Genetic Effects of Interferon and Ribavirin in Albino Mice

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Abstract: The resent study was conducted to investigate the effects of interferon alfa-2a or -2b separately and in combination with ribavirin in albino mice. Adult male mice were divided into nine groups, the 1st and 2nd groups were administered subcutaneously with interferon alfa-2a (0.03 and 0.05 IU/kg/week, respectively), the 3rd and 4th groups were administered subcutaneously with interferon alfa-2b (0.03 and 0.05 IU /kg/week, respectively), the 5th and 6lh groups were administered subcutaneously with interferon alfa-2a (0.03 and 0.05 IU/kg/week, respectively) plus administered orally with ribavirin (0.35 mg/kg/day), the 7th and 8th groups were administered subcutaneously with interferon alfa-2b (0.03and 0.05 IU/kg/week, respectively) plus ribavirin (0.35 mg/kg/day) and the 9th group was considered as a control group. The treated males with interferons for three months were mated with untreated females. On day 18 of gestation, the pregnant females and treated males were sacrificed and examined for sperm abnormalities, chromosomal aberrations and embryonic malformations. It was found that the separately treatments with interferon alfa-2a and -2b increased significantly the sperm abnormalities and chromosomal aberrations as well as the embryo-toxic effect; that was evident by total number of implantations, dead and live embryos; comparing with the control group. While in the combined treatments with interferon alfa-2a or -2b plus ribavirin, there was a highly significant increase in the sperm abnormalities, chromosomal aberrations, number of implantation, dead and live embryos when compared with the control. From this finding, it was shown that interferon alfa-2a and -2b have significant effects in treated mice and their fetuses. Also, the results of this work indicated that the combination between interferon alfa-2a or -2b plus ribavirin had a highly significant increase in sperm abnormalities, chromosomal aberrations and embryo toxic effects in treated mice and their embryos. [Journal of American Science 2010;6(9):43-51]. (ISSN: 1545-1003).

Key words: Interferon alfa-2a and -2b, ribavirin, sperm abnormality, mice, embryos.

1. Introduction

Hepatitis C virus (HCV) is a blood borne viral illness with significant morbidity and mortality. Hepatitis C is caused by an enveloped RNA virus. There are six genetic types of hepatitis C, the most common type in England is genotype I (40%) with the remainder being mainly genotypes 2 and 3. Genotypes 4, 5 and 6 are found predominantly in the Middle East and Egypt, because of the long latent period of infection/disease. The development of serious liver disease and its related mortality deaths from the complications of hepatitis C infection are likely to increase over the coming decades (Zhangf, 2003 and Booth et al., 2001). For clinical excellence, the National Institute endorsed the standard interferon alfa-2a (roferon) and interferon alfa-2b (viraferon). The two interferons are licensed bourse for hepatitis C, either as monotherapy or in combination with ribavirin (Malnick et al., 2000). The exact mechanism of standard interferon action is unknown, it may alter host cell metabolism and inhibit viral replication. Recently, Roch (2002) and Plough (2002) discovered two pegylated interferons; peginterferon alfa-2b (viraferonpeg) and peginterferon alfa-2a (pegasys). In pegylated interferons, the interferon moiety is conjugated with polyethylene glycol; pegylation reduces the clearance of interferon.

Peginterferon alfa-2a (pegasys) is indicated for the treatment of histologically proven chronic hepatitis C in adult patients with elevated transaminases and who are positive for serum (HCV) with liver compensated (Glesby et al, 2005), while peginterferon alfa-2b (viraferonpeg) is indicated for the treatment of adult patients with histologically proven chronic hepatitis C who have elevated transaminases without liver decompensation and they are positive HCV or anti-HCV. Rebetol capsule is an oral formulation of ribavirin; asynthetic nucleoside with broad-spectrum antiviral activity (Schering, 2001). Rebetol (ribavirin) monotherapy is not effective for the treatment of chronic hepatic C, so ribavirin is used in combination with interferon alfa -2a and -2b. Combination therapy with interferon and ribavirin has become the standard course in the treatment of chronic hepatitis infected patients (khalid, et al., 2009).

In the present study, we investigated the effects of interferon alfa-2a and -2b separately and in combinations with ribavirin on the fertility of male

mice and on embryos if female partners mated with male mice treated with hepatitis C drugs.

2. Material and Methods

Test drugs:

Interferon alfa-2a (from Roch) consists of molecule of interferon alfa-2a and interferon alfa-2b (from schering plogh) consists of molecule of interferon alfa-2b.

Interferon alfa-2a and -2b are available as lyophilized powder administered subcutaneously once weekly. Rebetol capsule is an oral formulation of ribavirin administered every day.

Animals and treatments:

Dilutions of different concentrations of tested drug were prepared by dissolving the synthetic powder of interferon alfa-2a and -2b and the oral capsules of ribavirin in distilled water.

Interferon alfa-2a or -2b were administered subcutaneously at two doses of 0.03 and 0.05 I.U/kg/week, while rebetol (ribavirin) was administered orally at a dose of 0.35 mg/kg/day. These doses of interferon alfa-2a, -2b and ribavirin are the recommended doses for human after modified to suit the small weight of albino mice (25 g) according to Pagat and Barnes (1964).

Animals:

Adult males Swiss albino mice each weighing 25g served as experimental animals were divided into nine groups.

Animals of the 1st and 2nd groups were administered subcutaneously by a single dose of interferon alfa- 2a (0.03 and 0.05 IU/kg/week, respectively). Animals of the 3rd and 4th groups were administered subcutaneously by a single dose of alfa-2b (0.03 and 0.05 1U/kg/week, respectively).

Animals of the 5th and 6th groups were administered subcutaneously by a single dose of interferon alfa-2a (0.03 and 0.05 IU/kg/week, respectively) plus 0.35 mg/day of ribavirin. Animals of the 7th and 8th groups were administered subcutaneously by a single dose of interferon alfa-2b (0.03 and 0.05 IU/kg/week, respectively) plus 0.35 mg/day of ribavirin.

Animals of the 9th group served as a control group were administered with the same volume of distilled water. The males were administered with interferons for three months and after that the treated males were housed with untreated females by ratio 1:3, respectively per cage. The day in which the females exhibited a vaginal plug is considered the first day of pregnancy.

The pregnant females were caged individually and on day 18 of gestation, the treated males and the pregnant females were sacrificed by cervical dislocation for studying sperm head abnormalities, developmental and cytogenetic effects of drugs on males and the embryos.

Developmental toxicity:

On day 18 of gestation, the females were sacrificed by decapitation, the uterus contents were evaluated for the number of implantation sites, resorbtion and live fetuses.

Sperm abnormality assay:

The treated males were sacrificed by decapitation, the cauda epididymis was removed and placed in physiological saline. It was minced into pieces with scissors and then left undisturbed for 20 minute for the diffusion of spermatozoa. The spermatozoa were spread on microscopic slides, air-dried, fixed in absolute methanol for 15 minute and stained with 1% aqueous eosin-y. In the next day, one thousand sperms from each animal were examined for the abnormalities in sperm head shapes following the method recommended by Wyrobek and Bruce (1975).

Chromosomes from embryonic cells:

Chromosomal preparations from embryonic cells were prepared according to Romagnano et al. (1985). Embryos were collected from each group and placed in 5 ml T.C.M. 199 media. Two ml of 0.05 colchicine were added for each tube and incubated at 37°C for 90 minutes, then 5 ml of hypotonic solution 0.56% KC1 was added to the pellet, and the cells were incubated at 37°C for 20 minutes. Five ml of fresh fixative (3 methyl alcohol: 1 glacial acetic acid) were added to the cells. After that two or three drops of the cells were dropped on the surface of clean slides, air-dried and stained with 5% Giemsa stain, and examined for chromosomal aberrations. Fifty metaphase spreads were examined for each embryo and the different types of chromosomal abnormalities were scored.

Statistical analysis:

The data of sperm abnormalities and chromosomal aberrations was statistically analysis by ANOVA Test according to Snedecor and Cochran (1980). Duncan's Multiple Range Test was used to compare between means according to Walter and Duncan (1969) at $p<0.05$.

The incidences of resorption, dead and live embryos between treated and control groups were calculated non-parametrically using Wilcoxon's Rank Sum Test (Siegal, 1956).

3. Results

I. Developmental toxicity:

The results presented in Table 1 showed that there were treatments-related effects on the fertility index, number of implantation, number of resorption, number of dead and live fetuses in all treated groups of interferon alfa-2a and -2b separately alone or combined with ribavirin when compared with the control, but these treatment-effects were more strong in combination's treatment groups (with ribavirin) than those in the single treatment group with interferon alfe-2a and -2b alone.

II. Sperm abnormality:

Means ± SD values of sperm abnormality results were showed in Table 2 and Fig. 1. Various forms of sperm heads, i.e., banana-shaped, dwarf, triangular, amorphous, hooked and double-headed were recognized in all the treated groups of interferons alone or plus ribavirin. Analysis of these abnormal sperm shapes showed that overall amorphous types, hooked and dwarf were more prevalent in different groups than banana-like heads, triangular and double-headed. Mono-treatment treatments of male mice for three months with interferon alfa-2a or -2b resulted significant increase in sperm abnormalities in the two tested doses compared with the control. The comparative analysis of sperm head abnormalities in mice treated with the two doses of interferon alfa-2a or -2b in combination groups with a single dose of ribvarin showed highly significantly increase in sperm head abnormalities when compared with control.

III. Chromosomal aberrations in embryonic cells:

The data in Table 3 presented the mean values of the total number of structural and numerical aberrations induced by hepatitis drugs in embryo cells. There were significantly increases in all treated groups; interferon alfa-2a, interferon alfa-2b, interferon alfa-2a plus ribavirin and interferon alfa-2b plus ribavirin; and these increases were dose-dependent when compared with control group.

The result indicated that hepatitis C drugs induced statistically significant increases in male mice and the total chromosome aberrations in the cells of the embryos were at a significant level ($p<0.05$) when compared with control group.

However chromosomal aberration analysis in treated groups with combined treatments (interferon alfa-2a or -2b plus ribavirin) showed highly significant increases in the total number of aberrations when compared with those in treated groups with mono-treatments (interferon alfa-2a or -2b alone).

4. Discusssion

Hepatitis C is a blood borne viral illness with significant morbidity and mortality. Patients with hepatitis C often have no symptoms; the average time of disease progression is 30 years and symptoms of chronic liver disease may only occur later on. The presence of symptoms in order marker of the severity of the disease, deaths from the complications of hepatitis C infection are likely to increase over the next two decades.

Interferon alfa-2a (roferon) and interferon alfa-2b (viraferon) are licensed for the treatment of chronic hepatitis C in adults in combination with or without ribavirin. The safety and efficacy of interferons with or without ribavirin has not yet been documented. Also their mutagenic and fertility effects have not been studied.

The present study was carried out in order to evaluate the mutagenic and embryos toxic effects of interferon alfa-2a and -2b with or without ribavirin in adult male mice and the embryos.

In the present study, the administrating adult male mice with interferon alfa -2a and -2b as a mono-treatment for three months by subcutaneous injection has produced a significant increase in the sperm head abnormalities in the two tested doses when compared with the control. However, negative effects were reported by Lindsay et al. (2001) who found that treatment with interferon alfa-2a did not affect fertility of male rhesus monkeys treated with a therapeutic dose for 5 months. Also, negative effects were reported by Reddy et al. (2001) who studied the genotoxicity of interferon alfa-2b in assays for gene mutations, chromosomal damage *in vitro* and micronucleus assay *in vivo* and they found that the genotoxicity was negative in all assays. Other negative effects were reported by Loginova et al. (1996) who found that peginterferon-2a is neither mutagenic nor clastogenic when tested in the Ames bacterial mutagenicity assay and in the *in vitro* chromosomal aberration assay in human lymphocytes.

However, positive effects were obtained by Matweeva (1982) who reported that treatment with standard interferon alfa-2b and -2a for three months increased the mice sperm head abnormalities and decreased the fertility in treated mice.

Also, positive effects were obtained by Taraka et al. (2004) who observed the interferon-alpha caused chromosomal abnormalities in leukemia patients.

In the present study, our results showed that there was a marked increase in the chromosomal aberrations and in the number of dead and resorbed embryos in treated mice with interferon alfa-2a and -2b.

Table 1: Effects of treatments with hepatitis C drugs on fetal malformations

Treatments groups	Control	Interferon alfa-2a I.U/kg/week		Interferon alfa-2b I.U/kg/week		Interferon alfa-2a I.U/kg/week + Ribavirin (0.35 mg/day)		Interferon alfa-2b I.U/kg/week + Ribavirin (0.35 mg/day)	
		0.03	0.05	0.03	0.05	0.03	0.05	0.03	0.05
Studied females	5	5	5	5	**5**	5	5	5	5
Pregnant females	5	5	4	5	4	3	2	3	2
Fertility frequency %	100%	100%	80%	100%	80%	60%	40%	60%	40%
Total no. of implantations	40	37	34	38	33	27	25	25	23
Total no. of resorption	(5) 12.5%	(5) 13.5%	(6) 17.7%	(5) 13.2%	(6) 18.2%	(6) 22.2%	(8) 32.0%	(5) 20.0%	(7) 30.4%
Total no. of dead fetuses	(3) 7.5%	(3) 8.1%	(3) 8.8%	(3) 7.9%	(3) 9.1%	(4) 14.8%	(4) 16.0%	(3) 12.0%	(4) 17.4%
Total no. of live Fetuses	(32) 80.0%	(29) 78.4%	(25) 73.5%	(30)78.9%	(24)72.7%	(17) 63.0%	(13) 52.0%	(17) 68.0%	(12) 52.2%

Numbers given are the absolute number of fetuses in that group with the indicated abnormality

Table (2): Effects of treatments with hepatitis C drugs on sperm abnormality in male mice

Treatments	Abnormal sperms Mean ± SD	Amorphous Mean ± SD	Banana Mean ± SD	Hooked Mean ± SD	Triangular Mean ± SD	Dwarf Mean ± SD	Double headed Mean ± SD
Control	$70.0^{H} \pm 0.07$	$40.0^{F} \pm 0.30$	$3.0^{G} \pm 0.63$	$10.0^{E} \pm 0.70$	$2.0^{C} \pm 0.05$	$15.0^{D} \pm 0.70$	0.0^{C}
Interferon alfa-2a (0.03 I.U/kg/week)	$77.0^{F} \pm 0.68$	$36.0^{G} \pm 0.51$	$7.0^{F} \pm 0.94$	$15.0^{C} \pm 0.41$	$2.0^{C} \pm 0.37$	$17.0^{C} \pm 0.50$	0.0^{C}
Interferon alfa-2a (0.05 I.U/kg/week)	$79.3^{E} \pm 0.45$	$42.0^{E} \pm 0.84$	$9.0^{E} \pm 0.83$	$10.0^{E} \pm 0.50$	$3.0^{C} \pm 0.31$	$14.0^{DE} \pm 055$	$0.1^{BC} \pm 0.40$
Interferon alfa-2b (0.03 I.U/kg/week)	$74.0^{G} \pm 0.0$	$30.0^{I} \pm 0.21$	$7.0^{F} \pm 0.83$	$21.0^{A} \pm 0.56$	$2.0^{C} \pm 0.31$	$13.0^{EF} \pm 0.83$	0.0^{C}
Interferon alfa-2b (0.05 I.U/kg/week)	$76.0^{F} \pm 0.51$	$33.0^{H} \pm 0.41$	$12.0^{D} \pm 0.30$	$17.0^{B} \pm 0.58$	$3.0^{C} \pm 0.44$	$11.0^{FG} \pm 0.89$	0.0^{C}
Interferon alfa-2a (0.03 I.U/kg/week) + ribavirin (0.35 mg/day)	$150.0^{C} \pm 0.73$	$100.0^{B} \pm 0.58$	$7.0^{F} \pm 0.89$	$13.0^{D} \pm 0.89$	$6.0^{B} \pm 0.70$	$21.0^{A} \pm 0.30$	$1.4^{A} \pm 0.50$
Interferon alfa-2a (0.05 I.U/kg/week) + ribavirin (0.35 mg/day)	$166.0^{A} \pm 0.73$	$112.0^{A} \pm 0.12$	$20.0^{C} \pm 0.58$	$10.0^{E} \pm 0.14$	$3.0^{C} \pm 0.44$	$19.0^{B} \pm 0.14$	$1.4^{AB} \pm 0.50$
Interferon alfa-2b (0.03 I.U/kg/week) + ribavirin (0.35 mg/day)	$140.0^{D} \pm 0.84$	$80.0^{D} \pm 0.78$	$27.0^{A} \pm 0.58$	$16.0^{BC} \pm 0.59$	$5.0^{B} \pm 0.83$	$11.0^{G} \pm 0.89$	$0.4^{BC} \pm 0.24$
Interferon alfa-2b (0.05 I.U/kg/week) + ribavirin (0.35 mg/day)	$157.0^{B} \pm 0.18$	$88.0^{C} \pm 0.38$	$23.0^{B} \pm 0.30$	$20.0^{A} \pm 0.30$	$7.0^{B} \pm 0.63$	$17.0^{C} \pm 0.78$	$1.2^{AB} \pm 0.58$

Different letters means significant at $p<0.05$

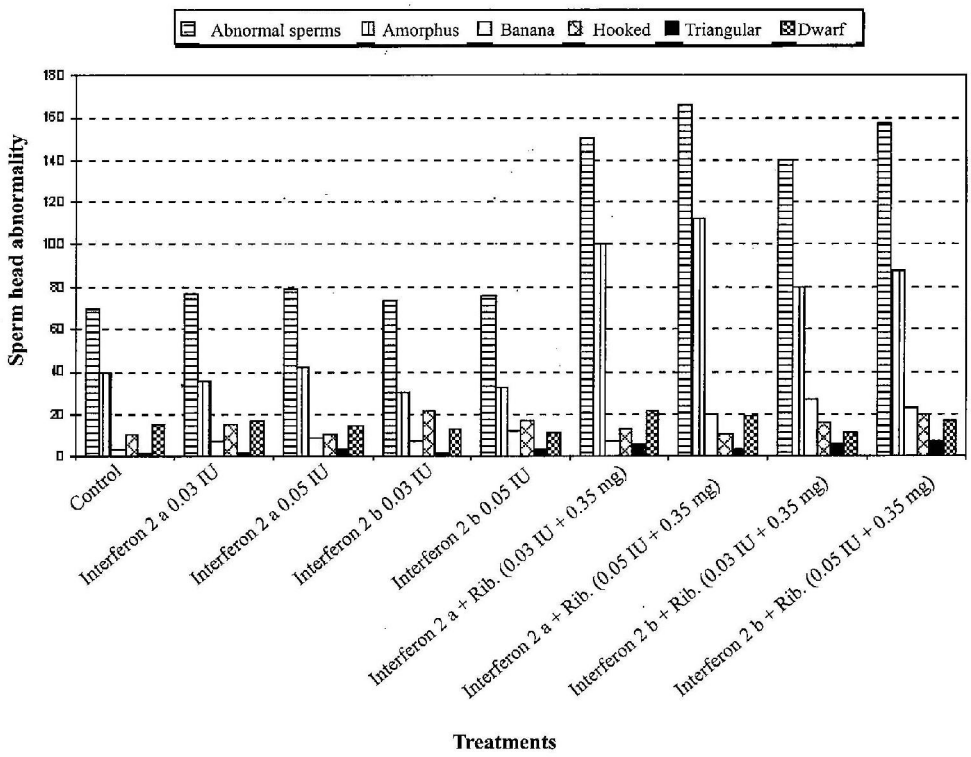

Fig. (1) Incidence of sperm head abnormality.

Table (3): Effect of treatments with hepatitis C drugs on the chromosomal aberrations of the embryonic cells

Treatment	Structural aberrations								Numerical aberrations		
	Chromatid gaps Mean ± SD	Chromosomal gaps Mean ± SD	Chromatid breaks Mean ± SD	Deletions Mean ± SD	Fragments Mean ± SD	Endomitosis Mean ± SD	Centromeric attenuations Mean ± SD	Total structural aberrations Mean ± SD	Aneuploidy Mean ± SD	Polyploidy Mean ± SD	Total numerical aberrations Mean ± SD
Control	$2.8^{D} \pm 0.73$	$4.5^{BC} \pm 0.89$	$1.5^{DE} \pm 0.32$	$3.5^{D} \pm 0.55$	$1.6^{C} \pm 0.4$	$1.4^{E} \pm 0.24$	$1.4^{D} \pm 0.24$	$16.7^{G} \pm 0.8$	$4.4^{D} \pm 0.51$	$0.6^{D} \pm 0.2$	$5.0^{G} \pm 0.31$
Interferon alfa-2a (0.03 I.U/kg/week)	$6.6^{BC} \pm 0.81$	$2.2^{BC} \pm 0.55$	$1.5^{E} \pm 0.32$	$3.0^{D} \pm 0.55$	$2.4^{BC} \pm 0.51$	$3.2^{D} \pm 0.66$	$2.6^{CD} \pm 0.81$	$21.5^{F} \pm 0.3$	$5.0^{D} \pm 0.63$	$2.4^{C} \pm 0.60$	$7.4^{F} \pm 0.24$
Interferon alfa-2a (0.05 I.U/kg/week)	$5.2^{AB} \pm 0.3$	$3.2^{AB} \pm 0.66$	$1.8^{DE} \pm 0.37$	$3.0^{D} \pm 0.63$	$3.0^{AB} \pm 0.63$	$3.4^{CD} \pm 0.51$	$3.2^{BC} \pm 0.58$	$22.8^{D} \pm 0.92$	$7.8^{C} \pm 0.09$	$2.0^{C} \pm 0.44$	$9.8^{DE} \pm 0.24$
Interferon alfa-2b (0.03 I.U/kg/week)	$4.4^{D} \pm 0.51$	$2.0^{C} \pm 0.22$	$3.2^{D} \pm 0.24$	$2.6^{CD} \pm 0.51$	$2.6^{BC} \pm 0.51$	$2.8^{D} \pm 0.66$	$2.8^{BCD} \pm 0.66$	$20.4^{F} \pm 0.45$	$7.6^{C} \pm 0.67$	$1.6^{CD} \pm 0.24$	$9.2^{EF} \pm 0.44$
Interferon alfa-2b (0.05 I.U/kg/week)	$6.4^{BC} \pm 0.92$	$2.6^{BC} \pm 0.24$	$1.6^{DE} \pm 0.24$	$2.6^{BCD} \pm 0.51$	$2.6^{BC} \pm 0.51$	$2.8^{D} \pm 0.58$	$2.6^{BCD} \pm 0.51$	$21.2^{E} \pm 0.9$	$8.2^{C} \pm 0.73$	$2.4^{C} \pm 0.40$	$10.6^{D} \pm 0.51$
Interferon alfa-2a (0.03 I.U/kg/week) + ribavirin (0.35 mg/day)	$6.2^{BC} \pm 0.48$	$3.2^{BC} \pm 0.48$	$6.6^{B} \pm 0.51$	$4.8^{ABC} \pm 0.48$	$4.8^{A} \pm 0.48$	$5.0^{AB} \pm 0.94$	$4.6^{A} \pm 0.40$	$35.2^{A} \pm 0.7$	$14.2^{B} \pm 0.28$	$3.6^{B} \pm 0.51$	$17.8^{C} \pm 0.15$
Interferon alfa-2a (0.05 I.U/kg/week) + ribavirin (0.35 mg/day)	$7.8^{A} \pm 0.37$	$4.4^{A} \pm 0.60$	$8.4^{A} \pm 0.60$	$6.2^{A} \pm 0.48$	$4.6^{ABC} \pm 0.40$	$4.8^{BCD} \pm 0.37$	$4.2^{A} \pm 0.51$	$40.4^{AB} \pm 0.11$	$15.6^{A} \pm 0.30$	$8.2^{A} \pm 1.37$	$23.8^{AB} \pm 0.93$
Interferon alfa-2b (0.03 I.U/kg/week) + ribavirin (0.35 mg/day)	$6.2^{CD} \pm 0.37$	$5.0^{A} \pm 0.44$	$4.0^{C} \pm 0.44$	$5.6^{AB} \pm 0.60$	$5.8^{A} \pm 0.37$	$5.6^{BC} \pm 0.51$	$1.7^{B} \pm 0.58$	$33.9^{C} \pm 0.06$	$15.8^{B} \pm 0.37$	$7.6^{A} \pm 0.74$	$23.4^{CB} \pm 0.51$
Interferon alfa-2b (0.05 I.U/kg/week) + ribavirin (0.35 mg/day)	$8.4^{AB} \pm 0.40$	$6.0^{A} \pm 0.44$	$7.4^{AB} \pm 0.51$	$4.2^{BCD} \pm 0.48$	$5.4^{A} \pm 0.51$	$6.2^{A} \pm 0.37$	$5.4^{A} \pm 0.60$	$43.0^{A} \pm 0.63$	$18.0^{A} \pm 0.94$	$8.4^{A} \pm 0.81$	$26.4^{A} \pm 0.51$

Different letters means significant at $p<0.05$

Our results are in agreement with Vogel (2003) and Wang et al. (2002) who observed that treatment with interferon alfa-2b produced significant abortifacient effects in Rhesus monkeys at doses equal to the recommended human dose.

Also, positive results were obtained by Bolzan et al. (2004) who observed that treatment with interferon alfa 2a resulted in a statistically significant increase of the chromosome aberrations and sister chromatids in hamster cells.

However, negative results were obtained by Sheliia et al. (1984) who demonstrated that administrations of standard interferons alfa-2a and -2b did not produce any teratogenic effects in the white mice offspring.

Also, in the present study the male mice treated with either interferon alfa-2a or -2b with ribavirin for three months produced a highly significant increase in the sperm head abnormalities of treated males and also produce embryocidal effects in the resulted embryos.

These results are in agreement with Sulkowski et al. (2000) who found that male mice administrated with doses of 15 to 150 mg/kg/day of interferons plus ribavirin for 3 months displayed abnormalities in sperms.

Also, positive results were obtained by Leyssen et al. (2003) who observed that the administration of interferon alfa-2b in combination with ribavirin has a reproductive toxicity effects in female and male mice.

In addition, Manns et al. (2001) observed that ribavirin alone caused a significant embryocidal and/or teratogenic effects in all animal species at doses below the recommended human dose. However negative studies by Glue et al., (2000) who demonstrated that the standard interferon alfa-2b is not mutagenic, but ribavirin increased the incidences of mutation and cell transformation multiple genotoxicity assays in the long-term studies in the mouse and rat (18-24 months old) with doses of 20.75 mg/kg/day. Fried et al. (2001) and Vogel (2003) demonstrated that there is a relationship between chronic ribavirin exposure and increase in the incidences of vascular lesions in mice. However in rats, retinal degeneration occurred.

Also, positive results were observed by Tedjaratis et al. (2002) and Craxi and Licata (2003) who found that ribavirin administration in a dose equal to the recommended dose in rats and rabbits caused increase in the incidences of malformations of the skull, palate, eye two, limbs and skeleton of the fetuses and this increase was dose-dependent.

5. Conclusion

In conclusion our results indicated that interferon alfa-2a or -2b had significant mutagenic and cytotoxic effects on both male fertility and the embryos and also these interferons have a slight increase in the total number of dead and resorped embryos when compared with control.

Also in the present study, we found that the treatment of mice with interferon alfa-2a and -2b plus ribavirin caused a highly significant increase in the sperm head abnormalities, in the chromosomal aberrations of fetal cells and embryos toxic effect at all tested doses of both drugs. This may be as a result of the highly teratogenic and mutagenic effects of ribavirin alone or in combination with interferon, which it may accumulate in intercellular components and affect somatic cells a well as germ cells thus causing sperm abnormalities which cause teratogenic and mutagenic effects on the fetal cells.

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