DOSE RESPONSE RELATIONSHIP FOR CISPLATIN INDUCED MICRONUCLEI IN BONE MARROW ERYTHROCYTES OF SWISS ALBINO MICE

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INTRODUCTION
Cisplatin is one of the most widely used chemotherapeutic drugs. The antitumor properties of this drug were reported. It is effective against a wide variety of animal tumors and human cancers (Calabresi and Parks, 1985) like ovarian, head, neck, bladder, cervical, testicular teratoma, non-small-cell lung carcinoma and lung cancers, both as a single agent and in combination with other agents (Muggia, 1984; Mugdy et al., 2003). CP is generally considered to exert its cytotoxic effect by binding its highly reactive hydrated platinum complex to DNA (Erickson et al., 1981) resulting in mutation induction (Fichtinger et al., 1984). Along with its therapeutic activity of this drug it is known for its toxic effects like nephrotoxicity, neurotoxicity, otoxicity, immunotoxicity, embryotoxicity, cytotoxicity, hepatotoxicity, mutagenecity, myelotoxicity, teratogenicity and severe nausea and vomitings (Lazar et al., 1978; Von Hoff et al., 1979; Sorsa et al., 1985; Anthony et al., 1988; Fillastre and Raguenex Viotte, 1989; Barbara et al., 1996; Vijayalaxmi and D’Souza, 2004; Nersesyan and Muradyan, 2004; Thomas et al., 2004; Yingjun et al., 2008) in different test systems. It is also a potential human carcinogen as it develops secondary malignancies in patients who have been treated with CP.

In the present investigation the incidence of micronuclei in bone marrow erythrocytes of mice by cisplatin was carried out.

MATERIALS AND METHODS
Swiss albino mice weighing about 22-24 g aged 8-10 weeks old were utilized in the present study. The drug was supplied by Reddy Labs Hyderabad. For each dose group of five animals were used. The animals were fed with 2.5, 5 and 10 mg/kg adriamycin intraperitonally in two installments within 24 hr interval. The control group of mice received 0.5 physiological saline simultaneously. The animals were scarified 6h after the last administration and smears were stained with Giemsa May Grunwald stain. The prepared slides were screened for the presence of micronuclei in polychromatic erythrocytes of mice. A significant increase in the frequency of micronuclei in poly chromatic erythrocytes was noted when compared with control values.

RESULTS
The results on the induction of micronuclei in bone marrow erythrocytes of mice were depicted in Table 1. In cisplatin treated animals there was an increase in the polychromatic cells with micronuclei (Table 1). The frequency in control
animal was 0.25% and the values were 0.60%, 0.85% and 1.20% after the treatment of 2.5, 5 and 10 mg/kg cisplatin respectively (Table 1). The percentage of normochromatic cells with micronuclei was 0.28% in control mice, while the frequencies were 0.62%, 0.81%, 1.24% at 2.5, 5 and 10 mg/kg cisplatin respectively (Table 1). The P/N ratio was 0.99 in control mice and it has decreased at all dose levels. The differences in the frequencies of micronuclei in polychromatic cells were significant between the control and cisplatin treated groups (p<0.01). Thus, the data showed dose response relationship in the frequency of micronuclei in polychromatic erythrocytes.

## DISCUSSION

The in vivo micronucleus test is one of best methods to screen the clastogenic effects of chemicals and drugs (Chaubey et al., 1978) using this procedure the mutagenicity of various alkylating agents (Maier and Schmid, 1976; Rudrama Devi et al., 1985, 1986, 1987) drugs (Rudrama Devi and Reddy, 1985, 1986, 1987) drugs (Rudrama Devi and Reddy, 1995) was also established.

The results clearly indicate that there was gradual increase in the frequency of various types of chromosomal aberrations with increase in dose and time intervals. Similar reports are also available on the toxic effects of platinum compounds. The chemotherapeutic agent cisplatin has been shown to be highly mutagenic, teratogenic, and carcinogenic (Morrison et al., 1981; Barbara 1996; Giri et al., 1998) in both in vitro and in vivo experimental models. Early activation of oncogenes was observed in human headneck tumors after treatment with the Cisplatin (Németh et al., 2002).

The cytotoxic and genotoxic effects of cisplatin was proved by using comet assay as an experimental protocol by Chandrasekar et al. (2006); Misra and Choudhury (2006); Premkumar et al. (2006), Sabry et al. (2008) and micronucleus Nersesyan and Muradyan, (2004) in different experimental models.

The frequencies of chromosomal aberrations increased non-linearly after intraperitonial administration of cisplatin with an average of 2.7 aberrations per aberrant cell in bone marrow of mice (Giri et al., 1998) independent cytotoxic agent (Würschmidt et al., 2000) Apoptosis of normal cells of the small intestinal epithelium and the bone marrow was also observed by Tamaki et al., (2003) after the administration of cisplatin in mice. The same was observed in the Hypopharyngeal Carcinoma Cell Lines by Byung et al. (2005). The present results are comparable with that of Choudhury et al., (2000) who reported the effect of Vinblastine (VBL); a vinca alkaloid is a chemotherapeutic agent in male mice for cytotoxicity. The results showed decreased mitotic activity of cells and significant decrease in the percentage of micronucleus in somatic cells of mice. Decarbazine (DTIC) a chemotherapeutic agent has been successfully applied to treat various types of cancer such as Hodgkin’s disease, malignant melanomas, soft tissue sarcomas and advanced neuroblastomas. In the bone marrow micronucleus assay DTIC induced micronuclei at range of 0-125 mg/kg. Present study proved the highly genotoxicity of CP in bone marrow cells of mice. Further observations are necessary to find out the mechanism by which cisplatin induce genetic damage.

### REFERENCES

Anthony, Gringeri, Peter, C. Keng, and Richard, F. Horch. 1988. Diethyldithiocarbamate Inhibition of Murine Bone Marrow Toxicity Caused by cis-Diaminedichloroplatinum (II) or Diammine – (I,I-


Lazar, P., Conran, C. and Damjanov, I. 1978. Embryotoxicity and teratogenicity of cisiamminedichloroplatinum (II). Experimental. 33:

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### Table 1: Results on the frequencies of micronuclei in the bone marrow erythrocytes of mice treated with various doses of cisplatin

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Micronuclei in polychromatic cells (P)</th>
<th>Micronuclei in normochromatic cells (N)</th>
<th>Micronuclei in total (P+N) cells</th>
<th>P/N ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>45/16000 (0.28)</td>
<td>15/16022 (0.09)</td>
<td>60/32022 (0.99)</td>
<td></td>
</tr>
<tr>
<td>2.5 mg/kg</td>
<td>99/16000 (0.62)*</td>
<td>34/16840 (0.26)</td>
<td>133/32840 (0.95)</td>
<td></td>
</tr>
<tr>
<td>5 mg/kg</td>
<td>130/16000 (0.81)*</td>
<td>45/17200 (0.26)</td>
<td>175/33200 (0.93)</td>
<td></td>
</tr>
<tr>
<td>10 mg/kg</td>
<td>198/16000 (1.24)*</td>
<td>70/20250 (0.35)</td>
<td>268/36250 (0.79)</td>
<td></td>
</tr>
</tbody>
</table>

*p<0.05


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