 1983; Musser *et al*. 1996).

To prevent repeated administration, to reduce the cost of treatment and to avoid stress condition, a long acting formulation of oxytetracycline was developed. The prolonged effect of this new preparation was claimed to be due to use of 2-pyrrolidone based formulation which should lead to provide prolonged circulating antibacterial concentration of the active agent for three to five days and controlled precipitation of oxytetracycline at the site of injection without significant tissue damage.

Wister Albino rats aged between two to three month old within body weight ranging from 150 to 200 g were procured from Small Animal House, Veterinary college, UAS, Bangalore. The animals were divided into eight experimental groups consisting of ten animals each group with equal number of male and female rats. Animals were housed in standard polypropylene rat cages and allowed for acclimatization for one week before the start of actual study and maintained hygienically under standard laboratory conditions by providing commercial pellet feed and water *ad libitum* (Alastrain and Warden, 1989).

Long acting oxytetracycline available as Oxytetracycline dihydrate injectable solution / L.A. (Oxytetracycline dihydrate 200 mg/ml in 2-pyrrolidone) manufactured by Pfizer Limited, Mumbai was used in the experiment. This preparation was further diluted with 2-pyrrolidone and a single administration to experimental animal by intramuscular route was carried out.

Structure of Oxytetracycline dihydrate



Experimental protocol

The animals were divided into eight experimental groups. The details of the treatments given were as follows.

Group I - Saline control (no treatment).

Group II - Vehicle control i.e. 2-pyrrolidone (0.5 ml) administered through intramuscular route.

Group III - Single dose administration of long acting oxytetracycline at 20 mg/kg body weight through intramuscular route.

Group IV - Single dose administration of long acting oxytetracycline at 40 mg/kg body weight through intramuscular route.

Group V - Administered 0.4 ml antigen on Day 0 and Day 7 intraperitonaelly.

Group VI - Administered 0.4 ml antigen on Day 0 and Day 7 intraperitoneally and 0.5 ml 2-pyrrolidone through intramuscular route.

Group VII - Administered 0.4 ml antigen on Day 0 and Day 7 intraperitoneally and long acting oxytetracycline at 20 mg/kg body weight through intramuscular route.

Group VIII - Administered 0.4ml antigen on Day 0 and Day 7 intraperitoneally and long acting oxytetracycline at 40 mg/kg body weight through intramuscular route.

Group I, II, III and IV were normal non antigen stimulated groups. In these Group I was Saline control, Group II was given vehicle i.e. 2-pyrrolidone control, Group III, and Group IV were given long acting oxytetracycline at 20 and 40 mg/kg body weight through intramuscular route, respectively.

The vehicle or long acting oxytetracycline given on Day '0'. These groups were used to assess the effect of long acting oxytetracycline on non-specific natural host defense mechanisms in rats.

Group V, VI, VII and VIII were antigen stimulated groups. In these, Group V was given antigen, Group VI was given antigen and pyrrolidone, Group VII was given antigen and long acting oxytetracycline at 20 mg/kg body weight through intramuscular route, Group VIII was given antigen and long acting oxytetracycline at 40 mg/kg body weight through intramuscular route. Antigen was given on Day '0', Vehicle and drug at two different doses were administered Day 1 after the administration of antigen. A second dose of antigen was given on Day 7 as a booster dose. These groups were used to assess the effect of long acting oxytetracycline on specific immune response.

Collection of blood samples

The rats were anaesthetized with diethyl ether and blood was collected from retro-orbital plexus.In all the groups blood was collected on Day '0' i.e. immediately before administering the drug/antigen and then on Day 1, 7, 14, 21, 28, 35 and 42 of the experiment.

Hematological Parameters

The blood was collected from retro-orbital plexus puncture method using microhaematocrit capillary tubes. Disodium Ethylene Diamine Tetraacetic Acid (EDTA) was used as anticoagulant. The following hematological parameters were determined following standard methods (Jain, 1990). In the present study estimation of the Total Leukocyte count (TLC) in non antigen and antigen rats.

Statistical analysis

The data generated from the experimental study was subjected to one-way ANOVA by statistical analysis (Snedecor and Cochran, 1976) using computerized Graph Pad Prism software.

RESULTS AND DISCUSSION

Table 1. The effect of long acting oxytetracycline on total leucocyte count (per cumm) in non antigen stimulated rats.

Time interval in days	Saline control (Group I)	Pyrrolidone control (Group II)	Low dose (20 mg/kg) (Group III)	High dose (40 mg/kg) (Group IV)
0	5400.00 ± 124.62	5631.02 ± 85.31	6231.00 ± 78.31	6184.00 ± 89.00
1	6421.04 ± 116.34	6689.32 ± 47.39	6789.67 ± 59.56	6438.04 ± 114.14
7	7392.04 ± 93.69	7603.00 ± 56.27	6478.00 ± 124.78	6032.45 ± 86.19
14	6939.12 ± 87.34	7036.31 ± 97.39	6834.14 ± 94.39	6934.47 ± 57.14
21	7043.14 ± 80.31	7183.28 ± 87.35	7030.15 ± 101.24	6988.08 ± 85.09
28	7130.28 ± 104.39	7208.98 ± 127.10	7124.15 ± 64.51	7200.24 ± 123.04
35	7210.39 ± 73.74	7314.20 ± 45.83	7222.24 ± 59.16	7290.24 ± 34.18
42	7258.48 ± 122.30	7400.00 ± 80.20	7330.00 ± 70.14	7380.09 ± 93.12

Values: Mean ± SE, n =10, P>0.05.

Table 2. The effect of long acting oxytetracycline on total leucocyte count (per cu.mm.) in antigen stimulated rats.

Time interval in days	Antigen control (Group V)	Antigen+ Pyrrolidone control (Group VI)	Antigen + Low dose (20 mg/kg) (Group VII)	Antigen + High dose (40 mg/kg) (Group VIII)
0	5549.00 ± 92.30	5963.02 ± 67.53	6096.18 ± 96.10	6011.42 ± 124.30
1	5770.67 ± 117.62	5849.00 ± 68.83	5710.00 ± 57.24	5934.00 ± 110.32
7	5652.00 ± 148.54	6280.34 ± 129.42	5524.21 ± 79.34	5498.02 ± 61.42
14	6337.33 ± 128.23	6210.20 ± 116.42	6012.24 ± 62.34	5958.00 ± 57.83
21	6242.24 ± 109.00	6894.27 ± 82.34	6380.30 ± 102.34	6329.21 ± 84.35
28	5893.00 ± 128.23	6681.12 ± 102.87	6150.32 ± 48.53	6080.18 ± 139.34
35	6299.24 ± 89.34	6505.60 ± 156.43	6320.00 ± 67.35	6200.16 ± 110.39
42	6853.41 ± 53.88	6538.00 ± 132.34	6610.18 ± 71.47	6570.10 ± 94.35

Values: Mean± SE, n =10, P>0.05.

Total leukocyte count (TLC)

The TLC (per cubic mm) values of all the experimental groups are depicted in Table 1 and 2.

The TLC in saline control group (Group I) ranged from 5400.00±124.62 to 7258.48±122.30 per cumm. The TLC in pyrrolidone group (Group II) ranged from 5631.02±85.31 to 7603.00±56.27 per cumm. The group given long acting oxytetracycline low dose (Group III) showed TLC values ranging from 6231.00±78.31 to 7330.00±70.14 per cumm. The group given high dose (Group IV) showed TLC values ranging from 6032.45± 86.19 to 7380.09±93.12 per cumm.

There was no significant (P<0.05) difference in TLC values in treatment groups (Group III and IV) when compared to saline control and pyrrolidone control groups (Group I and II).

Among antigen stimulated groups, the TLC values of antigen control group (Group V) ranged from 5549.00±92.30 to 6853.41±53.88 per cumm. In the group given antigen and pyrrolidone (Group VI) TLC values ranged between 5849.00±68.83 and 6894.27±82.34 per cumm. In the group given antigen and low dose group (Group VII) TLC values ranged between 5524.21±79.34 and 6610.18±71.47 per cumm. In the group received antigen and long acting oxytetracycline high dose group (Group VII), the TLC values ranged between 5498.02±61.42 to 6570.10±94.35 per cumm.

There was no significant (P>0.05) difference in TLC values in antigen plus long acting oxytetracycline low dose (Group VII) and high dose (Group VIII). There was decrease in the TLC values in treated groups (Group VII and VIII) when compared to antigen (Group V) and antigen plus pyrrolidone (Group VI) group.

There are no reports available on the effect of long acting oxytetracycline on total leucocyte count in rats.

On the contrary, Hussain *et al*. (2002) reported that oxytetracycline at the dose of 22.5 and 30.0 mg/kg body weight for a period of 42 days decreased the TLC in mice.

Trinica *et al*. (2015) reported that administration of oxytetracycline oral overdoses of 8 and 16 times the recommended dosage of the product over a period of 3 consecutive days in brioler chicken. There will be decreased in total number of leukocytes and lymphocyte level in the group treated with the highest dose can be attributed to a possible drug imunosupression.

CONCLUSION

The present study was conducted evaluate the effect of long acting oxyteracycline on humoral and specific immune response in rats. Sheep RBC used as antigen in this study. The Long acting oxytetracycline administered at 20 and 40 mg/kg body weight mg/kg body weight through intramuscular route in wistar albino rats. Sheep RBC used as antigen. The present study was conducted to study the hematological parameters like Total Leucocyte count (TLC) in rats. There was no significant (P>0.05) difference in TLC values in antigen plus long acting oxytetracycline low dose (Group VII) and high dose (Group VIII). There was non significant decrease in the TLC values in treated groups (Group VII and VIII) when compared to antigen (Group V) and antigen plus pyrrolidone (Group VI) group. This is suggestive that long acting acting

oxytetracycline at low and high dose does not affect the humoral and specific immnune response in rats.

REFERENCES

1. Alastrain, N. and Warden, 1989, Handbook of Laboratory Animals. Anmol Pub., New Delhi.
2. Goodman and Gillman, 2001, Tha Pharmacological Basis of Therapeutics, 10th Edn, McGraw- Hill Publication. Hudson, L. and Hay, F.C. Haemagglutination. In: Practical Immunology. 3rd Edn., Black Well Scientific Pub., Oxford/ London, 1989; 252-254.
3. Jain, N.C., 1990, Schalm's VeterinaIy Haematology. 4th Edn. K.M. Verghese Co., New Delhi.
4. Hussain, M.A., Awal, M.A., Mostofa, M., Ismalm, M.S. and Rahman, M.M., Effects of long acting administration of oxytetracycline on mice. Bangladesh Veterinarian, 2002; 19: 125-128.
5. Musser, J. , Mechor, G.D. , Gröhn, Y.T , Dubovi, E.J., Shin. S. Comparision of tilmicosin with long acting oxytetracycline for treatment of respiratory tract disease in calves. J. American. Vet, Med. Association. 1996; 208(1): 102- 106.
6. Snedecor, G.W. and Cochran, W.G., (1976), Statistical Methods. VI Edn., Oxford and IBH Pub. Co., Calcutta.
7. Swift, B, L. and Thomas, G.M. Bovine anaplasmosis: elimination of the carrier state with injectable long acting oxytetracycline J. American. Vet, Med. Associatio, 1983; 183(1): 63-65.
8. Trinca, S. F., Ognean, L., Chiurciu,V., Arion, A., Dumitru, I. The Influence of Different Doses of Oxytetracycline on Some Hematological Parameters in Broilers, Bulletin of University of Agricultural Sciences and Veterinary Medicine, 2015; 72(1): 19- 22.