Journal of Medical Sciences

ISSN 1682-4474

science alert

ANSInet
an open access publisher
http://ansinet.com

Research Paper

JMS (ISSN 1682-4474) is an International, peer-reviewed scientific journal that publishes original article in experimental & clinical medicine and related disciplines such as molecular biology, biochemistry, genetics, biophysics, bio-and medical technology. JMS is issued four times per year on paper and in electronic format.

For further information about this article or if you need reprints, please contact:

Seyed Jalal Hosseinimehr
Department of Medicinal Chemistry,
Faculty of Pharmacy,
Mazandaran University of Medical Sciences,
Sari, Iran

Fax: +98-151-3253189

J. Med. Sci., 6 (1): 48-51
January-March, 2006

The Protective Effect of Amifostine Against Radioactive Phosphorous (^{32}P) in Mice Peripheral Blood Cells

[1]Mohsen Froughizadeh and [2]Seyed Jalal Hosseinimehr

The use of radioprotectors such as WR-2721 in radioactive phosphorous is interested because radiation delivered to tumor organ therapy can induce side effects in the bone marrow and most normal organs. To evaluate the potential of WR-2721, we treated mice with this drug (400 mg kg^{-1}) prior administration of ^{32}P at dose 4 μCi g^{-1} intraperitoneally. We determined hematological parameters in treated mice with ^{32}P or WR-2721+^{32}P at twenty days after treatment. Administration of radioprotector failed to enhance RBC, WBC, PLT, Hb and Hct from ^{32}P. In contrast, this radioprotector provided significant protection from total-body gamma irradiation with a DRF 1.91. These results suggest that the potential role of WR-2721 in a ^{32}P therapy is limited because of the difficulty in achieving continuous protection with a single dose of this drug due to its short half-life.

Key words: Radioprotective, radioactive phosphorous, WR-2721, amifostine, hematopoietic

ANSInet
Asian Network for Scientific Information

[1]Novin Medical Radiation Institute, Tehran, Iran
[2]Department of Medicinal Chemistry, Faculty of Pharmacy, Mazandaran University of Medical Sciences, Sari, Iran

INTRODUCTION

The ionizing radiation can be given to treat malignant tumors are often limited by the side effects to normal tissues and organs. The availability of agents that could protect normal tissue from the damaging effects of radiation would allow increased doses and may improved the outcome of radiation therapy. The majority of chemicals that are tested for radioprotective activity are sulphydryl compounds. Amifostine (WR-2721) is the most radioprotective agent to use in clinical practice in radiation therapy[1-3].

The radioactive phosphorous, ^{32}P, is used widely in medicine and biochemical research. Phosphorous-32 is used as a therapeutic agent for the treatment of polycythemia vera in initial dose of 50 to 100 μCi kg^{-1} of body weight of an isotonic solution of $Na_2H^{32}PO_4$ given intravenously or orally. ^{32}P is a pure beta emitter with a maximum energy of 1.7 Mev with a physical half-life 14.3 days. When injected intravenously, ^{32}P has stayed several weeks in the body[4-6].

Although amifostine established to have protective effects against whole body irradiation but there is a little literature to study effects of amifostine against continuously irradiation such as phosphorous-32.

To define the ability of amifostine to ameliorate the effects of ^{32}P in peripheral blood cells in the present study we administrated WR-2721 to mice before administration of ^{32}P. In this line, we studied the protective effects of amifostine against whole body gamma irradiation for percentage survival.

MATERIALS AND METHODS

Animals: Eight-week old male NMRI mice (Razi Institute of Iran) weighing 28±3 g were used. A standardized pellete diet was given and tap water was *ad libitum*. The animals were housed for one week in a quarantine facility. All of the mice were kept under a controlled lighting conditions (light: dark, 12:12 h) and temperature (22±1°C) in the university animal house.

Irradiation and treatment: Radioactive phosphorous, ^{32}P, was prepared from Iranian atomic energy organisation (Tehran, Iran). This drug has high purity and is used for medical application in Iran. Solution Phosphorous-32 was the administrated ip at dose 4 μCi g^{-1} in mouse. WR-3721 was administrated ip at dose 400 mg kg^{-1} at 30 min prior ^{32}P injection.

Whole-body irradiation was performed with a cobalt-60 γ-radiation source (Theratron 780, Canada). Mice were placed in ventilated plexiglass cages and irradiated in groups of ten mice simultaneously. The source-to-skin distance was 80 cm with a dose rate of 1.075 Gy min^{-1} at room temperature (23±2°C).

Hematological studies: At twenty days after phosphorous-32 treatment, blood sample (0.8-1.2 mL) was obtained from anesthetized mice by cardiac puncture using a heparinized syringe attached to a 21-guage needle. White Blood Cell (WBC), Platelet (PLT) and Red Blood Cell (RBC) count and other parametrs were performed using a Hematology System (Abacus C, Austria). Eight animals were used for each experiment.

Survival studies: For survival studies two groups of animals were selected. A total of 100 mice were used for this experiment. Control irradiated group was only exposed different gamma irradiation, other groups was injected WR-2721 at dose 400 mg kg^{-1} before γ-irradiation. Survival was monitored on a daily basis and the number of animals 30 days post irradiation was recorded. The $LD_{50/30}$ and 95% confidence limits were determined from Probit curve fitting of the 30-day mortality data and were fitted to Probit curves. Each Dose Reduction Factor (DRF) was computed as the ratio of the radiation $LD_{50/30}$ value with the radioprotector to that without a radioprotector.

Statistical analysis: The LD_{50} values were determined using the Probit statistical analysis. The significance of the difference between the mean values obtained for hematological study end points was calculated using the student's t test. The values were considered to be significantly different if $p<0.05$.

RESULTS AND DISCUSSION

Administration of ^{32}P at dose 4 μCi g^{-1} significantly reduced WBC, RBC, PLT and Hct in mice compared to control group ($p<0.05$). These results showed which ^{32}P to have suppressive effects on peripheral blood cells (Table 1). The animals treated with WR-2721 at dose 400 mg kg^{-1} before ^{32}P treatment did not enhance total RBC, WBC counts, Hb concentration and Hct percentage.

Table 1: Variation in hematological parameters of mice treated with WR-2721 prior to ^{32}P treatment

	Groups		
Parameters	Control	^{32}P	WR+^{32}P
WBC (×10^3 μL^{-1})	5.8±1.35	3.9±2.1	2.40±0.92
RBC (×10^6 μL^{-1})	8.9±0.15	8±0.7	7.56±0.5
Hb (g dL^{-1})	14±0.32	13±1	13.2±0.9
Hct (g dL^{-1})	44±1.38	42±1.4	42.7±2.1
MCHC (g dL^{-1})	32±0.75	31±1.5	31.1±0.86
PLT (×1000 μL^{-1})	946±115	774±131	874±88

RBC = Red Blood Cell, WBC = White Blood Cell, Hb = Haemoglobin, Hc = Haematocrit, MCHC = mean cell haemoglobin concentration, PLT = Platelet

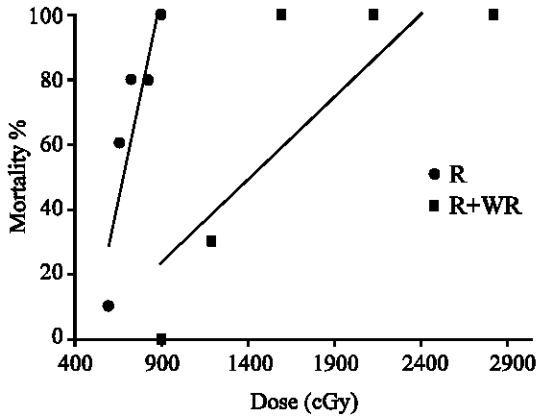

Fig. 1: Survival enhancing effect of WR-2721(WR) in mice that received gamma irradiation (R). Mice were administrated i.p with WR-2721 (400 mg kg^{-1}) or vehicle (normal saline) at 30 min prior to irradiation and exposed to various whole-body doses of radiation. The dose-response curves were fitted by Probit analysis

Although PLT value found to be increased in animals treated with WR-2721 compared to alone ^{32}P treatment but is not significant statistically. These results show which amifostine cannot induce any protective effects on mice treated with phosphorous radioactive. Treatment with amifostine failed to influence the development of peripheral blood cells. For survival study, mice treated with amifostine at 30 min prior whole body gamma irradiation. We found that the radiation dose for $LD_{50/30}$ for control irradiated mice to be 676 cGy. The $LD_{50/30}$ value for amifostine treated mice was 1294 cGy (Fig. 1). The Dose Reduce Factor (DRF) was calculated from the radiation-survival curve generated for this experiment to be 1.91. Administration of amifostine to mice before 9 Gy gamma irradiation to have a 100% survival compared to 0% survival in alone irradiated animals. These results showed which amifostine significantly enhanced survival against whole body gamma irradiation.

While the use of radioprotectors particularly amifostine in ^{32}P therapy is attractive, there is not experiment to show the effect of this drug. After administration of ^{32}P, it stayed more three weeks in the body and continuously emitted beta rays to organs[4-6]. In this study, DRF was 1.91 for amifostine using single optimal dose of radioprotector (400 mg kg^{-1}) against different total body gamma irradiation. However, continuously protection was not maintained since WR-2721 did not lead to enhance peripheral blood cell. Similarly, Lee *et al.*[7] failed to observe an effect WR-2721 given every 24 h on lethality from ^{90}Y-labeled antibody. Badger *et al.*[8] showed administration of WR-2721 at dose 200 mg kg^{-1} prior to ^{131}I-labeled antibody did not have any protection. Treatment with WR-2721 failed to prolong survival or delay myelosuppression from the ^{131}I-labeled antibody. ^{131}I-labeled antibody emits continuously gamma and beta rays for several days. Although administration of WR-2721 provided partial protection from a single treatment of 10 Gy total body gamma irradiation.

The duration of the protective effect of radioprotector is short (half-time 3-4 h for WR-2721) and there is substantial toxicity[9,10]. Other hand repetitive doses of WR-2721 increase toxicity[11]. Achievement of significant protection using amifostine in radioimmunotherapy to be unlikely because continues protection over a period of hours to day is difficult to achieve with tolerated dosed of drugs and presumably against low-dose-rate radiation is small[8].

Although amifostine had a good protection using a single dose against a single TBI, but single administration of amifostine has not any protection against continuous irradiation due to it's short half- life.

REFERENCES

1. Brown, D.Q., J.W. Pittock and J.S. Rubinstein,1982. Early results of the screening program for radioprotectors. Intl. J. Radiat. Oncol. Biol. Phys., 8: 565-570.
2. Hosseinimehr, S.J., A. Shafiee, H. Mozdarani, S. Akhlagpour and M. Froughizadeh, 2002. Radioprotective effects of 2-imino-3{(chromone-2-yl) carbonyl} thiazolidine against gamma irradiation in mice. J. Radiat. Res., 43: 293-300.
3. Anne, P.R. and W.J. Curran, 2002. A phase II prial of subcutaneous amifostine and radiation therapy in ppatients with head and neck cancer. Semin. Radiat. Oncol. 12: 18-19.
4. Spiers, F.W., A.H. Beddode, S.D. King and C.J. Hayter, 1976. The absorbed dose to bone marrow in the treatment of polycythaemia by ^{32}P. Br. J. Radiol., 49: 133-140.
5. Leichner, P.K., N. Rosenshein, A. Beible and S.E. Order, 1980. Distribution and tissue dose of intraperitoneally administered radioactive chromic phosphate (^{32}P) in New Zealand white rabbits. Radiology, 134: 729-734.
6. Silberstein, E., 2005. Teletherapy and radiopharmaceutical therapy of painful bone metastases. Semin. Nucl. Med. 35: 152-1583

7. Lee, Y.C.C., L.C. Washburn, T.T. Sun and B.L. Byrd, 1990. Radioimmunotherapy of human colorectal carcinoma xenografts using ^{90}Y-labeled monoclonal antibody CO17-1A prepared by two bifunctional chelated techniques. Cancer Res., 50: 4546-4551.
8. Badger, C.C., J. Rasey, C. Nourigat, D. Fisher and T. Edmond, 1991. WR-2721 protection of bone marrow in^{131}I -labeled antibody therapy. Radiat. Res., 128: 320-324.
9. Juhas, J.M., J.M. Spellman and F. Culo, 1980. The Role of WR-2721 in radiotherapy and or chemotherapy. Cancer Clin. Trail., 3: 211-216.
10. Srinivasan, V., J.A. Pendergrass, K.S. Kumar and M.R. Landauer, 2002. Radioprotection, pharmacokinetic and behavioural studies in mouse implanted with biodegradable drug (amifostine) Pellets. Intl. J. Radiat. Biol., 73: 536-543.
11. Travis, E.L., M. Fanf and I. Bsie, 1988. Protection of mouse bone marrow by WR-2721 after fractionated irradiation. Intl. J. Radiat. Oncol. Biol. Phys., 15: 377-382.