

TERATOGENIC EFFECT OF CISPLATIN-TREATMENT IN MICE FETUS

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ABSTRACT

Cisplatin is a drug with widespread use in the treatment of malignant tumors such as ovarian, testis, bladder cancers. The use of the drug is limited because of many side effects on different organs. Considering the above mentioned, the necessity of studying of destructive effects of the drug during pregnancy from maternal to fetal is raised. High dose of Cisplatin (10 mg/kg) was IP injected to the experimental group on sixth and twelfth days of pregnancy Control group received normal saline as IP injection on sixth and twelfth days of pregnancy. Data expressed as Mean±SEM T-test as well as SPSS software were used to data analysis and P<0.05 was considered. Analysis results of the obtained data related to fetal weight, CRL, Skull length and skull width measurement show that the experimental group CRL measure is lower meaningfully lower compared with the control group (P<0.05). It is suggested that cancerous patients doesn't use Cisplatin under any circumstance during pregnancy and lactating; so, if the use of the drug is necessary it is suggested that a complex of different anti oxidants such as arachidonic acid, ascorbic acid, super oxide dismutase, and antioxidants especially vitamins E and C be used.

Keywords: *Cisplatin, Mice Fetus, Teratogen*

INTRODUCTION

Birth defects, congenital disorders and anomalies are similar terms which are used to define structural, behavioral, functional and metabolic congenital disorders. The science studying these kinds of disorders is called Teratology. Congenital defects can be due to hereditary and environmental factors. One of the important environmental factors is the use of some drugs during pregnancy (Rahmani *et al.*, 2006). Cancer is occurred genetically or environmentally. Generally, most of cytotoxic drugs which are used due to chemotherapy affect directly the cancerous cells especially those cancers with high rate of cell proliferation. Some of the drugs cause basic changes in cells such that prevent their growth which is called "cell death planning" (Seaman *et al.*, 2003). Teratogenic effects of the drugs on fetus and causing fetal malformation (Longo *et al.*, 2011; Satoh *et al.*, 2003) is raised but our information about varieties of disorders in newborn mice isn't sufficient.

In a study conducted by McCuire *et al.*, (1996) it has been mentioned that ovarian cancer is the most common factor for women mortality. The congenital risk rate for ovarian cancer during life is about 1.5% and the mortality probability for women because of cancer in their life time is about 1% (McCuire *et al.* 1996). Cisplatin is a drug with widespread use in the treatment of malignant tumors such as ovarian, testis, bladder cancers. The use of the drug is limited because of many side effects on different organs like kidneys (Zahiri *et al.*, 2003). Due to widespread use of chemical drugs to treat cancers, their toxic effects on other cells have been proven. Cisplatin is one of the alkylating agents as well as one of the drugs used in chemotherapy. It is also known as a selective drug in treating the genital malignant tumors especially ovarian carcinomas. Considering the above mentioned, the necessity of studying of destructive effects of the drug during pregnancy from maternal to fetal is raised.

MATERIALS AND METHODS

Due to effects of Cisplatin on maternal mouse, the study was conducted as an experimental. The study was conducted on two groups: 1. Interventional (experimental) group, 2. Control group. 10 adult male and 30 adult female mice with average weight of 25 g were selected for mating and pregnancy. The mice were

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kept in a laboratorial animal cage at $25\pm 2^{\circ}\text{C}$ for 24 hours. Zero day of pregnancy was considered by observing the copulatory plugs formation. The pregnant mice were divided randomly into two, experimental and control, groups.

High dose of Cisplatin (10 mg/kg) (Seaman *et al.*, 2003) was IP injected to the experimental group on sixth and twelfth days of pregnancy (Hooser *et al.*, 2000). Control group received normal saline as IP injection on sixth and twelfth days of pregnancy. Near the end of pregnancy (day 19), the pregnant mice were anesthetized using ether. They were autopsied to remove fetus from uterine (Soysal *et al.*, 2011). The collected fetuses were evaluated in terms of anterior and posterior limbs' buds, tail, eyes, and skin defects, and the cases which were not normal for the fetus age, were identified and considered as abnormality (Hooser *et al.*, 2000; Longo *et al.*, 2011). The fetuses' weight was measured by digital scale with an accuracy of 0.01 gr and their length was measured based on their CRL using caliper with an accuracy of 0.1 mm.

- Fetal weights mean which is considered as a quantity variable.
- Fetal lengths mean which is considered as a quantity variable.
- Malformation rate which is considered as a quality variable.
- Fetal lethality rate which is considered as a quantity variable.

Statistical Analysis

The mean and standard deviation of newborns number were obtained. Offsprings length and weight and different abnormalities abundance in different groups were expressed as $\text{Mean}\pm\text{SEM}$ T-test as well as SPSS software were used to data analysis and $P<0.05$ was considered.

RESULTS AND DISCUSSION

Results

The Parameters of Fetal Growth

The collected samples were analyzed and fixed. In this study, quantitative growth parameters were considered. Parameters of weight, length of infant length, length and width of the skull in the control group, the intervention group were measured and recorded. Results are expressed as mean \pm standard deviation ($\text{SEM} \pm \text{Mean}$) was presented. The different between the different groups to test T-Test evaluated and analyzed using SPSS soft ware and the level of significance tests $P < 0/05$ was considered.

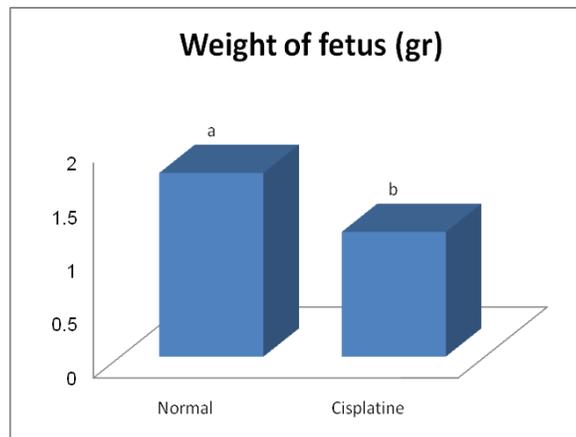


Diagram 1: Comparison of Mean \pm SEM of *weight of fetus*, followed by administration of Cisplatin, in Fetal Mice. The different letters show a meaningful difference of mean among groups ($P<0/01$)

Analysis results of the obtained data related to CRL measurement show that the experimental group CRL measure is lower meaningfully lower compared with the control group ($P<0.05$).

Analysis results of the obtained data related to fetal weight show that the experimental group weight is lower meaningfully lower compared with the control group ($P<0.05$).

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Analysis results of the obtained data related to skull length show that the experimental group skull length was meaningfully lower compared with the control group ($P<0.05$).

Analysis results of the obtained data related to skull width show that the experimental group skull width was meaningfully lower compared with the control group ($P<0.05$).

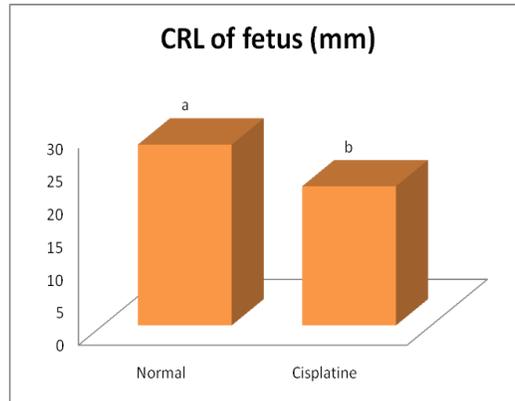


Diagram 2: Comparison of Mean±SEM of *CRL of fetus*, followed by administration of Cisplatin, in Fetal Mice. The different letters show a meaningful difference of mean among groups ($P<0/01$)

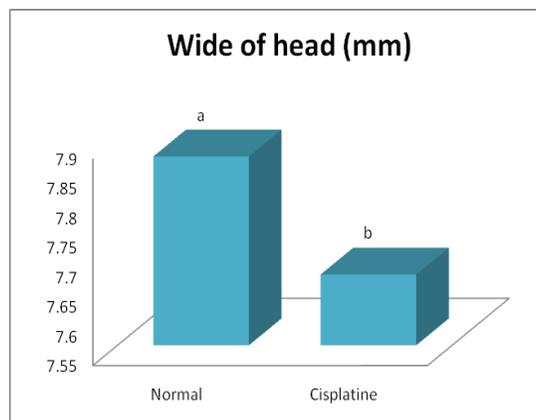


Diagram 3: Comparison of Mean±SEM of *width of head*, followed by administration of Cisplatin, in Fetal Mice. The different letters show a meaningful difference of mean among groups ($P<0/01$)

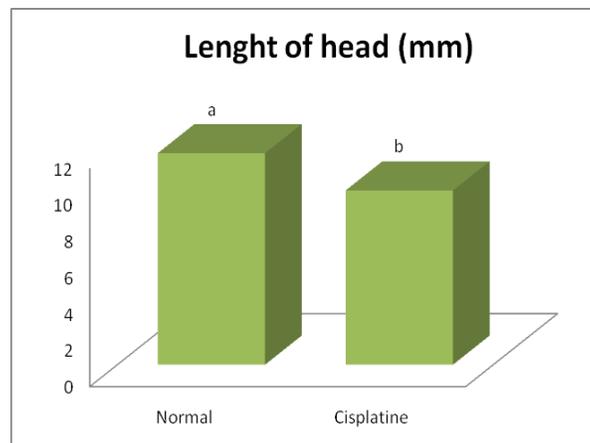


Diagram 4: Comparison of Mean±SEM of *Length of Head*, followed by administration of Cisplatin, in Fetal Mice. The different letters show a meaningful difference of mean among groups ($P<0/01$)

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Analysis results of the obtained data related to CRL measurement show that the experimental group CRL measure is lower meaningfully lower compared with the control group ($P<0.05$).

Analysis results of the obtained data related to fetal weight show that the experimental group weight is lower meaningfully lower compared with the control group ($P<0.05$).

Analysis results of the obtained data related to skull length show that the experimental group skull length was meaningfully lower compared with the control group ($P<0.05$).

Analysis results of the obtained data related to skull width show that the experimental group skull width was meaningfully lower compared with the control group ($P<0.05$).

The results of dead fetus counting demonstrated that the number of experimental group dead fetus was meaningfully high compared with the control group ($P<0.001$).

External Abnormalities

The obtained results from external abnormalities suggested that there were variety of abnormalities in experimental group such as: abnormal rotation of anterior and posterior limbs, wrinkled skin, local hemorrhage, tail and eyes diorderts, and lack of brain, that have been presented in table 1.

Table 1: Results the prevalence of fetal abnormalities in both the intervention and control groups

Groups variables	Control (n=40)	Intervention (n=40)
Abortion	0	3/3 * ± 0/2
Rotation of the hands and feet	1/7±0/5	9/6 * ± 1/2
Abnormal rotation of the body	0	2/4±0/6
Wrinkled skin and topical bleeding	1/8±0/3	5/5* ± 0/3
Failure tail	0	4/4*±0/2
Eye disease	0	1/6±0/7

* Significant differences between the control and intervention group ($P<0/05$)



Figure 1: Observation of aborted fetuses in Cisplatin group



Figure 2: Observation of mice fetus from experimental group (decreased growth, hand and foot abnormal rotation, and tail disorder)

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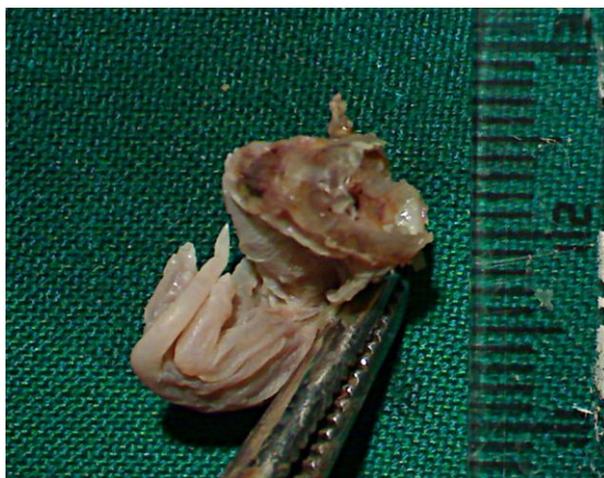


Figure 3: Observation of dead fetus with encephalic abnormality from experimental group

Conclusion

Since ovarian and cervix cancers are the most common causes of mortality among women, anti-cancer drugs, like Cisplatin, usage are increasing such that recently the concern about pregnant women's reproductive abnormalities has been increased.

Therefore, the present study aiming at the evaluation of the maternal use of Cisplatin effects during pregnancy on newborn maturation.

The obtained results suggest that the use of Cisplatin has irreversible teratogenic effects on fetus which are accompanied by the clinical symptoms. Our study is consistent with the studies on teratogenic drugs and their crossing from placental blood barrier and imposing birth disorders (Dudek *et al.*, 2001) It could be said that toxic effects of the drug have irreversible effects on fetuses like other teratogenic drugs (Naghizadeh *et al.*, 2007).

The obtained results from data analysis related to CRL measurement suggest that treatment with Cisplatin has a regressive effect on growth parameters (fetus length, weight, and width) which is considered as a destructive effect on fetus development. In study conducted by Hooser *et al.*, (2000), to evaluate the destructive effects of Cisplatin on newborn development, it has been reported that the drug has destructive effects on rats' fetus development which is consistent with the results of our study.

The obtained results from dead fetus counting suggest that Cisplatin imposes a meaningful difference between the control and experimental groups.

In a study conducted by Peterka *et al.*, (2002), on toxic effects of Cisplatin-procaine on chickens' fetus, the lethality and teratogenic effect of the drug on intra-egg period of chicks was evaluated which is consistent with the results of our study.

Based on the results obtained from external abnormalities observation it can be concluded that the use of Cisplatin during pregnancy can impose abnormal rotations of fetus body, abnormal rotation of fetus anterior and posterior limbs, wrinkled skin, local hemorrhage, tail and eye disorders, and lack of brain. In a study conducted by Peterka *et al.*, (2002), administration of Cisplatin and procaine in chickens' fetus caused some abnormalities such as micro ophthalmia, microcephalia, deformity of limbs and ets which is consistent with the results of our study.

Discussion

The occurrence of abnormalities in fetus from mothers treated by chemotherapy is not the result of the simple detectable and controllable factors but probably it is under the effect of more complex interactions among predisposing factors. It is suggested that cancerous patients doesn't use Cisplatin under any circumstance during pregnancy and lactating; so, if the use of the drug is necessary it is suggested that a complex of different anti oxidants such as arachidonic acid, ascorbic acid, super oxide dismutase, and antioxidants especially vitamins E and C be used.

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