

Embryotoxicity of cisplatin and a cisplatin-procaine complex (DPR) studied in chick embryo^{*}

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Received July 17, 2002

Cisplatin is widely used as an antitumor drug. To reduce its toxic side effects in patients, cisplatin has been bound with procaine in a cisplatin-procaine complex (DPR). The lethal and teratogenic effects of cisplatin alone and of complexed cisplatin were determined in the chick embryo *in ovo* in order to compare their influence on rapidly proliferating embryonic tissues. The embryotoxic (lethal + teratogenic) effect was examined after a single intra-amniotic injection of one of six different doses, ranging from 0.03 to 30.0 µg, on embryonic days (ED) 3, 4 or 5. The minimal embryotoxic dose was lower for cisplatin alone (0.03–0.3 µg) than for cisplatin in the DPR complex (0.3–3.0 µg), suggesting that cisplatin alone is more embryotoxic than complexed cisplatin. Both substances caused malformations in the surviving embryos evaluated on ED 9. These malformations included microphthalmia, microcephaly, hypoplasia of the upper and lower jaw, cleft beak, and haemocephaly. Moreover, heart septum defects and limb reduction deformities were found after exposure to the DPR complex. The embryotoxicity of complexed cisplatin exhibited a stage-response effect. It was highest on day 3 and gradually decreased until ED 5. Such an apparent stage-response effect was not observed for cisplatin alone. The embryotoxicity of procaine hydrochloride – a component of the complex – was also tested. Procaine hydrochloride alone did not produce any embryotoxic effect, not even after a single injection of the maximal tested dose (100.0 µg per embryo). We also examined the protective effect of procaine hydrochloride, whose separate administration at ED 4 was followed by the injection of 0.3 µg cisplatin. We did not observe any protective effect of procaine hydrochloride if injected separately.

Key words: Cisplatin, cisplatin procaine complex DPR, teratogen, malformation.

Cisplatin is an important antitumor drug widely used in the therapy of solid malignancies, particularly in ovarian and testicular cancers [22, 31, 32]. Significant side effects of nephrologic, neurologic and gastrointestinal origins may limit the dosage of cisplatin. Therefore, considerable effort has been made to reduce the toxic side effects of cisplatin in non-neoplastic tissues. Different chemoprotective agents have been used; para-aminobenzoic acid, procaine hydrochloride and procainamide have been used successfully to reduce cisplatin-induced nephrotoxicity

and hepatotoxicity [3, 4, 5, 37, 38, 39, 41]. Nimodipine and WR-2721 were also shown to have protective effects both in experimental *in vivo* models [10] and in patients [20].

Another option for circumventing the toxic side effects of cisplatin is the synthesis of less toxic cisplatin analogues [19] or cisplatin complexes. A platinum triamine complex containing cisplatin and procaine hydrochloride (DPR) was synthesized several years ago [2]. This complex exhibits an anticancer activity comparable with the original drug cisplatin, but its toxicity for non-neoplastic tissues appears to be considerably lower under *in vitro* and *in vivo* experimental conditions [2, 18, 35, 36, 40].

The experience of many laboratories [15, 16, 28, 29, 30], as well as our previous findings [23, 24, 25, 26], support the chick embryo *in ovo* as a convenient experimental system

^{*}This study was supported by a grant project KONTAKT, based on the Agreement on Scientific and Technical Cooperation between Italy and the Czech Republic and AVOZ5039906.