

Fourteen Days Oral Administration of Therapeutic Dosage of some Antibiotics reduced Serum Testosterone in Male Rats.

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ABSTRACT

Fourteen days oral administration of therapeutic dose of Ampicillin (4mg/100g/day), Cloxacillin (6mg/100g/day) and Tetracycline (12mg/100g/day) separately to healthy adult male albino rats significantly reduced their serum testosterone level as assessed by enzyme immunoassay. The control group received equal volume of the vehicle (Normal saline) throughout the period of the treatment. A significant reduction ($P < 0.05$) in testicular and epididymal weight was also produced by Cloxacillin, Cloxacillin and Tetracycline respectively. Ampicillin administration on the other hand significantly reduced ($P < 0.05$) prostate gland weight. After subjecting the treated animals to a recovery period ranging from 1-2 weeks, during which the drug administration was discontinued, all the animals recovered fully from the antifertility effect of these antibiotics on the serum testosterone level by the end of the second week. A significant recovery in the epididymal, testicular and prostate gland weight was also recorded in the Cloxacillin and Tetracycline, Cloxacillin, and Ampicillin treated animals respectively. The result suggests that the reversible antifertility effects of these antibiotics were produced via the disruption of testosterone hormone production process. This was also accompanied by reduction in the weight of some of the male reproductive organs.

KEY WORDS: Male Infertility, Antibiotics, Tetracycline, Ampicillin, Serum Testosterone, Male reproductive organs, and Antifertility.

INTRODUCTION

Antibiotics are some of the common drugs for therapeutic use. Ampicillin and Cloxacillin are broad-spectrum antibiotics that are used in the treatment of various bacterial infections. They belong to the Penicillin group of antibiotics. They also have prophylactic use in the pre-management of surgical operations (1). They are sometimes used along treatment of malaria, a dreaded disease of the tropics (2). It was reported to be among the commonly abused drugs in the society (3). Antibiotic therapy has been shown to adversely affect male fertility using such parameters as sperm count, motility, morphology, spermatogenic index and testicular

weight in both human and animal models. (4,5).

However, there is scanty information on the effects of antibiotics on circulating testosterone level in the male. The few reports indicated that some antibiotics have no effect (6,7). This study therefore, tends to update and validate previous reports on the effects of long term use of antibiotics vis-à-vis Ampicillin, Cloxacillin and tetracycline on circulating testosterone level in male using animal model.

MATERIALS AND METHOD

Healthy adult male Albino rats weighing between 200-240g were used for this experiment. They were kept in a metal cage in a healthy

environment in the animal house. The animals had free access to food and water throughout the period of the experiment. Smith Kline Beecham International manufactured the ampicillin tablets used, cloxacillin tablets by Maxheal Pharmaceutical India and tetracycline tablets by Gold Moore international. Fresh suspensions of the drugs were prepared in normal saline daily for use orally with the aid of an oral cannula. Weights of the animals were taken daily through out the period of the experiment.

At the end of the two weeks treatment with antibiotics, blood samples were collected using cardiac puncture technique with a 5ml needle into a sterile universal sample bottle and spun at 3000 r.p.m. for 5min to obtain the serum used for testosterone assay. The serum was stored at -20°C until the assay was carried out. Blood sample was taken from each of the rats in the treated groups,

their corresponding 1 and 2 weeks recovery and the control group. At the end of fourteen days drug administration, antibiotic therapy was discontinued for the animals. Blood samples were collected from control group as well as the treated group of ampicillin, cloxacillin and tetracycline. The period of recovery ranged between 7-14 days. Blood samples were collected from the recovery groups for testosterone assay at the end of the specified recovery period. The testosterone assay was carried out using enzyme immuno assay (EIA) system. A quality control sample was carried out at the beginning as well as at the end of the assay procedure to ensure acceptability with respect to bias and within assay variation. The testosterone assay kit was imported from Immunometrics U.K. Ltd.

The results were analyzed using ANOVA and presented as Mean \pm SEM. Level of statistical significance between groups and the control group was placed at $P < 0.05$.

Table 1: Animal grouping for control, treated, two weeks recovery groups and three weeks recovery groups of ampicillin, cloxacillin and tetracycline.

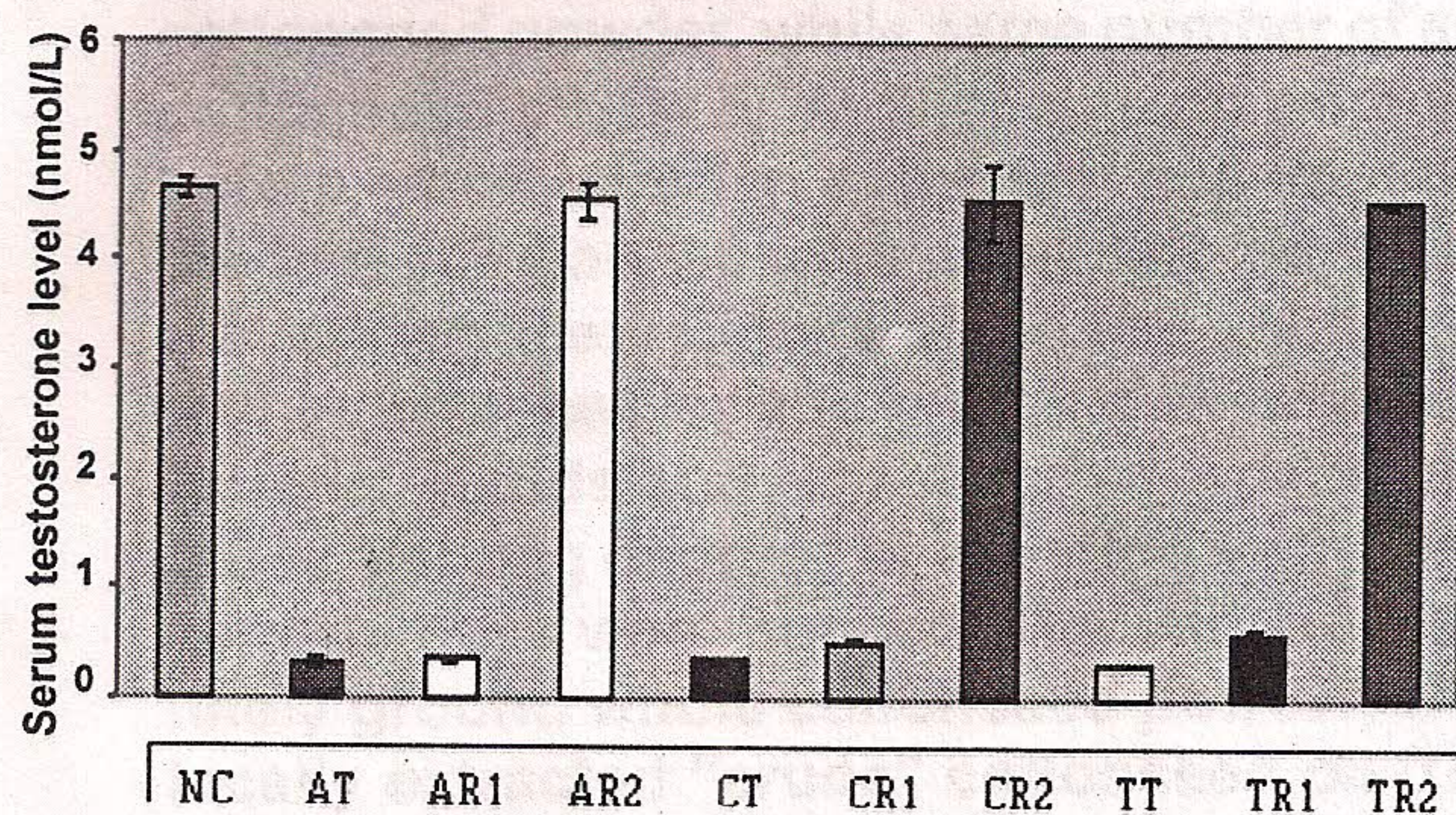
Group	Purpose	Number of animals	Dosage of Antibiotic for two weeks	Number of days for recovery
1.	Normal control "NC"	6	None	Nil
2.	Ampicillin treated "AT"	6		Nil
3.	1weeks Ampicillin recovery "AR1"	6	4mg/100g/day orally	7
4.	2 weeks Ampicillin recovery "AR2"	6	4mg/100g/day orally	14
5.	Cloxacillin treated CT"	6		Nil
6.	1 week Cloxacillin recovery "CR1"	6	6mg/100g/day orally	7
7.	2 weeks Cloxacillin recovery "CR2"	6	6mg/100g/day orally	14
8.	Tetracycline treated "TT"	6	12mg/100g/day orally	Nil
9.	1-week Tetracycline recovery "TR1."	6	12mg/100g/day orally	7
10.	2 weeks Tetracycline recovery "TR2"	6	12mg/100g/day orally	14

Table 2: Results of serum level of testosterone level in (nmol/L) in rats treated orally with ampicillin (6mg/100g body weight, cloxacillin (4mg/100g body weight) and tetracycline (12mg/100g body weight) for 14-days along with their corresponding one and two weeks recovery group compared with the normal control group.

Group	MEAN \pm S.E.M	Compared with Normal Control	Compared with Corresponding Treated group
Normal control "NC"	4.65 \pm 0.04		
Ampicillin treated "AT"	0.34 \pm 0.01	$P < 0.05$	
1weeks Ampicillin recovery "AR1"	0.31 \pm 0.00	$P < 0.05$	
2 weeks Ampicillin recovery "AR2"	4.56 \pm 0.09		$P < 0.05$
Cloxacillin treated CT"	0.34 \pm 0.01	$P < 0.05$	
1 week Cloxacillin recovery "CR1"	0.51 \pm 0.01	$P < 0.05$	
2 weeks Cloxacillin recovery "CR2"	4.57 \pm 0.17		$P < 0.05$
Tetracycline treated "TT"	0.30 \pm 0.00	$P < 0.05$	
1-week Tetracycline recovery "TR1."	0.59 \pm 0.01	$P < 0.05$	$P < 0.05$
2 weeks Tetracycline recovery "TR2"	4.57 \pm 0.01	$P < 0.05$	$P < 0.05$

Table 3: Mean \pm SEM values for initial and final body weight in grams, male organs (testis, epididymis) weight presented as percentage organ weight per body weight.

Group	Control	Ampicillin treated "AT" (4mg/100g/day)	1week Ampicillin recovery "AR1"	2 weeks Ampicillin recovery "AR2"	Cloxacillin treated "CT" (6mg/100g/day)	1 week Cloxacillin recovery "CR1"	2 weeks Cloxacillin recovery "CR2"	Tetracycline treated "TT" (12mg/100g/day)	1-week Tetracycline recovery "TR1."	2 weeks Tetracycline recovery "TR2"
Initial Body Weight	218.33 \pm 4.77	220.00 \pm 4.47	213.33 \pm 4.22	208.33 \pm 4.77	228.33 \pm 3.07	230.00 \pm 9.65	235.00 \pm 2.24	208.33 \pm 3.07	203.33 \pm 2.11	218.33 \pm 3.07
Final Body Weight	242.50 \pm 5.48	230.00 \pm 0.00	238.33 \pm 6.70	230.00 \pm 7.30	250.00 \pm 3.65	258.00 \pm 3.29	251.6 \pm 4.20	230.00 \pm 7.30	230.00 \pm 0.00	240.00 \pm 0.00
Testicular Weight	0.58 \pm 0.004	0.40 \pm 0.08	0.38 \pm 0.07	0.48 \pm 0.04	0.41 \pm 0.03*	0.43 \pm 0.02*	0.46 \pm 0.06*	0.53 \pm 0.04	0.54 \pm 0.04	0.48 \pm 0.04
Epididymis Weight	0.18 \pm 0.01	0.09 \pm 0.03	0.16 \pm 0.02	0.17 \pm 0.04	0.09 \pm 0.02*	0.12 \pm 0.00*	0.14 \pm 0.00***	0.15 \pm 0.00*	0.20 \pm 0.00**	0.19 \pm 0.01**
Prostate gland weight	0.13 \pm 0.01	0.08 \pm 0.00*	0.12 \pm 0.02	0.14 \pm 0.02	0.07 \pm 0.04	0.09 \pm 0.00	0.10 \pm 0.03	0.01 \pm 0.02	0.14 \pm 0.02	0.16 \pm 0.20

Keys* Significantly different from Normal Control ($P < 0.05$)** Significantly different from corresponding Treated group ($P < 0.05$)**Figure: Serum testosterone level in ampicillin (8mg/grm), cloxacillin (4mg/grm) and tetracycline (12mg/grm) treated rats along with their recovery groups compared with the control.****DISCUSSION**

The antifertility effect of these antibiotics: Ampicillin (4mg/100g/day), Cloxacillin (6mg/100g/day) and Tetracycline (12mg/100g/day) on male reproduction was confirmed by a significant reduction in serum testosterone level in all the treated rats after 14 days of treatment at the respective therapeutic dose (Ampicillin 0.34 ± 0.04 , Cloxacillin 0.34 ± 0.01 , Tetracycline 0.30 ± 0.00 nmol/L at $p < 0.05$). However, this was contrary to previous reports that some antibiotics such as fluphenazine and sulphasalazine have no effect on circulating serum testosterone level (6,7). The significant increase ($P < 0.05$) in the serum testosterone level recorded in all the animals treated

with Ampicillin, Cloxacillin and Tetracycline after two weeks of discontinuation of the antibiotic therapy showed that this antifertility effect is reversible. In addition, a significant change in testicular weight and Epididymal weight was recorded in all of Cloxacillin groups when compared with the Normal Control. One week and Two weeks recovery groups of Tetracycline also showed a significant increase in Epididymal weight when compared the corresponding treated group. Although, all the other rats in one and two weeks recovery groups of Ampicillin and Tetracycline showed an increase in both testicular and epididymal weight but they were not significant.

A recovery from the antifertility effect of these antibiotics was however established in all the two weeks recovery group of the three antibiotics used with a significant increase ($p < 0.05$) in serum testosterone level compared with the control. Pilot study carried out earlier had revealed that such antifertility effect on serum testosterone level in the antibiotics studied is not produced at therapeutic doses within the seven days treatment period for antibiotics. However, in situation of long-term administration of antibiotics e.g. in Gastroenterology patients, such antifertility effect will be expected.

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