Original Article

Quinolones on Sperm Count and Motility

Effect of Quinolones on Sperm Count and Motility in Male Albino Rats

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ABSTRACT

Objective: To determine the effects of quinolones on sperm count and motility in male albino rats.
Study Design: Experimental study
Place and Duration of Study: This study was conducted at the Pharmacology Department of Post Graduate Medical Institute, Lahore for the total duration of 84 days from May 2011 to July 211.
Materials and Methods: Eighty male albino rats were randomly divided into A,B, C and D groups each group having 20 albino rats. These groups were further subdivided into A1, A2, B1, B2, C1, C2, D1 and D2 having 10 albino rats in each group. Ciprofloxcin, ofloxacin and enoxacin dissolved in distilled water were given at 135mg/kg/day, 72mg/kg/day and 12.5mg/kg/day to groups A, B & C respectively for 12 weeks. Distilled water was given to group D being a control group for the same time period. The animals in subgroups Al, B1, Cl and D1 were sacrificed on 42nd day and samples were taken from epididymus. The caudal epididymus was dissected out and very small incision was made in the caudal epididymus. Seminal fluid was then squeezed on to the microscope slide. Epididymal sperms were assessed by calculating motile spermatozoa per unit area and expressed as percent motility. Epididymal sperm count were made by using haemocytometer and were expressed as million per ml of suspension.Rats in subgroup A2, B2, C2 and D2 were kept alive till 84th day after stopping drugs at 42nd day to find out if there was any reversible change in sperm count and motility after discontinuation of the treatment.
Results: Significant decrease in sperm count motility was observed as compared to the Control group. It was further noted that the values did not return back to normal even after the discontinuation of the treatment.
Conclusion: Quinolones reducesperm count and motility and should be used carefully for long term therapy.
Key Words: Floroquinolones,seminal fluid, sperm count,epididymus

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INTRODUCTION

Infertility affects approximately 15% of couples of reproductive age, and with nearly half of these cases resulting from male factor infertility this area of research is of great interest to both physicians and research scientists[1].

There are variety of prescription medications that can leads to male infertility, often temporary but sometimes permanently. These medications include, antidepressants, antihypertensives, H_2 receptor antagonists, disease modifying anti rheumatoid drugs, anti-cancer drugs and antibiotics. The antibiotics are often prescribed to deal with a variety of bacterial infections, often they are only taken for short period of time. Adverse effects on fertility are reversed after discontinuing the medication. Some of the antibiotics may be prescribed for longer time period which are suspected to interfere with the male fertility. These include nitrofurantoin, aminoglycosides, minocycline, macrolidies, sulfasalazine, and quinolones[2].

The fluoroquinolones are synthetic broad spectrum anti-microbial agents which are effective orally for a wide variety of infectious diseases. They are very potent agents having bactericidal activity against E.Coli, and different species of Neisseria, Enterobacter, Shigella, Campylobacter and Salmonella.Several new quinolones have activity against anaerobic bacteria[3]. The fluoroquinolones are very frequently prescribed for many clinical conditions. They have broad spectrum of antimicrobial activity and are considerably more potent for the infections of urinary tract[4]. They are effectively used for prostatitis caused by sensitive bacteria and in sexually transmitted diseases like gonnorrhoea. Ciprofloxacin, ofloxacin and enoxacin cure most of the patients with typhoid or enteric fever that is caused by Salmonellatyphi. They are also used in respiratory tract, bone, joint and soft tissue infections. These may be used as part of multiple drug regimens for the treatment of multiple drug resistant tuberculosis and atypical mycobacterial-infections[5].

The rate of multiplication of germ cells is very high that makes the reproductive system very sensitive to the toxic chemicals. Chemotherapy causes toxic effects on

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male gonad[6,7]. The toxic effects on reproductive system causes genetic damages which can be transferred from one generation to another. Keeping in view the mentioned facts it is very important to consider thesegenotoxic and cytotoxic effects of different agents[8]. It is noticed from previous years that there is marked decline in male fertility. Misuse of important drugs like antibiotics is one of the factors that caused this decrease in male fertility, such as ciprofloxacin, ofloxacin and enoxacin which come under the heading of fluoroquinolones[9]. Thus; in this study effects of ciprofloxacin, ofloxacin and enoxacin on sperm count and motility were evaluated.

MATERIALS AND METHODS

For the total duration of 84 days. Eighty male Albino rats, 7 weeks of age, weighing between 200-300 grams each were obtained from University of Veterinary and Animal Sciences Lahore. Ethical approval for animal study was taken from Ethical Committee of Post Graduate Medical Institute, Lahore. Animals were randomly divided into A, B, C and D groups having twenty albino rats each. These groups were further subdivided into A1, A2, B1, B2, C1, C2, D1 and D2 having 10 albino rats each in each group. Ciprofloxcin, ofloxacin and enoxacin dissolved in distilled water were given at doses of 135mg/kg/day, 72mg/kg/day and 12.5mg/kg/day to groups A, B & C respectively for 12 weeks.Group D served as control and was given 0.5ml distilled water orally for the same time period. Ciprofloxacin tablet of 500mg was dissolved in 5ml of distilled water.So, one ml contained 100mg of ciprofloxacin. Insulin syringe was used which has 100 sub divisions per ml and each sub division of 0.01 ml contained 1mg of ciprofloxacin. Then dosage for each albino rat was calculated according to the body weight according and given orally:

e.g. $\frac{135}{1000}$ x X b.w

Ofloxacin tablet of 400mg was dissolved in 4ml of distilled water so one ml contained 100mg of ofloxacin. Then dosage for each albino rat was calculated according to the weight as follows and given orally:

i.e. $\frac{72}{1000}$ x X b.w

Enoxacin tablet of 400mg dissolved in 4ml of distilled water so one ml contained 100mg of enoxacin. Then dosage for each albino rat was calculated according to the weight as follows and given orally:

i.e. $\frac{12.5}{1000}$ x X b.w

Standard doses were used as calculated by the above mentioned formulae. Standard doses are converted into mg/kg to adjust the dose according to the weights of the animals. The animals in subgroups Al, B1, Cl and D1 were sacrificed on 42nd day and samples were taken from epididymus. The caudal epididymus was dissected out. Very small incision was made in the caudal epididymus. Seminal fluid was then squeezed on to the microscope slide. Epididymal sperms were assessed by calculating motile spermatozoa per unit area and expressed as percent motility where as sperms were counted were by using haemocytometer and expressed as million per ml of suspension. Rats in subgroup A2, B2, C2 and D2 were kept alive till 84th day after stopping drugs at 42nd day to find out if there was any reversible change in sperm count and motility after discontinuation of the treatment.The tests were carried out in the Pharmacology Department of Post Graduate Medical Institute, Lahore. Statistical analysis was done by using SPSS version 16.ANOVA was used to compare the sperm count and motility in different groups and P value <0.05 was taken as significant.

RESULTS

The number of sperm ($10x^6$) in group A1 was 27.50 ± 4.90, in A2 was 30.24 ±7.746, in B1 was 24.24±3.24, in B2 was 26.05±5.56, in C1 was 27.37±6.31, in C2 was 26.96±6.05, in D1 was 56.70±9.56, in D2 was 56.68±11.63. The average number of sperm ($10x^6$) in control group was higher as compared to experimental groups sacrificed at 42nd and 84th day, p-value < 0.05. The pairs experimental vs control i.e A1 vs. D1, A1 vs. D2, A2 vs. D1, A2 vs. D2, B1 vs. D1, B1 vs. D2, B2 vs. D1, B2 vs. D2, C1 vs. D1, C1 vs. D2, C2 vs. D1, and C2 vs. D2 were statistically significant while the rest of the pairs were insignificant.

The motility in A1 was 32.51 ± 3.70%, in A2 was 31.67 ± 7.93%, in B1 was 24.46± 4.71, in B2 was 23.84 ± 4.60, in C1 was 27.40 ± 7.00, in C2 was 25.25 ± 3.78,where as in control sub groups, in D1 was 54.68 ± 4.78 and in D2 was 51.81±8.83. So, the mean motility was statistically higher in control group as compared to experimental groups sacrificed at 42nd and 84th day, p-value < 0.05. The pairs A1 vs. A2, A1 vs. C1, A2 vs. C1, B1 vs. B2, B1 vs. C1, B1 vs. C2, B2 vs. C1, B2 vs. C2, C1 vs C2, and D1 vs. D2 were statistically insignificant while all other pairs were significant in this study.

DISCUSSION

The fluoroquinolones are synthetic broad spectrum anti-microbial agents which are effective for a wide variety of infectious diseases. The therapeutic and adverse effects of fluoroquinolones have been well documented. However, the result of our experimental study revealed that prolonged administration of therapeutic doses of fluoroquinolones such as ciprofloxacin, ofloxacin and enoxacin promoted reproductive toxicity in rats. The reduction in sperm count and motility are the evidence for this toxicity.

It was seen by wait et al. (1989) that fluoroquinolone such as ciprofloxacin causing inhibition of oxidative drug metabolism has no anti-steroidogenicside effects[10]. Our study is congruent with the study of Zobeiri et al (2013) who proved that long time Ciprofloxacin administration in mice caused major alterations in Germinal Epithelium (GE) intracytoplasmic biochemistry leading to loss of physiological function and ultimately result in fertility problems. Ciprofloxacin is able to imbalance serum levels of gonadotropins and testosterone levels by affecting Leydig cells[11]. Khaki et al (2009) showed that Ofloxacin caused negative effects on testis architecture and germinal cells damages in rats, that can ultimately lead to infertility[12]. In a study male patients when given 250mg ciprofloxacin twice daily did not show difference in sperm quality and was without effect on spermatogenesis[13]. Our study is again in accordance with the study of King et al who proved that at pharmacologic concentration, ciprofloxacin adversely affected human sperm motility in vitro[14].

It has been reported that decrease in sperm count and motility are valid indices of male infertility. It is also stated that the disruption of seminiferous epithelium is indicative of male reproductive hazard. Therefore our experimental results suggest a gonadotoxic potential of fluoroquinolones.One of the reason for this toxicity could be explained on the basis that fluoroquinolones interfere with the energy production process required for sperm vitality and motility[15]. A study conducted by Demir et al.,(2007) had shown similar results that ciprofloxacin treatment for 10 days in rats resulted in marked reduction in sperm count and motility[16]. Nagai A et al(2002) have also reported that two weeks treatment with enoxacin is sufficient to detect toxic effects on reproductive organs in rats[17]. In another study showed that ofloxacin at a dose of 75mg /kg per day had almost the highest potential in terms of impairment of the rat testicular function[18].

The present study indicated that administration of fluoroquinolone for 42 consecutive days resulted in marked reduction in sperm countand motility as compared to respective control group (P<0.001)which is significant. Effect on sperm count was equal in all the experimental groups while ciprofloxacin caused the least reduction in spermatozoa motility. This is in agreement with that of Abdullrah et al and Khaki et al[19,20] who reported that ciprofloxacin administration for 15 days and 60 days respectively in rats caused a marked reduction in sperm counts and motility and it is seen that these changes, persist as such even after 42 days of withdrawal of drugs having P value <0.001 which is significant. These findings are in consistent with the current study.

It is evident that even after discontinuation of the drugs after 42nd day of treatment the sperm count and motility did not return back to the normal levels. This seems to be due to necrosis of the interstitial leydig cells[21, 22].

CONCLUSION

The present study concludesthat the use of quinolones results in reduction of sperm count and motility;however, more research work is required to find out the toxicity and exact mechanism operating at cellular level which suppresses the synthesis of sperms. This study however adds concern to the widespread and indiscriminate use of fluoroquinolones and recommends that these drugs should be used with great caution.

Conflict of Interest: The study has no conflict of interest to declare by any author.

REFERENCES

1. Sharlip ID, Jarow JP, Belker AM, Lipshultz LI, Sigman M, Thomas AJ, et al. Best practice policies for male infertility. Fertil Steril 2002;77(5):873-82.
2. 2. Demir A, Turker P, Onol FF, Sirvanci S, Findik A, Tarcan T. Effects of experimentally induced Escherichia coli epididymo-orchitis and ciprofloxacin treatment on rat spermatogenesis. International journal of urology : official J Japanese Urolog Assoc 2007;14(3):268-72.
3. Stahlmann R. Quinolones. Encyclopedia of Molecular Pharmacol 2008:1055-8.
4. Warren JW, Abrutyn E, Hebel JR, Johnson JR, Schaeffer AJ, Stamm WE. Guidelines for antimicrobial treatment of uncomplicated acute bacterial cystitis and acute pyelonephritis in women. Clin Infect Dis 1999;29(4):745-59.
5. Venugopal D, Kumar S, Isa M, Bose M. Drug resistance profile of human Mycobacterium avium complex strains from India. Ind J Med Microbiol 2007;25(2):115.
6. Horstman MG, Meadows GG, Yost GS. Separate mechanisms for procarbazine spermatotoxicity and anticancer activity. Cancer Res 1987;47(6): 1547-50.
7. Zhang QX, Yang GY, Li JT, Li WX, Zhang B, Zhu W. Melamine induces sperm DNA damage and abnormality, but not genetic toxicity. Regulatory toxicology and pharmacology: RTP 2011;60(1): 144-50.
8. Bairy KL, Kumar G, Rao Y. Effect of acyclovir on the sperm parameters of albino mice. Ind J Physiol Pharmacol 2009;53(4):327-33.
9. Abd-Allah AR, Gannam BB, Hamada FM. The impact of ofloxacin on rat testicular DNA: application of image analysis. Pharmacological research : the official J of the Italian Pharmacological Society 2000;42(2):145-50.
10. Waite NM, Edwards DJ, Arnott WS, Warbasse LH. Effects of ciprofloxacin on testosterone and

cortisol concentrations in healthy males. Antimicrobial agents and chemotherapy. 1989; 33(11):1875-7.
11. Zobeiri F, Sadrkhanlou R-A, Salami S, Mardani K. Long-Term Effect of Ciprofloxacin on Testicular Tissue: Evidence for Biochemical and Histochemical Changes. Int J Fertil Steril 2013; 6(4):294-303.
12. Arash Khaki AAK, Iraj S, Bazi P, Imani SAM. Comparative Study of Aminoglycosides (Gentamicin & Streptomycin) and Fluoroquinolone (Ofloxacin) Antibiotics on Testis Tissue in Rats: Light and Transmission Electron Microscopic Study. Pak J Med Sci 2009;25.
13. Merino G, Carranza-Lira S, Murrieta S, Rodriguez L, Cuevas E, Moran C. Bacterial infection and semen characteristics in infertile men. Archives of Androl 1995;35(1):43-7.
14. King A, Kolb U, Szuszkiewicz E. Why low-mass Black Hole Binaries are transient. The Astrophysical J 1997;488(1):89.
15. Folgerø T, Bertheussen K, Lindal S, Torbergsen T, Øian P. Andrology: Mitochondrial disease and reduced sperm motility. Human reproduction 1993;8(11):1863-8.
16. Demir A, Türker P, Önol FF, Sirvanci S, Findik A, Tarcan T. Effect of experimentally induced Escherichia coli epididymo-orchitis and ciprofloxacin treatment on rat spermatogenesis. Int J Urol 2007;14(3):268-72.
17. Nagai A, Miyazaki M, Morita T, Furubo S, Kizawa K, Fukumoto H, et al. Comparative articular toxicity of garenoxacin, a novel quinolone antimicrobial agent, in juvenile beagle dogs. J Toxicological Sci 2002;27(3):219-28.
18. Andreessen R, Sudhoff F, Borgmann V, Nagel R. Results of ofloxacin therapy in andrologic patients suffering from therapy-requiring asymptomatic infections. Andrologia 1993;25(6):377-83.
19. Abd-Allah AR, Aly HA, Moustafa AM, Abdel-Aziz A-Ah, Hamada FM. Adverse testicular effects of some quinolone members in rats. Pharmacological research 2000;41(2):211-9.
20. Khaki A, Heidari M, Ghaffari Novin M, Khaki AA. Adverse effects of ciprofloxacin on testis apoptosis and sperm parameters in rats. Int J Reproductive Bio Med 2008;6(2):71-6.
21. Kianifard D, Sadrkhanlou RA, Hasanzadeh S. The Ultrastructural Changes of the Sertoli and Leydig Cells Following Streptozotocin Induced Diabetes. Iranian J Basic Med Sci 2012;15(1):623-35.
22. Hong CY, Park JH, Ahn RS, Im SY, Choi H-S, Soh J, et al. Molecular Mechanism of Suppression of Testicular Steroidogenesis by Proinflammatory Cytokine Tumor Necrosis Factor Alpha. Molecular and Cellular Biol 2004;24(7):2593-604.