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EFFECT OF SOME ANTI - RHEUMATIC DRUGS ON MALE FERTILITY IN RATS

(With One Fig and 2 Tables)

By

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تأثير بعض أدوية علاج الروماتيزم على خصوبة الذكور في الفئران

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تم فى هذا البحث دراسة تأثير ديكلوفيناك الصوديوم، كيتوبروفين وبيروبروفين (على مستوى جرعتين لكل دواء) على الاعضاء التناسليه لذكور الفئران. وقد اعطيت الادوية لذكور الفئران بطريق الفم لمدة ٦٥ يوماً متتالياً ، ثم أخذ وزن الاعضاء التناسلية وصورة السائل المنوى ومستوى هرمون التستوستيرون فى الدم وكذلك الفحص الهستوباثولوجى للاعضاء التناسلية كخواص لتقييم كفاءة الاعضاء التناسلية للفئران.

وقد اظهرت النتائج ان اعطاء هذه الادوية لذكور الفئران لمدة ٦٥ يوماً متتالياً يسبب نقصاً فى وزن الاعضاء التناسلية وكذا فى عدد وحركة الحيوانات المنوية مصاحباً بزيادة فى نسبة الحيوانات المنوية الغير طبيعية . وقد أحدثت هذه الدراسة ايضاً نقصاً فى مستوى هرمون التستوستيرون فى دم الفئران المعالجة ، كما أظهر الفحص الهستوباثولوجى أن الادوية المستخدمة تسبب تغيرات اضمحلالية فى الأنيبيبات المنوية للخصية.

SUMMARY

The effect of some anti-rheumatic drugs on male reproductive organs and fertility was studied on 7 equal groups of 10 rats each. Oral administration of diclofenac sodium, ketoprofen and pirprofen in doses of 1.0 and 2.0; 1.0 and 2.0 and 7.5 and 15 mg/Kg b.wt, respectively for 65 successive days significantly decreased testicular weights, epididymal sperm cell concentration, progressive motility percentage and number of seminiferous tubules containing spermatozoa as compared with those of the control. These drugs significantly increased the total abnormalities of sperms. Moreover, the tested drugs significantly decreased testosterone level in rat's serum. Histopathological examination of testes and accessory glands of treated rats showed degenerative changes in some seminiferous tubules of the testes.

INTRODUCTION

Rheumatic diseases are chronic in nature as they persist for months or years and associated with excessive connective tissue formation. Several anti-rheumatics were introduced for treatment of rheumatic diseases as rheumatic fever, rheumatic arthritis, osteo-arthritis and ankylosing spondylitis *(CROSSLAND, 1980 and GOODMAN et al., 1985).* Long-term administration of anti-rheumatics resulted in hepatic and renal damages *(MEYERS et al., 1976).* Moreover, agranulocytosis, leukopenia, neutropenia, thrombocytopenia and gastro-intestinal disorders were also recorded *(WADE, 1977 and GOODMAN et al., 1985).*

However, chemical structure, pharmacokinetic, pharmacodynamic and toxicity of most anti-rheumatic drugs were investigated *(STIERLIN and FAIGLE, 1979; MIRALLES et al., 1987 and SMITH & REYNARD, 1992),* their effects on reproductive system and reproduction requires investgation. Therefore, the present work was designed to study the effect of prolonged administration of some newly unexpensive non-steroidal antirheumatics on the reproductive system and fertility of male rats.

MATERIAL and METHODS

Drugs:

1. Diclofenac sodium (Voltarin)$^{(R)}$; Ciba-GEIGY Limited, Basle, Switzerland was obtained as tablets of 50 mg each.

2. Ketoprofen (Profenid)(R); Alexandria Pharmaceutical Co. under licence of RHONE POLENC-Paris, France was obtained as capsules, containing 50 mg each.
3. Pirprofen (Rengasil)(R); Ciba-GEIGY Limited, Basle, Switzerland is available as capsules of 400 mg each.

These drugs were given in 2 dose levels (half therapeutic and therapeutic) and calculated from that of human according to *PAGET and BARNES (1964).*

Animals:

Mature male albino rats of about 10-12 months old and 200-220 g b.wt were used. Animals were fed on balanced ration watered ad-libitum.

Experimental:

Male rats of proven fertility were divided into 7 groups of 10 animals each. The first group was kept as a control while the others were orally given diclofenace sodium, Ketoprofen and pirprofen in doses of 1.0 and 2.0; 1.0 and 2.0 and 7.5 and 15 mg/Kg b.wt, respectively. Drugs were administered daily for 65 successive days to cover a complete spermatogenic cycle *(HERSHBERGER et al., 1969).* Blood samples were obtained from the orbital plexus of each rat at 0, 15, 30, 45 and 65 day of administration for obtaining clear serum to assay testosterone using radioimmuno assay method *(JEFFCOATE, 1971).* Rats were sacrificed by decapitation, their sexual organs were dissected out, dried between two filter papers and weighed. Weights of testes and accessory glands of each rat were calculated in relation to its body weight. The epididymal content of each rat was obtained by cutting the tail of epididymis and squeezed it in a clean watch glass. It was diluted 10 times with 2.9% sodium citrate dihydrate solution and thoroughly mixed to estimate the progressive motility and concentration of sperms *(BEARDEN and FLUQUARY, 1980).* Eosin-nigrosin stained smears were made to determine the sperm abnormalities. Testes, epididymis, prostate glands and seminal vesicales of each rat were taken for histopathological examination using Harris Haematoxylin and Eosin method *(LUNA, 1968).* The percentage of seminiferous tubules containing spermatozoa were also determined *(YUNDA and KUSHNIRUK, 1974).*

RESULTS

The effects of prolonged administration of the studied drugs on male fertility in rats were recorded in Table 1.

Oral administration of diclofenac sodium, ketoprofen and

pirprofen in doses of 1.0 and 2.0; 1.0 and 2.0 and 7.5 and 15 mg/Kg b.wt, respectively for 65 successive days significantly decreased the weights of testes. A significant decrease in the weights of accessory glands was induced only by the large doses of ketoprofen and piroprofen. The tested doses of drugs significantly decreased sperm cell concentration and percentages of sperm motility and seminiferous tubules containing spermatozoa but increased the total abnormalities of sperms. Sperm abnormalities were characterized by bent tails and headless sperms. Moreover, the tested drugs significantly decreased testosterone level in rat's serum (Table 2). Histopathological examination of testes and accessory glands of treated rats showed that diclofenac sodium, ketoprofen and pirprofen induced degenerative changes in some seminiferous tubules of the testes (Fig. 1). Few sperms were seen in epididymal tubules of teated rats. Seminal vesicles and prostate glands showed degenerative changes and the lumen did not contain abundant secretions as in the control.

DISCUSSION

Diclofenac sodium, ketoprofen and pirprofen are proved to be highly effective in treatment of rheumatic arthritis, osteo-arthritis and ankylosing spondylitis *(CROSSLAND, 1980);* attacks of biliary colics *(KARACHALIOS et al., 1988)* and severe pain after orthopedic surgery *(FORTUNATO, 1988).* No available literature could be obtained on the effect of studied anti-rheumatics on reproductive organs and fertility of males.

The results revealed that oral adminsitration of diclofenac sodium, ketoprofen and pirprofen to mature male rats for 65 successive days significantly decreased testicular weight, sperm cell concentration and percentages of sperm cell motility and seminiferous tubules containing spermatozoa. Moreover, a decrease in palsma testosterone levels was observed in the treated groups as compared to control one. Histopathological examination of testes, seminal vesicles and prostate glands shwoed degenerative changes.

The present decrease in epididymal sperm characters could be explained by the degenerative changes seen in testicular and accessory glands structures or by the decrease in testoseterone levels. These effects could impaire or even stop the process of spermatogenesis resulting in infertility of male rats. *PAZ et al. (1978)* attributed the reduction in sperm motility to a decrease in testosterone production.

The decrease in progressive motility and increase in sperm abnormalities may be attributed to the inhibitory effect of the studied drugs to prostaglandine synthesis *(KOICHI et al., 1988)*

or their crossing the blood-testes barrier as their molecular weights are less than 600 *(HARBISON et al., 1975)*.

It is concluded that the studied anti-rheumatic drugs when given orally to male rats causes in a dose-dependent fashion, a decrease in their reproductive efficiency. Therefore, great attention should be taken during their prolonged administration to males used for breeding.

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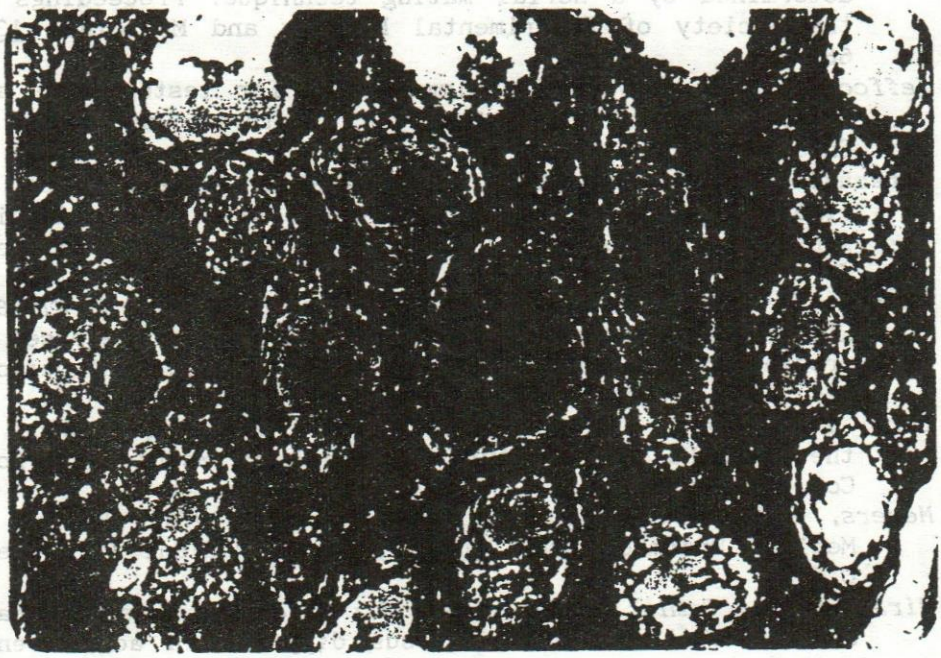

Fig. 1: Showing degenerative changes in some seminiferous tubules in testes of a rat given diclofenac sodium in a dose of 2 mg/Kg b.wt. for 65 successive days.

Table 1: Effect of prolonged oral administration of some antirheumatic drugs (for 65 successive days) on the weight of sexual organs, epididymal sperm characters and number of seminiferous tubules containing spermatozoa. (n=10).

Drugs	Dose in mg/kg b.wt.	weight of sexual organs (g/100g b.wt)			Epididymal sperm characters			Number of seminiferous tubulus containing spermatozoa %
		Testes	seminal vesicles	prostate gland	concentration (10^6/ml)	Motility (%)	Abnormality (%)	
Control	0	1.76±0.04	0.38±0.007	0.23 ±0.014	288.3±11.0	83.3±1.6	2.1±0.16	93.0±2.21
Diclofenac sodium	1.0	1.52±0.07**	0.37±0.019	0.21±0.018	221.0±7.4***	53.3±3.3***	3.3±0.21**	84.0±2.87*
	2.0	1.5±0.02***	0.36±0.026	0.22±0.014	193.0±14.9***	35.0±2.2***	4.8±0.16***	79.0±2.71**
Ketoprofen	1.0	1.48±0.11*	0.33±0.027	0.21±0.016	231.0±7.6***	51.66±3.0***	4.5±0.22***	83.0±2.33**
	2.0	1.4±0.12**	0.31±0.026*	0.15 ±0.009*	213.3±9.8***	45.0±3.8***	5.1±0.3***	81.0±2.37**
Pirprofen	7.5	1.47±0.07**	0.35±0.025	0.22±0.016	195.0±11.4***	48.3±3.8***	10.5±0.5***	79.0±2.44**
	15	1.47±0.09**	0.31±0.025*	0.17 ±0.015*	185.0±12.5***	40.0±3.6***	20.8±0.54***	77.0±2.16***

* Significant at $p<0.05$.

** Significant at $p<0.01$.

*** Significant at $p<0.001$.

Table (2): Effect of oral administration of some anti-rheumatics on serum testosterone of male rats (Radioimmunoassay).(n = 10).

Drugs	Dose (mg/kg b.wt)	Level of testosterone (ng/ml serum) after (days)				
		0	15	30	45	65
Control	0.0	7.30 ± 0.24	7.58 ± 0.25	7.52 ± 0.20	8.34 ± 0.29	9.17 ± 0.31
Diclofenac	1.0	7.21 ± 0.28	6.72* ± 0.20	5.25*** ± 0.32	4.82*** ± 0.28	3.62*** ± 0.24
Sodium	2.0	7.84 ± 0.26	6.16** ± 0.29	4.93*** ± 0.30	4.25*** ± 0.20	3.08*** ± 0.28
Ketoprofen	1.0	8.15 ± 0.32	6.58** ± 0.24	5.14*** ± 0.26	4.71*** ± 0.29	3.51*** ± 0.26
	2.0	7.95 ± 0.28	6.14** ± 0.26	4.90*** ± 0.20	4.17*** ± 0.25	2.87*** ± 0.23
Pirprofen	7.5	7.56 ± 0.32	6.51** ± 0.29	5.10*** ± 0.28	4.62*** ± 0.32	3.50*** ± 0.28
	15.0	7.92 ± 0.20	6.22** ± 0.30	4.87*** ± 0.24	4.12*** ± 0.26	2.84*** ± 0.20

Significant at :

* $P < 0.05$
** $P < 0.01$
*** $P < 0.001$