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Glycogen Contents and Related Enzymes in Placenta and Fetal Organs of Rats Exposed to Titanium Trichloride During Pregnancy

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ABSTRACT

Exposure of Titanium trichloride to pregnant rats during the period of organogenesis is known to induce sever embryotoxicity, however, the overall mechanism is not know. Disturbances in the carbohydrate metabolism in the exposed mother may induced physiological alteration which are transported to the fetuses through Placenta. Glycogen contents, the activity of adenosine triphoshatase and glucose-6-phosphatase play an important role in inducing embryotoxicity and therefore these parameters has been studied in placenta, fetal liver and kidney of rats exposed to titanium trichloride. Doses of titanium trichloride 0.72 ml/kg of the body weight (1/5th of LD_{50}) were administered orally once on 9th and 15th day of gestation p.c. and 0.36 ml/kg of the body weight (1/10 of LD_{50}) were also administered orally daily from 6th to 14th day and 15th to 20th day of gestation. It has been observed that at doese of 0.72 and 0.36 ml/kg of body weight during organogeneises (from 6th to 14th and 9th day of gestation) and fetal development (from 15th to 20th and 15th day of gestation) significant depletion in glycogen contents in fetal livker and placenta was observed, however, no changes were recorede in kidney. The activity of ATPase during organogenesis and fetal development causes significant increase in kidney and liver. However, the activity of Glucose-6-phosphatase during organogenesis was significantly decreased in liver, kidney and placenta. Disturbances in carbohydrate metabolism in treated animals may become the cause of embryotoxicity.

Key words: Biochemical alteration during pregnancy, placenta, fetal organs, Titanium trichloride.

1. INTRODUCTION

Titanium which is a grey metal, highly flammable, mustering nature and corrosive resistance, so used in explosives, electronic devices, paints, and cosmetics, It is also used in the confectioning, food and dairy industries as a potential additive. It's general toxicity is well established and it also known to induce mutagenicity but its effect on reproduction during pregnancy is not yet known in laboratory animals. Scheroedar and Mitchener reported same basic studies of titanium salts and its toxic effect on the reproduction of mice and rate through three generation (from F1 to F3 generation). Titanium was toxic with a marked reduction in the number of animals surviving to the third generation. The ratio of males and female pups was reduced and number of runts increased. During fetal development and organogenesis the supply of nutrients in the form of glycogen place a major supportive role and it has been reported that number of metals like lead and cadmium deplete the carbohydrate metabolism enzymes with result fetuses does not get full nutrients from the mother. Keeping in view this fact embryotoxic mechanism under the influence of titanium trichloride as reported earlier. Being evaluated in the present investigation selection of glycogen contents and related enzyme in mother and fetuses which may enlighten the mechanism of action.

2. MATERIALS AND METHODS

Adult Wistar female rats (140 .... 10g) of proven fertility were selected from the departmental animal colony. These animals were maintained under uniform husbandry conditions of light and temperature (26 ... 1ºC) and were given Hindustan Gold Mohur rat palliated diet and water ad libitum. Females were caged with males in the ratio of 2:1 Mating was confirmed by the presences of a vaginal plug and spermatozoa in the vaginal smear and the day considered as day one of pregnancy. Doese of titanium trichloride, 0.36ml/kg (LD50 4.30g/kg which was equivalent to 3.6ml/kg)2 were prepared in aqueous medium and were administered orally daily from 6th to and 15th to 20th day of gestation. Titanium trichloride was also administered at single dose orally at 0.72ml/kg on 9th and 15th day of gestation to separate sets of animals.

Animals were sacrificed on day 21st of gestation (a day before delivery). The uterine horms were removed by cesarean section, placenta and liver, kidney of fetuses were removed. Cleared tissues were weighed on monopan balance to nearest of mg and thereafter these were processed for biochemical for biochemical estimation, glycogen, the activity of G-6- Pase and At Pase. The results obtained were analysed statistically using analysis of variance (ANOVA).

3. RESULTS AND DISCUSSION

3.1 Effect on the glycogen contents

Findings revealed that 0.72 ml/kg of dose of titanium trichloride once on 9th and 15th day of gestation and 0.36ml/kg of dose daily from 6th to 15th to 20th day of gestation showed significant reduction in glycogen contents in liver and placenta (Table 1)

3.2 Effect on the activity of glucose-6 phosphatase

Results revealed that 0.72ml/kg and 0.36ml/kg dose of titanium trichloride on 9th and 6th to 14th day of gestation caused significant reduction in the activity of glucose-6-phosphatase in liver, kidney and placenta, where as during fetal development (15th to 20th day of gestation) no significant changes observed.(Table 1)

3.3 Effect on the activity of adenosine triphosphatase

Result showed that once 0.72ml/kg dose of titanium trichloride on 9th and 15th day of gestation and 0.36ml/kg of dose daily from 6th to 14th and 15th to 20th day of gestation showed significant increase in the activity of ATPase in liver and kidney. However, no significant changes were observed in placenta as compared to control. (Table 1)

Table 1 Effect of Titanium trichloride in fetal organs during a different period of gestation in rats

Parameters	Dose and period of administration	Treatment**	Liver	Kidney	Placenta
Glycogen (mg/100gm)	0.36ml/kg (6th to 14th day)	C	2645 ±159	146.2±1 1.3	985±52. 5
		E	1002±79 *	145.3±1 0.4	515±38. 2*
	0.72ml/kg (9th day)	E	127±79*	152.9±1 6.6	516±35. 8*
	0.36ml/kg (15th to 20th day)	E	1105±63 *	139.1±8. 37	694±34. 7*
	0.72ml/kg (15th day)	E	1970±15 2*	133.4±1 2.5	445±27. 5*
G-6 Pase (mgiP/100g/ min)	0.36ml/kg (6th to 14th day)	C	7.11±0.3 7	6.21±0.3 7	5.80±0.2 9
		E	4.70±0.3 2*	1.59±0.0 9*	1.27±0.0 8*
	0.72 ml/kg (9th day)	E	2.23±0.3 4*	1.93±0.1 3	2.53±0.2 0
	0.36 ml/kg (15th to 20 th day)	E	8.08±0.7 2	7.61±0.6 7*	6.53±0.6 8*
	0.72 ml/kg (15th day)	E	6.82±0.5 4	5.02±0.3 0	5.62±0.2 8
ATPase (mgiP/100g/ hr)	0.36ml/kg (6th to 14th day)	C	48.3±2.8 9	40.8±2.0 4	70.6±4.2 3
		E	65.3±3.8 7*	63.7±5.0 9*	80.6±6.4 4
	0.72ml/kg (9th day)	E	65.2±4.5 6*	62.5±3.7 5*	88.1±7.0 4
	0.36ml/kg (15th to 20th day)	E	85.2±6.8 1*	85.5±5.9 8*	89.4±7.1 5
	0.72ml/kg (15th day)	E	67.7±4.7 3*	63.1±3.7 8*	88.2±5.9 6

Under constant or sustained toxicological stress, innumerable biochemical and morphological changes occur in the vital and reproductive organs inducing serious pathological lesions. Due to the presence of fetus and placenta the metabolism and physiology of pregnant rats differs considerably to that of normal cyclic rats. Indu Bala reported that metabolism of carbohydrates, proteins and fats is altered in the presence of placental hormones. It is also known that the immediate source

of glucose for fetus is the maternal blood. The liver is an active metabolic organ even in the fetus as it synthesises compounds which are necessary for fetal homeostasis. Ghosh *et al* have observed that fetal liver glycogen was found to be usually stable where as glycogen in adult is relatively unstable. Numbers of workers have reported depletion of hepatic glycogen due to various chemicals, metals and their salts. After prolonged exposure to cadmium chloride Sporn *et al* observed loss of glycogen in liver. Exposure of metals like lead, mercury, copper, chromium molybdenum, and manganese treatment in the rats observed disturbance in liver glycogen Benable *et al* have reported the reduced glycolysis rate due to influence of high concentration of vanadium. Mathur *et al* and other workers have reported that glycogen contents of maternal liver, placenta, uterus and fetal liver were significantly reduced. In the present study administration of titanium trichloride during organogenesis (from 6th to 14th and 9th) and fetal development (15th and 15th to 20th day of gestation) showed significant depletion in glycogen contents in fetal liver and placenta. Hazelhoff Roelfzema *et al* have reported that in normal pregnancy, the glycogen contents of the placenta is increased from day 14 to 19, and decreased on day 20. Decreased in the level of glycogen as observed in liver and placenta may be due to the inhibition of the activity of certain enzymes in carbohydrates metabolism viz, glucokinase, hexokinase, phosphoglucomutase or glucose 6-phosphates dehydrogenase. So decreased glycogen level in the placenta indicates that titanium is acting on the placental membranes and is unable to supply nutritive material freely to the fetus. Consequently the fetus regulates its own blood glucose concentration therefore its level is decreased in the fetal liver. The mechanism for doing so do develop in the fetus, but is largely dormant until birth, when the supply of glucose from the placenta ends abruptly.

Abram *et al* reported that the activity of AT Pase increases in liver of rats when treated with chromic chloroquinine. Slater reported increased AT Pase activity after treatment with carbon tetrachloride. It has been reported that activity of AT Pase is inhibited by vanadium in higher animals. This supports our study that when titanium trichloride administered during organogenesis and fetal development caused significant increase in the activity of ATPase in kidney and liver. By workers significant decreased in the ATPase in maternal and fetal organs reported when beryllium nitrate was exposed to mother on 16, 18 and 20 day of pregnancy. In present studies the activity of ATPase was increased which may be due to the phosphorylation reaction of Na+, K+ and ATPase. It can also be correlated with decreased glycogen level. This may also be explained on the fact that titanium administration accelerates the break down of glycogen for which high energy ATPase is required.

The activity of glucose-6- phosphatase regulates the blood glucose level from the liver. The enzyme activity of glucose-6-phosphatase was found to be either absent or very low during fetal life and increase soon after birth or at adult stage. Number of workers reported that the activity of glucose-6-phosphatase in human fetal liver is about one third than that at the adult level. In fetus the enzyme activity starts to increase from 12 week of gestation. This supports our study that when titanium trichloride administered during period of organogenesis caused significant decrease in its enzyme activity glucose-6-phosphatase in liver, kidney and placenta however, its administration during the period of feta development did not show any change. These finding clearly show that clearly shown that energy producing substance are constantly utilised for which high ATPase is observed to catalyze the metabolic activities.

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REFERENCES

1. Lorenz K and Maga J. Functional and sensory properties of titanium dioxide as a flour and bread additive. Food Prod Develop 1973: 7: 93-98.
2. Kosikowski FV and Brown DP. Application of titaniumdioxide to White mozzarella cheese. J dairy Sci 1969; 968-970.
3. Schroeder HA and Mitchener M. Toxic effect of trace elements on the reproduction of mice and rats. Arch Env Health 1971;23;102-106.
4. Malik B. Embryo and fetotoxic effects of titanium compound in rats exposed during a pregnancy. Ph.D. these Jiwaji University Gwalior India 2000.
5. Malik B and Prakash AO. Estimation of Lethal dose (LD_{50}) of titanium salts in rats. Asian J Exp Sci 1996;10: 85-90.
6. Indubala. Study of Ane-Partum and Post serum cholesterol level in normal and toxemia of pregnancy. J Obst Gyna 1983;33.
7. Ghosh S, Thakurta GG and Mukherjee KL. Glycogen content and structure and some enzymes of glycogen metabolism in human foetal organs. India J Med Res 1989;90:147-153.
8. Rana SVS, Prakash R, Kumar A and Sharma CB. A Study of glycogen in the liver of metal Fed rats. Toxicol lett 1985;29: 1-4.

9. Mathur S, Sharma S, Prakash AO and Mathur R. Beryllium induced glycogen mobilization in albino rats. Asian J of Exp Sci 1987;3: 37-44.
10. Rastogi S and Rana SVS. Influence of parathyoidectormy on liver glycogen in rats treated with Carbon tetrachloride. Ind J Exp Biol 1990;28: 794.
11. Sharma A Mathur R and Shukla S. Hepatoprotective action of a propriety of herbal preparation against Carbon tetrachloride intoxication. India Drugs 1994;32:120-125.
12. Spron A, Dinu I. and Stoeneseu I. Influence of Cadmium administration on carbohydrates and cellular energy metabolism in the rat liver. Rev Roum Biochem 1970;7:299.
13. Benabe JE, Echegoyen LA, Pastrana B and Murtinez- Maldonado M. Mechanism of inhibition of glycolysis of Vanadate. J Biol Chem 1987;262:9555-9560.
14. Mathur S, Mathur R and Prakash AO. Possible mechanism of transplacenta absorption of beryllium in rats. Trace and toxic elements in nutrition and health (proceeding of the fourth International Conference on Health and Disease) New Delhi, feb-12, Wiley Eastern Limited (New Age International published) 1993;129
15. Mathur S, Flora SJS, Mathur R, Kannan GM and Das Gupta S. Beryllium induced biochemical alteration and their prevention following co-adminitration of Meso-2,3 dimercapto succinct acid 2,3 dimercato propane Sulphonats in rats. J Appl Toxicol 1994;14:263.
16. Hazelhoff Reolfzema W, Roelofsen AM and Copius Peereboom-Stegeman JHJ. Glycogen contents of placenta and of fetal and maternal liver in cadmium exposed rats. 1:A descriptive light microscopic study. Placenta 1987;8:27-36.
17. Aldridge WN and Thomas, M. The inhibition of phosphoglucomutase by beryllium. Biochem J 1966;98:100
18. Hassan S S and Kushwasha AKS. Effect of Datura (seeds) on rats brain and unitary metabolites. Pol Pharmac of Pharma 1986;38:329-344.
19. Austin C R and Short RV. Embryonic and Fetal Development, Cambridge University Press, London. New York. Melbourne;1975.
20. Abraham R Golbery L and Grasso P. Nature, 215:194 Cited by Gerald Weimann In: Lysosomes in Biology and Pathology, Part 1, (Dingle, J.T. and Tell, H.B.eds.) North-Holland publication, Amsterdam, London. 1967;276
21. Slater TF. Lysosmes and experimentally induced tissue injury cited by Dingle, J.T. and Fell, H.B. (1973) "Lysosomes in biology and pathology "Part 1 (Dingle, J.T. and Fell, H.B. eds.) North-Holland Publishing Company, Amsterdam, London 1973;467.
22. Jandhyala BS and Hom GJ. Physiology and Pharmacological Properties of Vanadium. Life Sci 1983;33:1325.
23. Jones CT. Comparative aspect of hepatic glucose metabolism during foetal development. Biochem Soc Trans 1981;9:375.
24. Auricchio S and Rigillo N. Glucose-6- Phosphates activity of the human foetal liver. Biol Neonat 1960;2:146.