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To Study Genesis of Testis Tissue in Male Infants of Rat That Were Born from Mothers Who Were Cared by Haloperidol Drug

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Abstract: *This paper is to evaluate the effect of Haloperidol drug in changes of tests tissue and spermatogenesis process. 40 pregnant female rat of vistar race were studied empirically in 5 groups with 8 members and 8 male infants out of each group were tested randomly. There was not medicine care on control group. Control group recived 1 cc distilled water as drug's solvent. Empirical group received Haloperidol drug 12/5, 25, 50 mg/kg every day for 28 days orally. Hemat imeteric was performed when time period was over and after drug prescription. Hematosmples were gathered in order to measure serum concentration of hormones: LH, FSH and testosterone by radio immunocy method. According to results, 25, 50 mg/kg Haloperidol consumption has led to lose weight and testosterone reduction while, has increased LH, FSH concentration. To evaluate on testis tissue showed some obvious reduction in cell sequence of spermatogenesis by dose of 25, 50 mg/kg.weight – average of testes had not meaningful difference between control group and empirical group who had received drug. Eating 50 mg/kg Haloperidol reduced erume concentration of testosterone hormone and attenuate production process of spermatogenesis cells, whereas it increases serum concentration of LH, FSH hormones and this show remedy activity by hippofiz to exudte gonadoteropins. Hence, if this drug is used in long time, generation efficiency will bedecreased.*
Key words: *haloperidol, testosterone, gonadotropin, testicles, rat*

INTRODUCTION

Depression is a psychiatric desease that leads to feel continuous worry and grief and to loss interests, sometimes, many people feel to fall in worry, depression and grief, to feel sorrow and depression is normal reaction of body to life problems and to loss things and whom are related to them. But if feeling to fall insever grief, hopelessness, helplessness and valuelessness is long some days and some weeks, depression disease is appeared.

A kind of depression which is under this scale is named melancholia or depression, but reaction depression is often a sever reaction to tension- earing events that is out of control by person. Sometimes, pharmaceutical treatment leads to depression. Depression is one of the most important mood disordersin psycho herapy. Reason of depression is not known, but there may be an action interference between psychical mechanism and bio-chemical mechanism or may act separately each on.

In other words, depression is a physicl and mental disease. Many of people not only have mental symptoms but also they have physicalsymptoms of this disease, but its nature is different from person to other. Various symptoms of depression are seen more and less in every patient. There may be no syptom of depression in some persons, but their behavior begins to change abnormally. Haloperidol is used to treatment of

schizophrenia, quarrelling, harshness, psychical disorders, mania, and it decline severity of some psychical diseases like madness and other psychical diseases that mania is seen in them and it decline wisdom- wane and imaginations.

Haloperidol is used to aid and to treat in short time, psychical movemental excitation, to control ticks or involuntary movements and to control chronic sichotik disorders in patient's who need to long term treatment and to treat irritability and disturbance and it is also effective in treatment of sever worry in short term. Although, it cannot cure disease completely, but it decreases worry, it means that Haloperidol don't treat initial disease but, rather, it moderate disturbance- bearing symptoms. Haloperidol is used to treat delirium, movemental ticks and unwanted sounds in Tourette syndrome. It is also applied to treat children who have sever abnormality and problems in theirbehaviours and is used to short term treatment for hyperactive children with violent movement actions and it's used to treat resistant hiccup.

Nervous abnormalities including cerebral injuries, nervous infection , poly-kind sclerosis, Parkinson, worry, depression, catalepsy, … and according to report by global health organization, the most common nervous disorders is worry and depression.

According to studies by sanberg andco-workers in 1988, Parkinson disease (PD) is one of the diseases in nervous transmutation which is seen is form of selective losing of Dopamin neuron (DA) incondence part of black body. Katalepcy means before recovering normal situation by animal. Haloperidol is and anti- psychosis drug that is prescribed mainly to treat schizopherny and other abnormalities and can be used to form catalepsy in animals. Genitals (system) in adult male rat is testis, meanwhile, the gonad that is affected by Y chromosome is trapped in some complex events that lead to genesis male genitals. Testosterone production by lydic cells results to mature velphin canal in form of male genital canal epididom, vazedpharan, spermatocyst, and simultaneously, production of controlling materialof mulerin canal through testis leading to to degenerate mulrin canals, these canals (Mulrin) has role in forming uterus and phalluptube.

Rudketaz Alfa-5 enzyme turn testosterone to dehydrotestosterone and this leads to masculation external system, for example: scrotum and penis. Testes sizes are different if rats and are based on body weight. Testes come down in a non- adult rat that has 4- 6 weeks' age and testes can move up inside stomach fossa across rat's life, this results to open groin canal. Inside each testis, there are many sperm tubes, sperm is produced in these sperm tubes.

Humen have two testes. Each testis has 4-5 c-m long, 2-5 cm wide and 3 cm wide on the average. Left testis is 1cm farther down right testes. The testes have 2 key function:

1) To prepare suit space to spermatogenesis process and

2) To exude testosterone that control body changes related to generational acts.

Testes are 2 and are placed in scrotum and out of stomach fossa, where, heat is 1-2 c lower than body heat, since, in mammals, spermatogenes is sensitive to heat up 37 c and they may be dead, so scrotum has many glands in its wall that produce sweat and maintain testes in desired heat (18,4).

Sperm is formed in spermatogenesis tubes of male rat in 12 days and 48 days take to perfect spermatogenesis in genitals (system) of male rat, so there is took 60 days and 35 days since beginning spermatogony cells activity to forming active sperm in human and rat respectively. (11)

Research method:

Tested animals were infants of male rat from vistar race with 80 c approximate weight and 22 days age, their mothers were took from nurse center of animals in Azad university of Kazeroon and they received Haloperidol drug in gestation. After birth, male infants were selected to main in that center.

Room temperature was constant 2 t 22 cat day and night. They spend 12- hours in the dark and 12 hours in the light at optical period. Uniform and indirect Radiation was diffused from lab windows. And they were feed within period of experiment without any limitation.

Experiment had been done from first days of Mordad 2014 till middle days of Bahman in 2014. At first, female rat was weighted in order to grouping, the rat was put in restricting system, because they cannot move around, when they were weighted. And then, net weight of them was determined by digital balance with 0/001 g precision.

After that, female rats were grouped in 5 groups including 8 members, each one of them had 20 ± 220 g weight approximately in poly- carbonat cages and as control group, witness group and empirical group, after gestation and, male infants were grouped.

In 5 group including 8 members and their weight were 20 ± 80 g approximately. Control group did not receive any drug or non-drug nurcing.

Witness group were receiving distilled water 1c-c orally as drug solvent every day. Empirical group 1,2,3 were receiving Haloperidol drug one time every day on value 12/5, 25 , 50 mg-k-w respectively and this drug has been feed to them through feeder syring for 28 days. After 28 days, infants were anesthetized by ether and blood was sampled from their heart. About 2 c.c blood of infants was sampled in strilled test tube that hadn't anti-coagulant. Sampled blood was centrifuged with 5000 circulations in minute rate (velocity) for 15 minute, in order to separate serum from clot. Then, the samples were used to measure serum concentration of LH, FSH, testosterone hormones. Radio immunocy method was applied to measure hormones.

After cleavaging animals stomach, both testes were come out in all of 5 group and were weighted and after preparing tissue's sections and staining with hematoxlin- Auzin dye by calibrated slide which is special to measurement (Graticol) every changes in numbers of chain, curtail and interrelated cells of spermatogenes at control group and empirical group were studied in tissues by optical microscop.

Data [gathered data] were analyzed by spss software and statictic tests of variance analyze and monitoring tests of tike.

Results:

According to results, there was meaningful decrease in serume's concentration of festosterone hormone in day 29, in the group that receiving 25, 50 m-g-drug on kg of body weight comparing control group ($p \leq 0/005$) there was meaningful increase in serume's concentration of LH and FSH hormones in 29 the day, in the group that receiving 25, 50 mg drug on kg of body weight comparing control group ($p \leq 0/005$) (table 1) .

Studying on testis biopsy showd, in the group with 25, 50 m.g on kg, initial spermatogony cells and spermatocit cells were decreased meaningfully and in empirical group with 12/5 , 25, 50 mg on kg there was meaningful decrease in lydig and spermatid cells compare to control group and witness group ($p \leq 0/005$). Sertoly cells didn't differ among empirical group and control group and witness group after 28 days. ($p \leq$

0/005). Control group and witness group also didn't no diffrences and changes in above parameters (p ≤ 0/005) (table 2 and pictures 1-3) .

Table 1:avarage and reference deviation in plasma concentration of testosterone, LH and FSH hormones are compared after prescribing Haloperidol orally in studied groups:

LH Global unit on liter	FSH Global unit on liter	Testosterone (Nanogram on desilite)	Group
7/68 ±0/18	4/08 ± 0/26	5/02 ± 0/08	Control
7/65± 0/30	3/98 ± 0/28	4/90 ± 0/22	(solvent value)
8/44 ± 0/33	4/66± 0/14	4/44 ± 0/16	Empirical1
9/38 ±0/30 *	5/56 ± 0/23 *	3/84 ± 0/11 *	Empirical 2
11/20 ± 0/39 *	6/30 ± 0/16 *	3/22 ± 0/11 *	Empirical 3

*meaningful difference to control group and witness group

Table 2: average and reference deviation in sperm cells, sortie cells and lydig cells are in compared same tube, after Haloperidol drug prescription in from of orally among studied groups:

Numbers of lydig cells	Numbers of sertdi cells	Numbers of spermatid cells	Numbers of initial spermatocit cells	Numbers of spermatogony cells	Group
22±0.63	18.12±0.72	128.5±0.87	76.12±0.95	53±1.04	Control
20.50±0.57	18.12±0.99	127/8±0.75	75.25±1.06	52.12±0.99	Witness
18.25±0.49*	18.98±0.92	122.5±1.27*	72.50±1.05	49.25±3.69	Empirical 1
12.38±0.56*	19.38±0.50	112.1±0.97*	65.25±0.80*	42.38±0.65*	Empirical 2
10.75±0.75*	17.62±0.94	107/8±0.88*	59.75±0.59*	37±0.70*	Empirical 3

*meaningful difference to control and witness groups (p ≤ 0/005)

Picture 1: photomicrograph on spermatogenesis- duct tube in control group , magnitude : 40 x

Picture 2 : photomicrograph on spermatogenesis- duct tube in witness group, magnitude : 40 x

Picture 3 : photomicrograph on spermatogenesis tube (duct) in empirical group 1, with 12/5 m-g doze of drug on kg- magnitude: 40x

Picture 4: photomicrograph on spermatogenesis duct (tube) in empirical group 2, with 25 m.m doze of drug on kg. magnitude: 40 x

Picture 5 : photomicrograph on spermatogenesis duct (tube) in empirical group 3, with 50 mg doze of drug on kg, magnitude: 40x

Discussion:

The main objective of this paper is to evaluate Haloperidol drug effect on sex hormones (Gonadotrop and testosterone hormones) and changes in testes tissue of male rat in fants.

According to results of this study, prescription of Haloperidol drug orally, decrease testosterone and increase FSH and LH meaningfully. Histology on testis tissue revealed, excellent decrease in spermatogenesis cellular chain (sequence). Plasmic concentration average of testosterone was more decreased in empirical groups (that received drug) compare to control and witness group. This difference is mainly seen in the group that received maximum doze of drug. ($p \leq 0/005$) Inhibitor drugs in reabsorption of servtion, decrease production of testosterone hormone and this lead to derange sex behavior (34,35).

The results show, prolactin stimulate testosterone secretion by fixing number of LH acceptors in lydig cells area (24). To secret this hormone increasingly, will be lead to decrease sensivity of these acceptors toward LH secretion and control on LH acceptors will be decreased (42).

According to results, prolactin slow steroidogenesis and testosterone production in testis by inhibitory affecting on secondary messengers (signals) (15).

According to negative feedback control, If testosterone is secreted bellow standard, hipotalamus will secret high GnRH and consequently, LH testosterone will be secreted highly from anterior hypophysis and testis respectively.

But decreasing testosterone may have negative feedback not only on hypotalamous but also on anterior hypophysis directly and some believed this hypophysis feedback increase LH secretion specially, and this effect on lydic cells and testosterone will be increased.

In this search, we conclude that Haloperidol drug affection gonadotrop cells of hypophysis in brain directly, and LH hormone will be increased. (30, 31,33) Haloperidol drug effect on dopamine metabolism indirectly, as testosterone will be decreased, on one hand, testosterone is an inhibitor factor to monoamino oxidaz enzyme, this enzyme has role in dopamine catabolism and decrease dopamine level in synapse part (17, 15).

Therefore, testosterone increase dopamine level indirectly by decreasing this enzyme, here this inhibitory effect on monoamino oxidaze is decreased by testosterone decreasing and dopamine concentration is also decreased (32).

According to studies, based on negative feedback, if testosterone is secreted below standard, hypothalamus will secret GnRH highly, consequently, FSH will be secreted highly from anterior hypophysis and on one hand, based on negative feedback if testosterone is secreted lowly, anterior hypophysis gland will be affected and FSH will be secreted highly, eventually it effect on sterol cells and spermatogenesis process will be increased (10).

This study showed, Haloperidol may decrease spermatogenesis process and sperm production, it increases FSH secretion. Decreasing in testosterone concentration and it's production site in lydig cells, spermatogenesis process will be problem and sperm condensation will be decreased, so it is told that high value of Haloperidol may decrease sperm condensation in Lumen space (23, 7).

According to results of this study average numbers of primary spermatogony and spermatocit cells was decrease mainly in ($p < 0/005$). In empirical group with 25, 50 mg/kg doze and also spermatid was decreased in empirical group with 50, 25, 12/5 mg/kg doze compare to control group and witness group. Spermatogony cells are set on membrance of spermatogen tubes' support chainly and during divisions; spermatogony cells are not separated completely and boned to eachother by cytoplasmic bridges. Source of primary spermatocit cells is spermatogony cells (8).

Spermatid cells are the smallest cells in spermatogenic class and have 7-8 micron diameters (22). Testosterone hormone cause to mature circle spermatids easily and their transformation to long spermatids is done easily too. High concentration of testosterone has role in division of spermatocytes and spermatogony. In this study, concentration of testosterone hormone was decreased meaningfully in empirical group with consumption 25,50 doze of drug , probably, Haloperidol drug consumption, may decrease testosterone concentration and after that cellular division will be decreased. FSH hormone is necessary to start spermatogenesis or on the other hand to start miosis division on spermatogony cells (36-1)

Thus, abnormality in FSH hormone level, lead to abnormality in spermatogenesis process. (13-1) based above, Haloperidol drug decrease LH, FSH, and testosterone levels and decrease also spermatogony cells, primary spermatocit and spermatid. There has been observed meaningful decrease in intermediate cells (lydig)in empirical group with 50 , 25 mg/kg doze of drug compare to average numbers of these cells in control and witness group ($p <0/005$) . Lydic cells are set in interspace of spermatogenesis tubs and will synthesize testosterone in purity. Nonseparated cells in intermediate tissue are affected by LH hormone that is secreted

from anterior hypophysis and turn to intermediate cells. Intermediate cells have ability to synthesize testosterone out of cholesterol (27) since, testosterone level has been decreased mainly in this search, and testosterone is secreted from intermediate cells, hence, it's evident to decrease numbers of intermediate cells across testosterone decreasing.

Condensation of sperms was decreased in empirical group respectively and min level of sperm was seen in max group, but its noteworthy that sperms condensation is normal in control group and witness group. First reason is related to testosterone decreasing which is necessary to spermatogenesis and this lead to decrease sperms condensation. Second reason is related to numbers of cells that making spermatogony, primary spermatocit and spermatid, that eventually decrease sperms 'condensation (23, 7).

One another change in this research is to decrease spermatogenesis tubes. In empirical group 2, there was increased gap of spermatogenesis tubes and their condensation was decreased but in empirical group 3, spermatogenesis tubes not only were decreased but also, they were degraded and perturbed.

Another change is related to empty spaces in spermatogenic cells and some picnoz core is inside spermatogenesis tubes. These show inhibitory effects of drug on spermatogenesis process. Abnormality in testes tissue is related to effect of drug on cellular bonds.

As Haloperidol drug increase LH, FSH hormones and decrease testosterone, and their effects on different stages of spermatogensesis, there is negative effect on testes tissue. As beginng and maintaining spermatogenesis requires to normal value of LH and FSH pre and post sex purity as separating, spermatocytes of stage and spermatids of stage 7 are affected by testosterone directly (20)

Conclusion wholly, Haloperidol drug increase plasmic concentration of LH, FSH hormones and decrease serume concentration of testosterone hormone and decrease condensations of spermatogony, primary spermatocit , spermatid and lydig cells but does not decrease sertoly cells. As seemingly, decreasing of androjens like testosterone can effect on spermatogenesis process and decrease sperm production in seminiphoz tubes.

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