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EFFECT OF OPIOID ANTAGONISTS ON HORMONAL MODULATION AND THEIR RELATION TO MALE REPRODUCTION

By

EL-SHELTAWI, M.;ESSAWY, S.A.; GALILA A. EL-RAFEY; ABD-EL-MALAK, G. and MAKKAR, N.N.*

Animal Reproduction Research Institute
* Fac. Med., Zagazig Univ. (Benha).

SUMMARY

Twenty four adult male Sprague Dawley rats were used to study the effect of acute administration of naloxone (4mg/kg b.w.) on FSH, LH and prolactin levels and its relation to male sexual functions on both intact (n=12) and castrated male rats, (n=12) In case of the intact male rats FSH and LH levels were increased significantly (P<0.01) in the treated group. However, prolactin level was decreased significantly (P<0.01). Also, the viability and motility percentages of the epididymal sperm were decreased significantly in treated group. Minimal degenerative changes were observed in treated group. No significant changes were observed in exploratory and sexual behavior parameters in both groups.

In sexually inactive rats (chronically castrated 2 weeks before experiment then treated with 2.5 mg/kg. b.w. testosterone propinate). Both FSH and LH levels were increased significantly (P<0.01) in treated group. However, the prolactin level was decreased significantly (P<0.01). The explopratory and sexual behaviour parameters were not changed.

INTRODUCTION

The neuroanatomic pathway and hormonal changes through which opioids and opioid antagonists exert their action on sexual functions. are poorly understood (Pfaus and Gorzy active and inactive male alka, 1987; Agmo and Paredes, 1988). Naloxloxone is, the N-allyl derivative of narcotic analgesic oxymorphone, an opioid antagonist (Martin et al., 1976). The present study is an attempt to clarify the effect of active administration of naloxone on gonadotrophins and prolactin levels and its relation to the male sexual performance on both sexually active and inactive male rats.

MATERIALS AND METHODS

Twenty four adult male Sprague-Dawley rats weighting 130-140 grams weight were used in this study. Animal were fed on mixed cereal diet together with green fodder and dried skimmed milk (Waynforth, 1980).

Experiment I:

Twelve intact male rats were used to study the acute administration of naloxone and its relation to hormonal changes. Animals were subdivided into two equal groups: group A (6 rats) were injected (i.p) with 1.5 ml saline, 15 minutes before the beginning of the experiment. On both groups; exploratory behavior parameters (Sniffing, rearing and investigating movement) were recorded for both latency (is the time ellapsed from the begning of the experiment until

end of acting performance by the rate expressed in seconds) and frequency (is the number of acts performed by the rat per 30 minutes) according to Clark et. al., (1988). Sexual behavior parameters (mounting, intromission and ejaculation) were also recorded for both latency (expresed in minutes) and frequency according to Meyerson et al., (1988).

At the end of 30 minute-behavioural test, animals of both groups, were sacrificed and trunk blood samples were collected , serum was separated and kept at -20°C until hormonal assay. LH and FSH hormones were assayed by direct radioimmunoasay according to the method of Davidson and Henry (1974) and prolactin was assayed following the method of Djusing (1981).

Epididymal spermatozoa were collected and examined for sperm concentration (million/ml), sperm motility (%) and live (%). Abnormal forms (%) were dtermined from film stained with eosin & negrosin (Blom, 1983).

Samples of testes from both groups were processed for histological examination and stained with Periodic Acid Schiff (PAS) technique for detection of neutral mucopolysaccarides (Pearse, 1985).

Experiment II:

Twelve male rats were used to study the effect of acute administration of naloxone in a model of sexually inactive rats which performed through chronic castration (2 weeks before experment) and treatment with a low dose of Testosteron Propionate (TP) (2.5mg/Kg B.W.) Animals were subdivided into two equall groups:

Group.I: (6 rats) were treated with TP in Sesame oil 48 hrs before saline (I/P) injection.

Group.II: (6 rats) were treated with TP an naloxone (4 mg/KgB.Wt). I/P.

Both exploratory and sexual behavio parameters were recorded. Bloo sample were collected for hormon assay.

Statistical analysis:

Student "t" test was applied to compare betwee the terated and the control groups in each experiment (Snedecor and Cochran, 1967).

RESULTS

Experiment I:

Effects of acute naloxone administration on exploratory behavior casterated male rats are shown in Table, 1. The changes in latency and frequency of sniffing, rearing and investigating movement are insignificant between treated (group B) and control (group a).

Parameters of sexual behavior are presented in Table 2. The difference between the treated and the control group were insignificant.

LH, FSH and Prolactin levels in both groups are shown in Table (3) . It was obvious that, there was a significant ($P< 0.01$) increase in the level of both FSH and 4H in treated group (group. B), but the level of prolactin decreased significnalty in treated compared to control gorup.

Charecters of epidedymal sperm (count, viability %, motility % and abnormal forms %) were tabulated in Table 4. The differences between treated and control groups were significant for both viability and motility while there were no significant differences in sperm count and the percentage of abnormal forms.

Table 1: Parameters of exploratory behavior in treated and control groups

Parameters	GroupA (control)		GroupB (expermental)	
	Latency	Frequency	Latency	Frequency
Sniffing	2.00±0.00	49.00±0.73	2.00±0.00	50.00±0.73
Rearing	7.00±0.58	52.00±0.86	8.00±0.73	50.00±0.73
Investigating mov	145.00±0.73	8.00±0.37	155.00±0.73	9.00±0.52

Values are represented as mean ± S.E.

Parameters of sexual behavior are presented in Table 2. The difference between the treated and the control group were insignificant.

Table 2: Parameters of sexual behaviour in treated and control groups

Parameters	GroupA (contrl)		GroupB (expermental)	
	Latency	Frequency	Latency	Frequency
Mounting	5.23± 0.07	7.00± 0.37	5.25 0.08	7.00 ±0.37
Intromission	5.67 ±0.10	8.33± 0.33	5.55± 0.11	8.83± 0.31
Ejaculation	14.85± 0.1.	--	14.93 ±0.39	--

Values represented as mean±S.E.

LH, FSH and Prolactin levels in both groups are shown in Table (3). It was obvious that, there was a significant ($P<0.01$) increase in the level of both FSH and LH in treated group (groupB) ,but,the level of prolactin decreased significantly in treated compared to control group.

Table 3: LH, FSH and prolactin levels (ng/ml) in naloxone treated male rats.

Hormones	GroupA (control)	GroupB (expermental)
LH (mIU/ml)	2.33 ±0.12	6.30 ±0.13*
FSH (mIU/ml)	2.52 ±0.08	6.50±0.13*
Prolactin (ng/ml)	5.83±0.13	3.00 ±0.08*

Values represented as mean±S.E.
*Significant $P<0.01$

Table 4 Characters of epidedymal sperms

charecter	Group A (control)	Group B(expermental)
Sperm Count in 10^6	86.50±2.16	86.00±1.38
Viability %	83.33±2.35	17.67±1.16*
Motility %	76.67±1.25	11.67±0.68*
Abnormal Forms %	17.83±1.14	17.50±1.01

values are represented as mean - S.E.
* Significant at $P < o.o1$

Table 5: Parameters of Exploratory Behavior in Both Treated and Control Groups

Parameters	Group I		Group II (treated)	
	Latency	Frequancy	Latency	Frequancy
Sniffing	2.00±0.00	53.0±0.97	2.00±0.00	50.0±0.58
Rearing	6.00±0.26	52.0±0.58	8.00±0.37	56.0±0.58
Investigating	149.0±0.93	9.0±0.26	144.0±0.93	8.0±0.73

.Latency was expressed in seconds.

sexual behavior activities were tabulated in table 6. It was clear that,the acute adminstraton of naloxonc lead to an insignificant changes in sexual activities.

Table 6: Sexual Behavior Activities in Treated and Control Groups

ACTIVITY	Group I		Group II(treated)	
	Latency	Frequency	Latency	Frequency
Mountain	1.78±1.13	2.33±1.50	1.80 1.14	2.50±1.59
Intromission	1.92 ±1.21	3.00±1.91	1.93±1.22	3.17±2.01
Ejaculation	5.29±3.32	----	5.15±3.26	----

Table 7: Scrum FSH, LH and Prolactin Levels in Treated and Control Groups

Hormonc	Group I	Group II (treated)
FSH (mIU/ml)	2,23±0.06	6.20±0.10*
LH (mIU/ml)	2.10±0.07	6.12±0.10*
Prolactin (ng/ml)	5.55±0.08	3.20±0.06*

* Significant at P<0.01

Naloxone administration in a dose of 4 mg/kg b.w., i.p. caused a minimal degenerative changes in the seminiferous tubules included small areas of epithelial disorganization or missing cells and chromatin margination of spermatoocytes (Fig. 1. The interstitial cells appeared normal with PAS positive fine granules and vaculated cytoplasm (Fig. 2).

Expermint II:

Effects of acute naloxone to castrated TP primed rats on exploratory behavior are shown in table 5. There were no significant differences in all parameters of exploratory behavior.

Sexual behavior activities were tabulated in table 6. It was clear that, the acute adminstraton of naloxone lead to an insignificant changes in sexual activities.

FSH, LH and prolactin levels, in both treated and control groups were tabulated in table 7. The leves of FSH and LH were significantly increased in treated groups ($P < 0.01$). However, prolactin level was significantly decreased ($P < 0.01$).

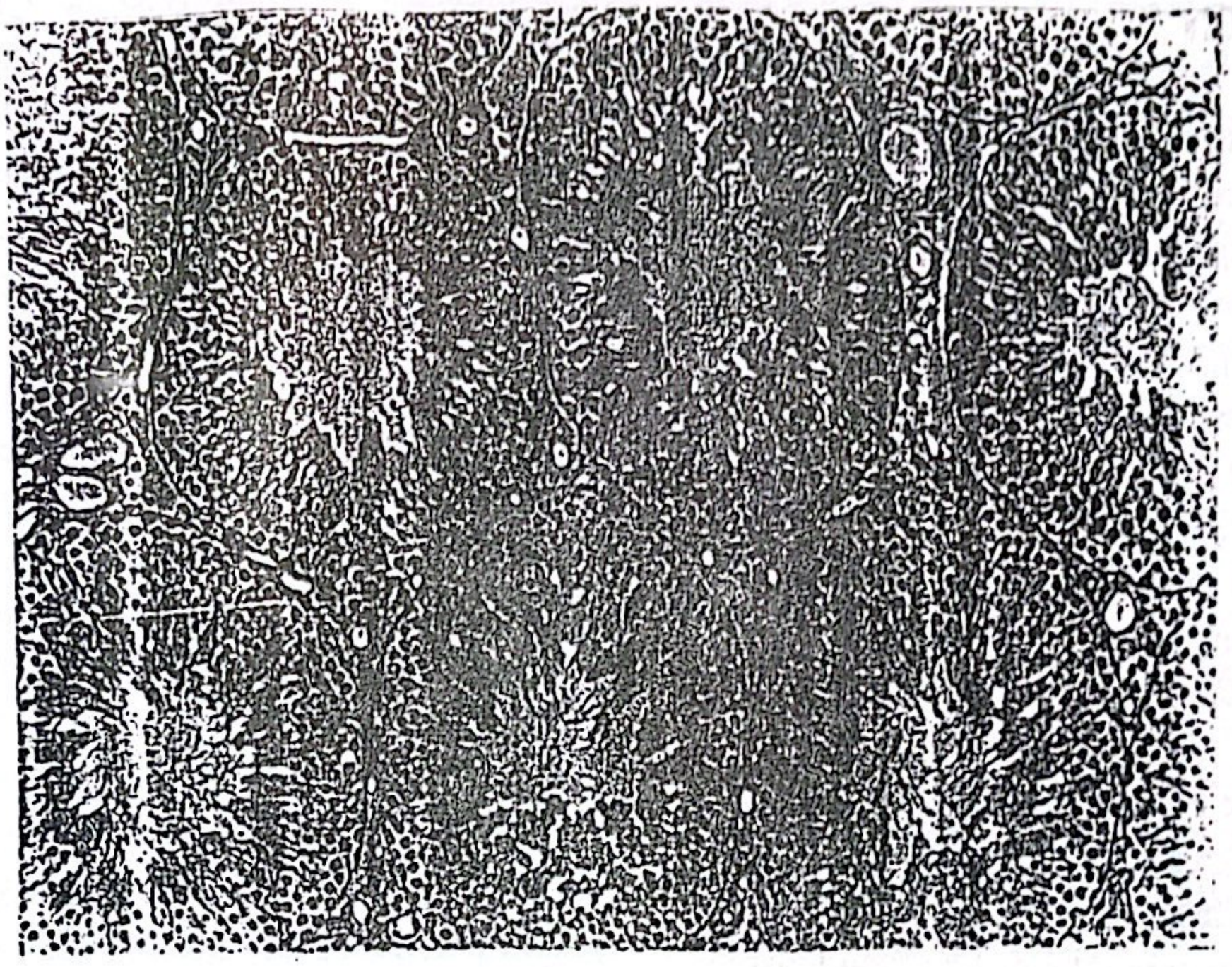

Fig. 1: Cross section in the testes of a rat injected with Naloxone showing normal seminiferous epithelium with minimal degenerative changes appeared in the form of epithelial disorganization or missing cells. Stain: PSA X 200.

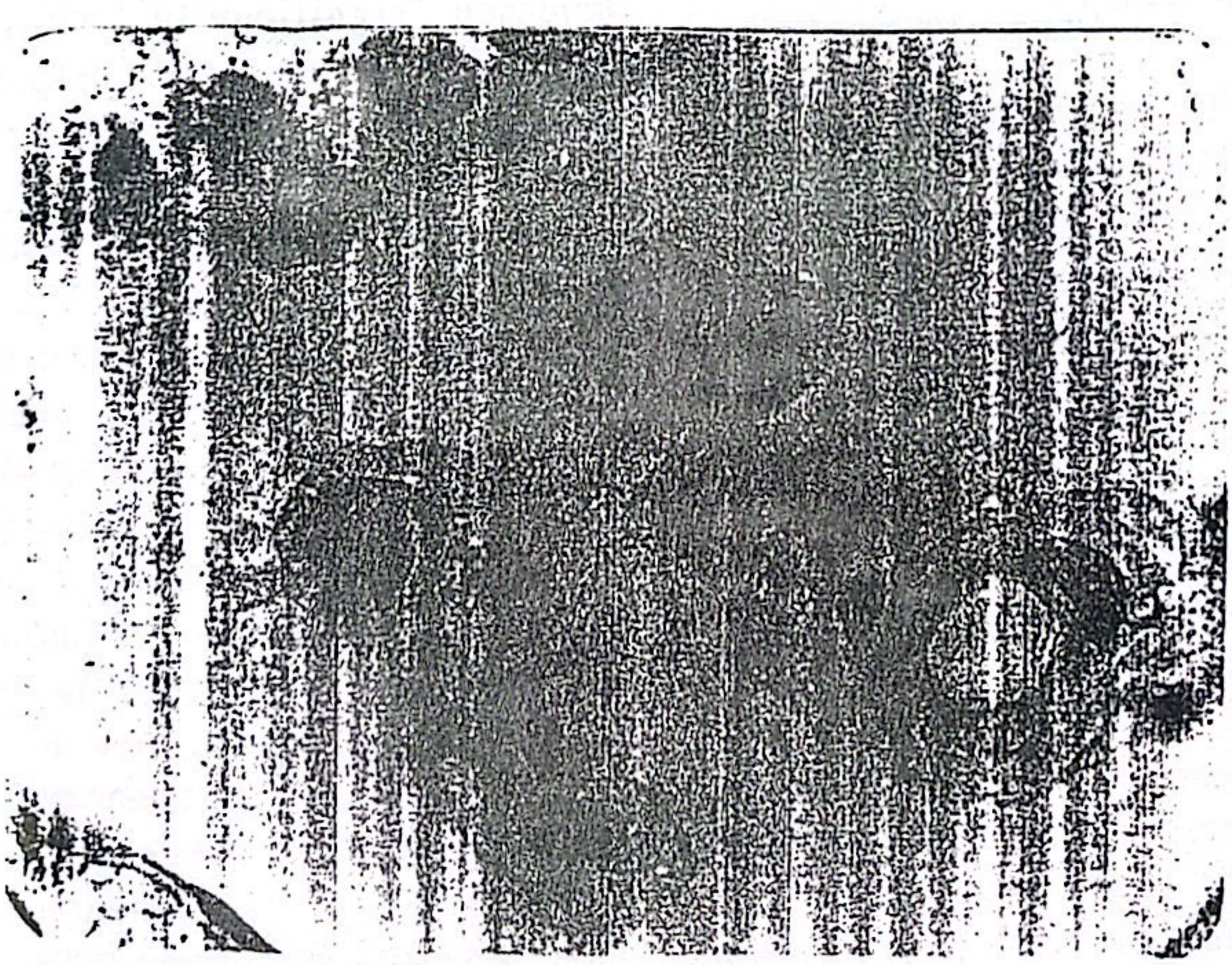

Fig. 2: Cross section in the testes of a rat injected with Naloxone showing normal interstitial cells with fine PAS positive granules and vacuolated cytoplasm. Stain: PSA X 1000.

DISCUSSION

In the present study 4 mg/kg b.w. naloxone administrated i.p. to intact sexually active rats caused no significant changes in both exploratory and sexual behavior parameters. These results are in line with those presented by many other authors (Gess et al., 1979; Myers and Baum, 1979; Sachs et al., 1981; Pfaus and Gorzalka, 1987; Deviche and Moore, 1987). This could be attributed to preferantially block opioid receptor (mu subtype) more than any other receptors (Rothman and Westfall, 1983).

Furthermore, acute naloxone administration resulted a significant increase in serum FSH and LH levels. These are in agreement with those of other investigators (Blank et al., 1979; Cicero et al., 1979; Grossman et al., 1981). This increase could be due to the effect of naloxone in stimulating the release of GnRH, norepinephrne and dopamine from adult male rat brain (Rasmussen et al., 1988).

Serum prolactin level was reduced significantly in treated group and this is in line with many other investigators who reported a significant depression of prolactin level in sera of both rat and monkey after naloxone administration (Bruni et al., 1977; Gold et al., 1979; Rossier et al. 1980). This effect may be due to inhibition of the tonic PRL effect of endogenous opioid peptides (EOPS) and or through antagonising the hypothalamic serotonergic prolactin releasing effect (Perersen and Barraclaugh, 1986).

Acute I.P. naloxone administration caused a significant decrease in sperm motility and viability, while sperm concentration and morphology were not affected. These changes are in according with those of Davies (1983) who proved that acute administration of narcotic drugs caused inhibition of sperm motility and marked reduction in live/dead ratio in rat and mice epididymal seminal fluid.

The minimal degenerative changes in the testes following nalocone administration may be due to the inhibition of microtubule formation in Sertoli cells and in the mitotic divisions of the germ cells (Hess et al., 1990) Sertoli cells contain an abundance of microtubules in their apical cytoplasm, which appear to be involved in the control of spermiation and any disturbance in these microtubules may cause disorganization in the seminiferous epitheilium (Russel et al., 1981).

In the present study, naloxone had no effect on exploratory or sexual behavior in testosterone propionate primed castrated male rats in relation to control group. These data are in line with the results reported by Myers and Baum (1979) and Agmo and Paredes (1988).

In the present study, serum FSH and LH increased significantly after naloxone administration in testosterone propionate primed castrated male rats, while the prolactin level decreased in relation to control group. These results are in line with the results of other investigators who reported the ability of naloxone to stimulate FSH and LH and inhibit Prolactin after steroid administration to castrated rats (Sylvester et al., 1982; Masotto and Negro-Vilar, 1988).

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