Effects of Poly-plat, SSP, SAP and Cisplatin on the Testis of Rats

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ABSTRACT

In the present study, Polyglat, SSP and SAP are second generation analogs of cisplatin, the main and most active representative of antitumor platinum complexes. On the morphology of the testis of male rats were investigated histologically by examining semithink section. Polypoly, SSP and SAP administered in doses of 10 mg/kg and applicable as a single intraperitoneal injection. For comparison purpose, the parent compound Cisplatin [cis-diamminedichloroplatinum (II)] as administered at equitoxic doses (1.8 mg/kg) at various intervals between days 1, and 30. Four compounds effected structural alteration of Sertoli cells, disrupted the blood testis barrier and impaired the processes both of spermatogenesis and spermiohistogensis in different degrees. The structural damage in testis following treatment by mean of Polyplat, SSP and SAP were at least as pronounced as that occurring under the influence of equi toxic doses of cisplatin. Results show Polyplat, SSP, and SAP were changed small structure of testis in morphology comparing with control group. But all side-effects of them are more small than cisplatin.

Key Words: Cisplatin, Ply-plat, SSP, SAP, Morphology, Platinum compound, Cell structure

Introduction

Since the detection of antitumor properties of some platinum compounds by Tosemgerg and coworker in 1969 **[9]**. The inorganic complex cisplatin [cis-diamminedichloroplatinum (II)] has been confirmed to be one of the most board spectrum anticencer drugs available today for the clinical treatment of human solid carcinomas **[10, 13]**. It is proven to be effective in the treatment of testicular, ovarian, uristat , bladder, head, neck ,lung cancers and enhanced immuno stimulation **[1, 3, 6, 10, 14, 15, 17, 18]**. However, as cisplatin is burdened by severe side-effects. Such as nephrotoxicity, neurotoxicity, gastrointestinal irritation and a profound disturbance of the patients' general behavior, the clinical administration of cisplatin can be actually limited by the interference of non-tolerable side-effects **[2, 4, 7]**. Poly-plat Poly-[(trans-1,2-diaminocyclohexane) platinum]-carboxyamylose, SSP [(5-sulfosalicylato-trans-(1,2-diaminocyclohexane) platinum] and SAP [(4-hygroxy-α-sulfonylphenylacetato (Trans-1,2-diamonocyclohexane) platinum II)] as well as carboplatin are second generation analogsofcisplatin with higher efficacy and potency, while eliciting less toxicity **[16]** No invention is yet available can concerning the influence of Poly-plat, SSP and SAP. Considering these increasingly replaced cisplatin in clinical chemotherapy, we investigated in the present study the influence on the Poly-plat, SSP and SAP appearance of the testis of rat in comparison to cisplatin applied at equitoxic dose levels.

Materials and Methods

Male Waster rat, purchased from Animal Center of Michigan State University, were kept under standard conditions and received food and rap water. At the beginning of the experiment, they were 132-148 g and were treated with single intraperitoneal injections of Poly-plat (10mg/kg), SSP (10mg/kg), SAP (10mg/kg) and cisplatin. 0.18mg/kg/day at intervals of 1 and 5 days, then killed by 7, 12, 20, 30 days after first injection. Two animals of each group were anaesthetized and perfused with a fixative solution. For this purpose, the thorax of the animals was opened, injected 0.9 ml of equithesin I.P. for each rat, insert the needle into left ventricle and cut the right atrium. Perfused with normal saline for 2 min and put 4% paraformaldehyde 15 min. Thereafter, the testis were removed. Immersed for another 24 h in fixative solution. postfixed with a 1% solution of osmium tetroxide in cacodylate buffer, dehydrated. Semitic sections of 8 um were prepared, mounted and stained with pigment.

Influence of cisplatin upon testicular morphology

When cisplatin was applied as the dosage (1.8 mg/kg), the rat's intraepithelial intercellular spaces enlarged and the contact between the cells loosened within 7 days. Whereas the germinal epithelium of untreated seminiferous tubules consisted of densely arranged spermatogenic cells in different developmental stages. Basement membrane encircling the seminiferous tubules became irregularly wound and germine epithelium was no longer straight. Cleft , vacancy and necrotic cells appeared in the middle and luminal areas of occupied spermatids as well as in the base layer around spermatogonia and Stertoli cells. Spermatogonia are neither clear nor regular on the germine epithelium comparing to control group. These results are same Petra's report **[19]** [low dose of cisplatin (3 mg/kg) was sufficient to induce profound structural alterations within the testicular tissue of mice].

Following days, the shape of cells of testis were changed seriously. The layers of Spermatogonia, primary spermatocytes, and seminiferous tubules were confused till 12 days. The malformed spermatozoa occurred in luminal regions and necrotic spermatogenic cells were seen.

A great number of abmortal mitotic figures with condensed chromosomes in a the primary spermatocytes **[11, 12]** were are easily identifiable by their typical propahist nucleus. Arrested mitotic figures were detected between and above primary spermatocytes, and other cells with three or four nuclei were found.

Beginning between day 13 and 20, The cell began to recover, but still had a few cleft , vacancy and necrotic cells appeared in the middle and luminal areas of occupied spermatids. Till day 20 to 30, most cells in testis were not all recovered like before. The layers of Spermatogonia, primary spermatocytes , and seminiferous tubules didn't distinguish. A lot of Stertoli cells occurred in all middle and luminal areas.

Influence of Poly-plat, SSP and SAP upon testicular morphology

Poly-plat:

Basement membrane encircling the seminiferous tubules became irregularly wound and was no longer straight. Cleft, vacancy and necrotic cells appeared in the middle and luminal areas o f occupied spermatids as well as in the base layer around spermatogonia and Stertoli cells. Spermatogonia are more clear and no regular on the germine epithelium than control group. Arrested mitotic figures were detected between and above primary spermatocytes and other cells. The nuclei were found abnormal mitoses of spermatogonia with condensed chromosome.

SSP:

Basement membrane encircling the seminiferous tubules became irregular swell and not long strait and smooth normal like control group as cycle. Cleft , vacancy and necrotic cells appeared in the middle and luminal areas of occupied spermatids as well as in the base layer around spermatogonia and Stertoli cells. Spermatogonia seem to be more layers than normal structure. Arrested mitotic figures were detected between and above primary spermatocytes. The nuclei were found abnormal mitoses of spermatogonia.

SAP:

Basement membrane encircling the seminiferous tubules looked regular and was cycle like normal group. The cleft, vacancy and necrotic cells seldom appeared in the middle and luminal areas of occupied spermatids as well as in the base layer around spermatogonia and Stertoli cells. Spermatogonia seem to be regulation on the germine epithelium comparing to control group. Arrested mitotic figures were not detected between and above primary spermatocytes and other cells. The nuclei were found normal mitoses of spermatogonia.

Conclusion

Compare to cisplatin treatment there are much less damages of testis's structure for the above three platinum compound (Poly-plat, SSP and SAP) though their structures have a little different change. The degrees of damage and toxicity on morphology are Cisplatin>Poly-plat>SSP> SAP. The second generation analog of cisplatin is really less side-effect on rat's testis. But no toxicity no experiments evidence completely.

Discussion

As the second generation analog of cisplatin, Polyplat's toxicity was only 1/15 of cisplatin

according to research report. But to rat's testis it was disappointing the conclusion. From a morphological point of view, Polyplat and SSP had a toxicity for the layers of Spermatogonia, primary spermatocytes, and seminiferous tubules as well as mitoses of spermatogonia. These compounds witch contained Pt can damage and penetrate the blood/ testis barrier. It can connect with DNA and make degeneration of rats.

For Spermatogonia and spermatocyte's damage, Cisplatin and carboplatin were reported. As consequence of the violation of this barrier, the processes of spermatogenesis are disturbed by the cytotoxic action of Polyplat and SSP like cisplatin. The numerous arrested mitotic figures appearing during spermatocytogensis and both meiotic divisions following applications of Polyplat and SSP are probably the consequence of molecular interactions between Platinum-containing consecutive products of them and DNA molecules, witch was same cisplatin and carboplatin. Besides impairing spermatocytogenesis and meiotic divisions, both compound were also able to disturb the differentiation processes of spermiohistogenesis like vincristine, procarbazine [5]. But SAP showed a normal structure and exciting result like normal group. It may provide a less side-effect. Platinum hsa stronger effective for anti cancer than Polyplat and SSP. This may have relationship with its structure. It is difficult to explain the real reason. Therefore as a medicine for anti cancer, the first choose should be as follow: SAP [(4-hygroxy-α- sulfonylphenylacetato (Trans-1,2-diamonocyclohexane) platinum II)], SSP [(5-sulfosalicylato-trans-(1,2-diaminocyclohexane) platinum, Polyplat (Poly-[(trans-1,2-diaminocyclohexane) platinum]-carboxyamylose), Carboplatin [diammine (cyclobutane-1, 1-dicaroxylato) platinum (II)] and Cisplatin [cis-diamminedichloroplatinum(II)].

References

1. Hansen PV, Trykker H, Helkjaer PE, Andersen J (1989) Testicular function in patients with testicular cancer treated with orchiectomy alone of orchiectomy alone or orchiectomy plus cisplatin-based chemotherapy. J Natl Cancer Inst 81: 1246-1249

2. Harrap KR. Jones M, Wilkinson CR, Clink HM, Sparrow S, Mitchley BCV, Clarke S, Veasey A (1984) Antitumor, toxic and biochemical properties of cisplatin and eight other platinum complexes. Dev Onvol 17: 193:-212

3. Hodel C, Ettlin RA, Zschauer A (1984) Morphological changes produced in rat testis by anticancer drugs. Arch toxical Suppl 7:147-150

4. Pogach LM Lee Y. Gugkui W, Naymff M, Huang HFS(1989) Zinc acetate pretreatment ameliorates cisplatin-induced Stertoli cell dysfunction in Sprague-Dawley rats. Cancer Chemother Pharmacol 24: 177-180

5. Pogach LM. Lee Y., Gould S, Giglio W, Meyenhofer M, Huang HFS function in rodents. Toxicol Appl Pharmacol 98:350-361

6. Rose WC, Schurig JE, Haftalen JB, Bradner WT (1982) Antitumor activity and toxicity of cisplatin analogs. Cancer Treat Rep 66:135-146

7. Vawda AI, Davies AG. Effects of cisplatin on the mouse testis. Acta Endocrinol 112: 436-441

8. Williams SD , Nichols CR, Jansen J (1989) Use of carboplatin in the treatment of testicular cancer. Semin Oncol 16 [Suppl 5]: 42-44

9. Rosenberg B, Van Camp L, "Trosko JE, Mansour VH (1969) Platinum compounds: a new class of potent antitumour agents. Nature 222; 385-386

10.Rosenzweig M, Von Heff DD, Abele R, Muggia FM (1979) Cisplatin, Cancer Chemotherapy. The EORTC Cancer Chemotherapy Annual 1. Experpta Medica, Amsterdam, pp 107-125

11.Sundquist W, Lippard SJ (1990) The coordination chemistry of platinum anticancer drugs and related compounds with DNA. Coord Chem Rev 100:293-322

12.Terheggen PMAB (1990) Interaction of cisplatin and carbplatin with cellular DNA. Pharm Weekbl [Sci] 12:119-120

13. Von Hoff DD. Rosenzweig M (1990) cis-Diamminedichloroplatimum (II): a metal complex with significant anticancer activity. Adv Pharmacol Chemother 16:273-298

14. Herther J Muenchen, Surinder K Aggarwal, Hemant K misra and Peter J Andrulis, Enhanced immunostimulationby novel platinum anticancer agerts. Anti-cancer Drus 1997, 8. pp 323-328

15. Nicolini M. Platinum, and other metal coordination compounds in cancer chemotherapy. Boston: Martinus Nijhoff; 1990

16. Fiebig HH, Dress M, Ruhnau T, Misra HK, Andrulis P, Hendriks HR. GB-21, a movel platinum complex with antitumor activity in

human renal and mammary xenograft. Proc Am Ass Cancer Res 1996; 37:297

17. Mangione C, Bolis G, Pecorelli S. Bragman K, Epis A, Favalli G, Gambino A, landoni F, Presti M, Torri W, Vassena L, Zanaboni F, Marsoni S (1989) Randomized trial in advanced overran cancer comparing cisplatin and carboplatin. J Natl Cencer Inst 81: 1464-1471

18. Pekham MJ Horwich A, Hendry WF (1958) Advanced seminoma: treatment with cis-platinum-based combination chemotherapy or carboplatin (JM8). Cancer 52: 7-13

Petra Kopf- Maier, Effects of carboplatin on the testis. Cancer Chemother Pharmacol (1992) 29: 227-235