Radiation Effect on Body Weight and Hematological Changes of Hybrid Mice by Conventional Fraction, Large Abdominal Field Irradiation*

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Radiation effect on mammals, especially on hematologic changes, has been studied since discovery of x-ray. Various experimental animals were tried for radiobiological studies. 72 hybrid mice with conventional ffaction (5X/week), large abdominal field (2X3cm, from symphysis pubic to xyphoid process) were used. Body weight was declined gradually by increasing irradiation doses, nadir was about 29.7% in male; 30.4% in female at 6000 rad irradiation group. Hemoglobin value was nearly normal throughout entire treatment. Significant dropping of WBC count was noted to 40-50% of pretreatment values by only 1000 rad irradiation. Change of differential count was interesting; lymphocyte proportion showed gradual reduction, instead of gradual increasing of segmented neutrophil. Those proportion were reversed after 6000 rad irradiation. Urinary protein tests showed + − +++, showing no correlation with dosage. Application. of our study in clinical combination therapy (radiation + chemotherapy) was discussed.

Key Words: Radiation effect, Hematologic changes, Large abdominal field.

INTRODUCTION

Radiation effect on living mammals has been studied sporadically since Roentgen discovered x-ray. Massive radiobiological researches had been tried since World War II, 1st trial of Atomic Bomb on Hiroshima and Nagasaki. Among the series of small animal experiments, radiation response on hematologic changes are still interested and studied until present time.[1-5] The radiation effect on body weight, hematological changes and urinalysis in large abdominal field were studied since abdominal field is the most radiosenstivie portion of the body.[2] Damage and recovery studies are also examined for clinical application of our data, especially for combination therapy.

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MATERIAL AND METHOD

Animals were 25±2gm in male, 23±1gm in female, 30±3 days old, 72 hybrid mice which were based on our preliminary experiment. Conventional fractionation (200 rad × 5/week), 2×3cm field (whole abdomen, from symphysis pubis to xyphoid process) were used from Monday to Friday. Remainder of the body were shielded with 4 mm lead. Used machine was 250 KV, 25 mA. (Coronado, Westinghouse, England), and calibration was confirmed every Saturday. During treatment, all mice were placed in a specially designed wooden boxes with limited mobility. All mice were given slight Ether anesthesia. Mice to be irradiated were devided into 10 groups from 1000 rad group to 10000 rad group, 6 mice for each group (3 males and 3 females). Every Friday, after radiation body weight were scaled and Hb, WBC, differential counts were examined by puncture of tail vein. Urinalysis was performed

simultaneously. Stool examination for occult blood were studied for further evaluation.

RESULT

1. Weight; Pretreatment mean was 26.5 gm in male, 22.8gm in female. All post-treatment mean were decreasing with increment of exposure dose. Up to 6000 rad, body weight was decreasing gradually inspite aging. Maximum reduction was 6000 rad group, 29.7% in male, 30.4% in female, of normal growth value at same age respectively. Above 6000 rads, body weight of experimental animals reached to plateau with some fluctuation. Those data suggested that susceptibility of the mice to reduction of weight may be influenced by irradiation especially in moderate doses. (Table 1, Fig. 1).

Table 1. Body Weight Change at Different Dose & Age

Date	Dose (rad)	Weight ♂ (gm) Mean	S.D.	Weight ♀ (gm) Mean	S.D.
4/ 8	pretreatment	26.5	±2.65	22.8	±2.59
4/12	1000	26.1	±3.19	22.3	±2.25
4/19	2000	25.7	±3.42	22.2	±2.48
4/26	3000	23.9	±3.84	21.5	±2.44
5/ 3	4000	22.8	±3.51	20.2	±2.46
5/10	5000	22.8	±3.47	19.6	±1.93
5/17	6000	22.5	±3.40	19.4	±2.16
5/24	7000	23.3	±3.39	19.9	±1.78
5/31	8000	23.8	±3.94	19.9	±1.61
5/ 7	9000	23.7	±4.47	21.9	±2.41
6/14	10000	22.0	±1.78	21.3	±2.25

Table 2. Hb. Change at Different Dose

Dose (rad)	Hb (♂) Mean	S.D.	Hb (♀) Mean	S.D.
pretreatment	15.1	±1.72	15.5	±0.93
1000	15.0	±2.04	15.4	±1.60
2000	15.1	±2.04	15.4	±1.38
3000	15.3	±2.20	15.6	±1.10
4000	14.9	±2.20	15.5	±1.70
5000	14.8	±2.20	15.0	±1.10
6000	14.2	±1.90	14.7	±1.20
7000	14.0	±2.60	14.6	±1.40
8000	14.0	±3.70	14.2	±1.20
9000	14.4	±2.40	14.6	±1.70
10000	14.7	±2.30	15.3	±1.40

2. Hb: Pretreatment values were 15.1 gm% in male, 15.5 gm% in female. Nadir of treatment group was 8000 rad group in both, male and female. (14.0 in male, 14.2 in female). Throughout all group of irradiation, Hb. value shwed no significant difference in dose increment and sex. (Table 2, Fig. 2).

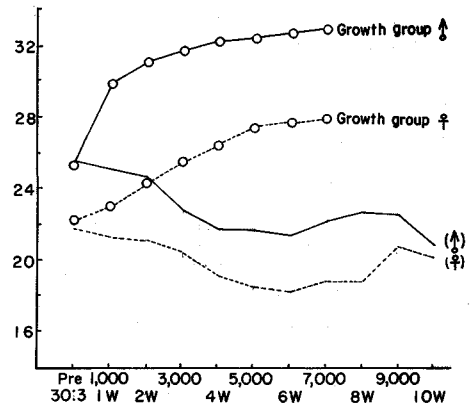

Fig. 1. Body wt. change at different dose & age.

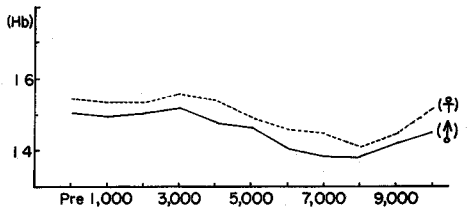

Fig. 2. Hb. change at different dose

Table 3. WBC Change at Different Dose

Dose (rad)	WBC ♂ (×10³) Mean	S.D.	WBC ♀ (×10³) Mean	S.D.
pretreatment	9,16	±2.62	8.44	±2.22
1000	4.47	±1.98	5.02	±1.37
2000	4.09	±1.22	3.52	±1.31
3000	4.51	±1.29	4.29	+1.33
4000	4.20	±1.58	4.11	±0.81
5000	3.53	±0.97	3.77	±1.15
6000	4.28	±1.02	4.76	±1.24
7000	3.80	±1.16	3.93	±1.14
8000	3.38	±1.02	3.12	±0.71
9000	3.22	±1.01	3.00	±0.86
10000	3.20	±1.01	3.00	±1.00

3. WBC: Pretreatment normal mean were 9160 in male, 8440 in female. WBC count after 1000 rad irradiation showed marked reduction to 4470 in male (48.8% of pretreatment value), 5020 in female (59.9% of pretreatment value). Thereafter, the value showed gradual reduction with mild fluctuation and reached to 3200 (34.9%) in male, 3100 (36.7% of pretreatment value) in female at 10000 rad irradiation. (Table 3, Fig. 3).
4. Differential count: In pretreatment normal group, mean lymphocyte was 71% (male), 73% (female), segmented neutrophil was 22% (male), 19% (female), degenerates was 6.3% (male), 6.7% (female).
   The proportion of lymphocytes was reduced progressively but segmented neutrophil showed gradual increasing with dose increment. Those proportion were reversed at 6000 rad irradiation level. At 10000 rad irradiation, the differential count showed lymphocyte: 40% in male, 36% in female, segmented neutrophil: 53% in male, 55% in female, and the proportion of degenerates observed no significant changes throughout entire treatment. (Table 4, Fig. 4).
5. Urinalysis: In pretreatment examination, urine protein was + − +++. There were various changes even in repeated tests of same animal, but those datas showed no significant changes by radiation doses.

DISCUSSION

In the practice of radiation therapy, large abdominal field is troublesome by nausea, vomiting and other symptoms due to various organ damages. Bond et al.[2] demonstrated LD 50 was 1950 r. in air for rats with abdomen shielded, and 1025r. for those receiving only abdominal exposure. He also reported that peak death was 9-12 days of exposure for abdomen shielded group, 4-5 days for abdomen only exposure group in his experiment. He observed animals daily over a period of 28 days after irradia-

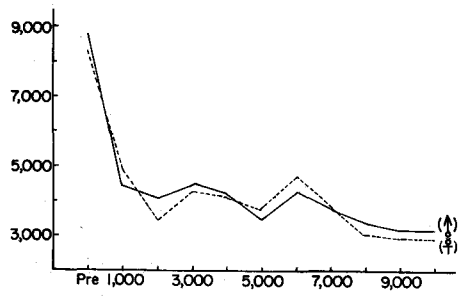

Fig. 3. WBC change at different dose

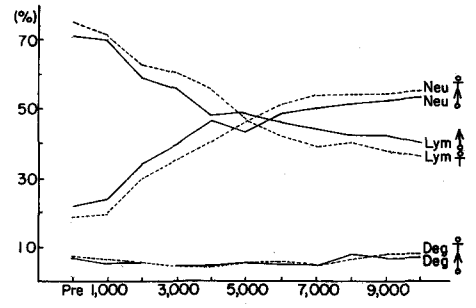

Fig. 4 Differential count change at different dose

Table 4. Differential Count Change at Different Dose

Dose (rad)	Diff. ♂ (%)			Diff. ♀ (%)		
	Lym.	Neu.	Deg.	Lym.	Neu.	Deg.
pretreatment	71± 7.0	22± 6.2	6.31	73± 5.4	19± 3.5	6.72
1000	70± 6.2	24± 5.2	5.41	71± 6.0	20± 4.6	6.33
2000	59±12.3	34±13.4	5.79	62±11.0	30±10.0	6.12
3000	56±10.4	39±10.0	4.62	60± 8.8	35± 8.4	4.96
4000	48±15.3	46±15.2	4.67	55±13.4	40±12.7	4.20
5000	49± 8.6	43± 7.8	5.50	47±14.9	46±13.6	5.76
6000	46±11.3	48±11.8	5.15	42±15.4	51±15.0	6.00
7000	44±12.4	50±12.7	5.00	39±10.4	54± 9.5	5.00
8000	42±10.0	51±10.9	8.25	40±11.8	54±14.4	6.88
9000	42±10.8	52±11.2	7.00	38±11.0	54±12.4	7.83
10000	42±10.4	53±10.4	7.67	36±10.6	55±12.0	8.00

tion for clinical symptom and mortality. He found the mortality was 33% at 2000 rad in abdomen shielded, 55% at 1000 rad in abdomen exposure only. He concluded that abdomen was the most radiosensitive portion of the body and resulting syndrome were a function of the region irradiated rather than of the total energy received. He also insisted that intestine was the largest contributory factor in producing the syndrome of "radiation sickness" even though anorexia, nausea and vomiting can be obtained in the absence of gut irradiation in therapeutic irradiation of human.

Abrams et al.[7] observed 30 days after irradiation and found that highest mortality was 30 day old group and the mortality decreased progressively with age thereafter. He concluded that susceptibility of mice to the lethal effects of irradiation was strikingly influenced by age at the time of exposure and selection of proper age was one of the most important factor in experiment, at least with mice.

Melamed et al.[2] demonstrated that WBC counts reached to 40% of control at 10 days after irradiation of 20-50 rad, fractionated, whole body, low dose irradiation. He reported hematocrit and hemoglobin value remained near the range of control, in spite of consistently depressed bone marrow cells throughout 19 weeks after irradiation in his experiment (Normal life span in mice is 40.7 ± 1.9 days).[9]

Einhorn[9] suggested that the degree of anemia in malignancy is significantly correlated to the serum folate clearance rate, the serum iron and ESR, This suggested that another possibility for the cause of secondary anemia following radiation therapy.

In WBC study, Jankins et al.[1] reported mean lymphocyte mumbers decreased to 31% of the pretreatment value during therapy and whole blood cultures decreased to 15 to 39% of pretreatment value.

Blogren et al.[3] insisted that the peripheral blood lymphocyte population after radiation therapy was reduced to 30-50% of pretreatment value in his experiment.

Our data showed extensive reduction of the weight compared to normal growth group. This suggested that the reason may be mice factor which we selected in our series, i.g., 30 ± 3 days old, the most radiosensitive age.

No significant change of Hb. in our experiment supported Melamed et al.[2]. No difference between our data and Jenkins' observed in WBC study which showed 40-50% of normal value at 1000 rad level and 34.9% (male), 36.7% (female) at 10000 rad irradiation. In differential count studies, the proportion of segmented neutrophil observed progressive increasing with increment of total dose while the proportion of lymphocyte was decreased gradually. Those data supported Storer[11] who insisted that granulocytes were slightly more resistant but also showed a marked drop after moderate dose. He also insisted that initial granulocytosis preceding the decline represented an accelerated release from marrow.

Our data is interesting observation for clinical extrapolation of combination therapy, especially chemotherapy and radiation therapy, since hematologic change during treatment is important factor which can modify the treatment course and the results.

CONCLUSTION

Our results can be summerized as follows:

1. Body weight was decreased progressively with increment of radiation dose, the nadir was 6000 rad irradiated group.
2. No significant variation in Hb. value observed throughout all experiment, although nadir was 8000 rad group.
3. WBC count showed extensive reduction even at 1000 rad irradiation group to approximately 40-50% of pretreatment mean value.
4. Proportion of lymphocyte was reduced progressively with dose increment while the proportion of the segmented neutrophil was increased gradually. Those proportion were reversed from 6000 rad irradiation group.
5. Urinalysis showed various urine protein value (+ − +++) throughout all experiment.

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=국문초록=

고식적 분할조사시 전복부 조사량에 따른 잡종 백서의 체중과 혈액상의 변화에 관한 연구

영남대학교 의과대학 치료방사선과학교실

이 성 헌 · 신 세 원 · 김 명 세

X-선의 생물체에 대한 효과는 뢴트겐이 X-선을 발견한 이래 산발적으로 보고되어 왔으나 원폭의 인류사용을 계기로 하여 급격히 진행되어 왔다.

여러가지 동물을 사용하여 수많은 연구가 계속되어 왔으나 그중에서도 혈액상에 관한 변화는 근래에까지도 계속되고 있다. 저자들은 잡종 백서를 사용하여 방사선에 가장 감수성이 예민한 부위인 전복부에 고식적인 분할조사를 실시하여 체중과 혈액상의 변화를 분석하였고 이 결과를 토대로 하여 화학요법등과의 병합 치료에서 혈액상의 변화를 비교할 수 있는 기본자료로 이용하고저 한다.