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Zinc Protection against Cadmium Effects on Preimplantation Mice Embryos

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Cadmium, one of the most toxic heavy metals, produces congenital malformations, developmental retardation and death in amphibians (Perez Coll et al 1986) as well as in laboratory mammals (Ferm 1972). Although the development of preimplantation mammalian embryos may be impaired by environmental chemicals (Blackburn and Clegg 1979) it has been reported that Cd^{++} administered on day 3 of gestation exerts only a slight reduction in the number of blastocysts (Giovani et al 1980). However, the further capacity of those embryos to hold postblastocystical development was not evaluated. On the other hand, the protective role of Zn^{++} against several etiological agents which produce malformations (Dreosti et al 1986, Eckert and Hurley 1979) including Cd^{++} (Hackman and Hurley 1981) is well documented. The present study was undertaken to investigate the effects of Cd^{++} administered in preimplantation stages of mouse embryos and to evaluate the eventual protective role of Zn^{++} against the efects of Cd.

MATERIAL AND METHODS

Sixty virgin female Rockland mice from our own stock, 30-35 grs body weight, maintained under laboratory conditions, were caged overnight with males of the strain; the morning on which a vaginal plug was found was designated as the first day of pregnancy. The mated females were randomly divided into 4 groups and treated intraperitoneally with 0.5 ml/animal of saline, 2.2 mg/Kg Zn SO_4 and 2.2 mg/Kg Cd Cl_2 + 2.2 mg/Kg Zn SO_4, respectively, on day 5 of gestation. The cadmium dose level was nearly equivalent to the teratogenic dose 50 for rats (Mc Clain and Becker 1975). Females were sacrificed at 15.00 hr. on day 9 of gestation. The uterine horns and ovaries were exposed and the number of corpora lutea and implanted embryos were recorded. Considering that in about 33 °/o of the females treated with Cd Cl_2, an almost total reduction in the number of implanted embryos was recorded we also compared this group with the rest (67 °/o) of the cadmium treated females. Data were analysed by the Mann-Whitney test.

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table 1: effects of $CdCl_2$ and its prevention by $ZnSO_4$ in pregnant mice on day 5 of gestation.

treatment	number of experimental females	mean number of luteal corp ± SD	mean number of fetus ± SD	number of fetus
saline (0.5ml/animal)	15	11.00 ± 2.13	9.46 ± 2.47	142
$CdCl_2$ (2.2mg/kg)	15	11.46 ± 2.44	7.00 ± 5.52*	104
$CdCl_2$+$ZnSO_4$ (2.2mg/kg resp)	13	10.69 ± 1.65	11.15 ± 1.77	144
$ZnSO_4$ (2.2mg/kg)	15	11.93 ± 1.70	9.13 ± 2.82	137

* $p < 0.04$ Mann-Whitney test.

table 2: number of implanted embryos in (A) slightly affected and (B) severely affected females.

	luteal corp		fetus	
	group A	group B	group A	group B
number of experimental females	10	5	10	5
mean	11.70 ± 2.58	10.90 ± 2.33	10.30 ± 3.05	0.20 ± 0.44

RESULTS AND DISCUSSION

The results of this investigation are shown in Table 1. Although the administration of cadmium at nearly equivalent to teratogenic dose 50 in pregnant rats at preimplantational stages of embryonic development exerted only a very slight reduction in the number of blastocysts (Giavini et al 1980), in mice, according to our results a very significative reduction in the number of implanted embryos was obtained. This difference could be attributed to I) the number of blastomeres is not in the case of cadmium effects a good indicator of embryonic damage II) a differential susceptibility between rats and mice in the teratological effects of cadmium and III) the administered on day 3 of pregnancy exerted a reduced effect when compared with day 5 of pregnancy. According to preliminary data obtained in our laboratory, this last assumption appears as most probable. It is note worthly that the reduction in the number of implanted embryos occurs drastically but only in some of the cadmium treated pregnant females (Table 2). Therefore it seems that at this early stage of mouse development if cadmium toxicity results in lethality, almost all the embryos will be affected. The administration of zinc simultaneously with cadmium prevents its harmful effects. It is widely accepted that zinc is an essential element for the biological activity of several metalloenzymes such as RNA and DNA polymerases as well as other enzymes involved in protein synthesis and degradation (Vallee and Falchuk 1981, Cousins 1986). It is noteworthly that the protective effect of zinc against cadmium toxicity in mouse embryos is achieved at a ratio 1 Cd^{++} /1 Zn^{++} while in free living embryos (Bufo arenarum) the proportion for protection was 1 Cd^{++}/8 Zn^{++}(Herkovits et al 1987). Therefore, it seems that the evaluation of the protective effect of zinc against cadmium toxicity requires a very careful study in each species and even in each developmental stage in order to detect the proper ratio between both heavy metals capable of preventing the teratological and lethal effects exerted by cadmium.

The question whether cadmium effects are exerted directly on the embryo or by alterations of the maternal tissue or by both, could be evaluated at this early stage of development by embryonic transplantation experiments which are in progress.

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