

The Effect of Total Dose of Radiation on Normal Colon of Hybrid Mice

Gil Ho Jho, M.D.* and Myung Se Kim, M.D.

Department of Diagnostic Radiology, Department of Therapeutic Radiology
College of Medicine, Yeungnam University*

Optimal balance between control probability and risk of complication is emphasized even in present time. Although certain incidence of intestinal injury is accepted as an inevitable consequence after abdominopelvic irradiation, these complications still remain as problems.

60 mice were irradiated with 250 kVp orthovoltage x-ray machine and 200 rad \times 5/wk regimen. Histopathologic findings of colorectum and the relationship with occult blood test were analyzed and possible tolerable dose which would be safe from permanent complication was also estimated.

Followings are the results:

Mild mucosal and submucosal edema were observed in 1,000 rad irradiated group.

Congestion of small vessels was prominent in 2,000 rad irradiated group and infiltration of inflammatory cells was observed in 3,000 rad irradiated group.

Denuded mucosa was observed in 3,000 rad irradiated group.

Occult blood test is not a proper indicator for rectal denuding or rectal ulcer, but our results suggest the possibility of using this as a relative scale of intestinal damage.

Mitotic figures of crypt cells were observed even in 5,000 rad irradiated group, these suggest that the repair capacity of crypt cells are still functioning.

Key Words: Radiation colitis, Radiation pathology.

INTRODUCTION

Radiation therapy is a powerful and effective modality for treatment of cancer in present time. But probability of tumor control by irradiation is very small at low dose, rises steeply at moderate doses, and reaches a plateau at high dose. Similar curve is presumed for the probability of complication as a function of dose because administration of sufficient dose to eradicate viable tumor cells may produce damage to normal tissue within treatment field, unfortunately.

Therefore, radiation therapy is necessary to achieve an optimal therapeutic balance between control probability and risk of complication. The study of radiocurability as well as tolerance of normal tissue has continued to the present time¹⁻⁴⁾ to maximize tumor destruction and minimize normal tissue damage.

Many experimental and clinical studies on various factors of intestinal complication have been

published, but systemic study on the effect of total dose, fractionation and dose rate, which is similar to clinical practice, has not been performed so far.

The purposes of this study are:

- 1) to analyze histopathological change of colorectum at various total dose of irradiation,
- 2) to investigate the correlation with the extent of histopathology of colorectum and occult blood test of stool,
- 3) to estimate the maximum tolerable dose of colorectum respectively,
- 4) hopefully those data could be used as the basic data in clinical practice.

MATERIAL AND METHOD

Used animals were 60 mice (30 male, 30 female) derived from "brother-sister or offspring-young parents" mating. Selected animals were 30 ± 3 days old, weighing 25 ± 2 gm (δ), 23 ± 2 gm (φ), which were based on previous experiment in radiobiology laboratory of Yeungnam

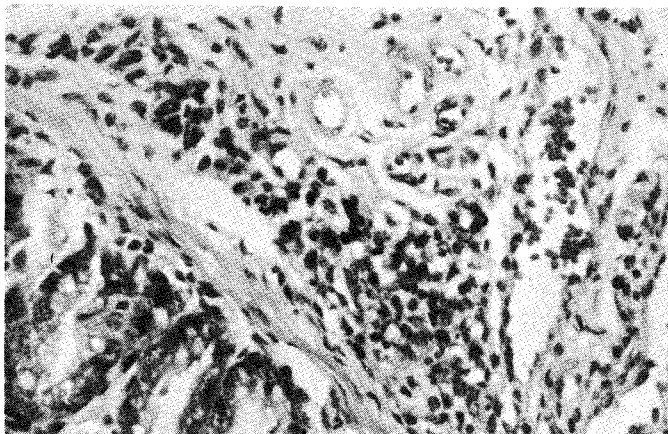


Fig. 1. Slight vascular congestion and dilatation in 1,000rad irradiated group (H&E, X100).

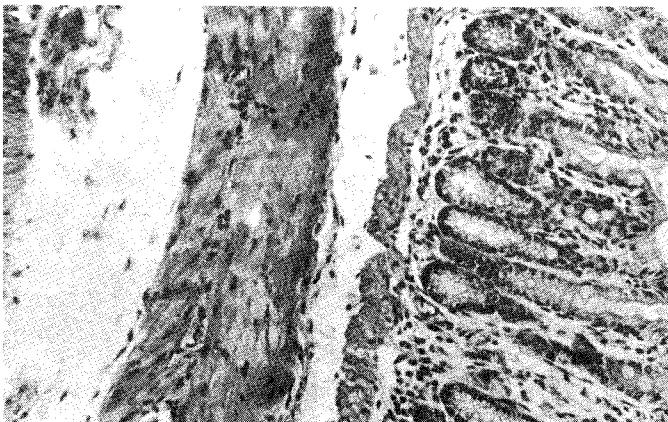


Fig. 2. Increased mucosal and submucosal edema, but intact crypt cells in 2,000rad irradiated group (H&E, X40).

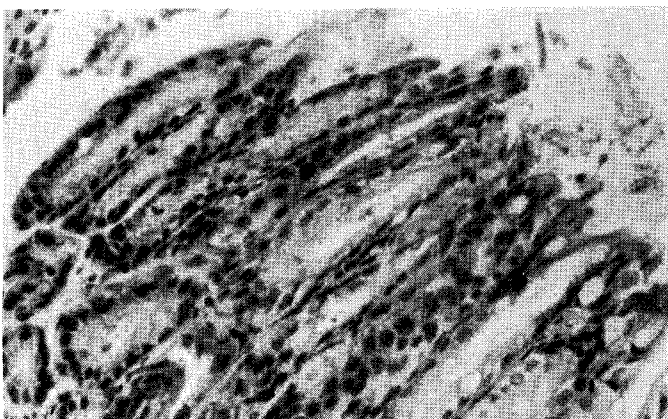


Fig. 3. Denuding of mucosal surface in 3,000rad irradiated group (H&E, X200).

were evident. Submucosal edema and capillary congestion were more prominent than 1,000 rad group (Fig. 2). Occult blood test showed (+) in 93 % of male, in 71% of female, (++) in 29% of female.

In 3,000 rad irradiated group, marked mitotic figures of crypt cells and denuding mucosa (Fig. 3), prominent capillary congestion and infiltration of inflammatory cells were observed. Occult blood test showed (+) in 56% of male, in 58% of female, (++) in 38% of male, in 25% of female, (+++) in 6% of male, in 17% of female.

In 4,000 rad irradiated group, slight decreased mitotic figures than 3,000 rad group, but similar degree of capillary congestion and submucosal

edema were observed. Mucosal atrophy with focal denudation of mucosa (Fig. 4), and marked infiltration of inflammatory cells were also observed in some specimens. Occult blood test showed (+) in 22% of female group only, (++) in 56% of male, in 44% of female, (+++) in 44% of male, in 33% of female group.

In 5,000 rad irradiated group, mucosal denuding, capillary congestion and atrophy of mucosa were increased than 4,000 rad irradiated group, but mitoses of crypt cell similar to 4,000 rad irradiated group (Fig. 5). Occult blood test showed (+++) in 57% of male, in 88% of female, (++++) in 43 % of male, in 12% of female group. (Fig. 5)

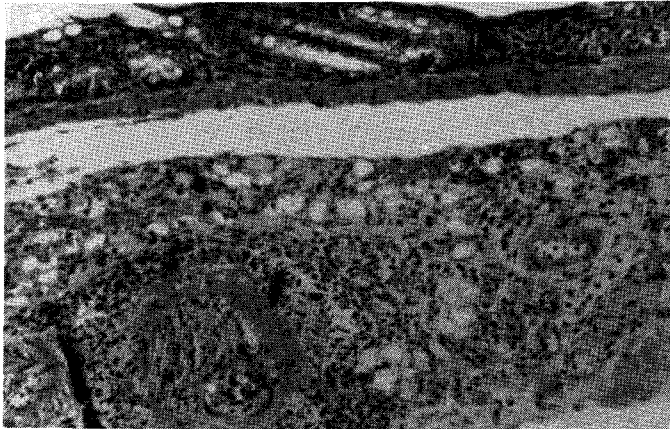


Fig. 4. Moderate mucosal atrophy, vascular congestion and perivascular inflammatory cell infiltration in 4,000rad irradiated group (H&E, X40).

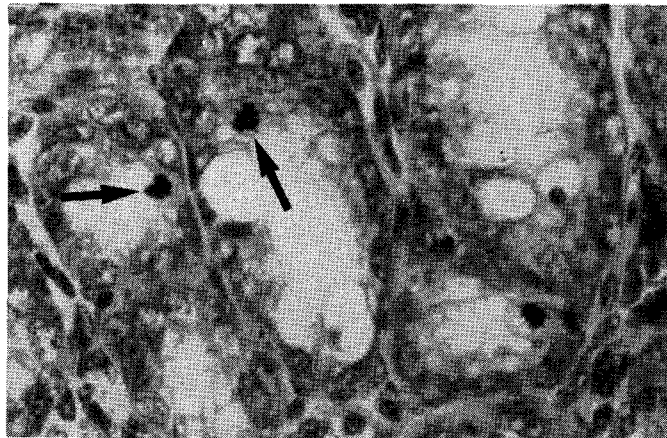


Fig. 5. Many mitotic figures in the mucosal epithelium in 5,000rad irradiated group (H&E, X400).

DISCUSSION

Uterine cervical cancer, bladder cancer, seminoma, renal cell ca, malignant lymphoma and colorectal cancer, which need abdominopelvic irradiation, have been known as curable with radiation therapy with or without surgery and chemotherapy.

Development of supervoltage machine may minimize skin reaction and make it possible to deliver greater tumor dose and incidentally greater tissue dose to the deep seated normal tissue, especially intestine.

Therefore, certain incidence of intestinal injury came to be accepted as an inevitable consequence, particularly after combination of intracavitary and external irradiation for uterine cervical cancer. Especially when the large intestine was fixed by adhesion after previous surgery, PID, or tumor, the problems are serious because these condition induce malposition of the radioactive source so that created deviation of the dose, causing hot spot.

Such GI complications following abdominopelvic irradiation are reported in 0.5%⁷⁾ to 23.9%⁸⁾ of patients.

The influencing factors for those GI complications are:

1) total radiation dose, overall treatment time, number of fraction, size of fraction, quality of radiation, and size of field,^{7,10~13)} 2) number or previous laparotomy and extent of surgery,^{12~14)} 3) hypertension, diabetes, cardiovascular disease,^{8,12~15)} 4) age, sex and thin or obese physique,^{13~15)} 5) stage of disease.^{8,12,13)}

Smith et al⁸⁾ observed mucosal edema, capillary thrombosis, hyalinization of small arteries and infiltration of non-specific inflammatory cells in his autopsy specimens which were irradiated 2,500-4,500 rad on abdomen. Our result showed submucosal edema and capillary congestion in 2,000 rad group, non-specific inflammatory cell infiltration in 3,000 rad irradiated group, these are compatible with the report of Smith et al.

Weisbrolet al¹¹⁾ reported prominent eosinophilia and numerous mitosis in autopsy specimens which were irradiated 1,300-2,500 rad. Withers and Elkind¹⁶⁾ reported that crypt cell of mice has a big capacity for recovery of sublethal injury in his single (2075 rad) and split dose (660+1415 rad) experiments and also insisted that almost all the recover occur within 8 hours after irradiation even

though various differences existed in different species. Our result showed mitosis of crypt cells in 5,000 rad irradiated group, and these suggest big capacity of recovery.

Although mucosal ulcer after irradiation reported even in whole body irradiation of 400 rad only,¹¹⁾ Kieszel et al²⁾ reported severe ulcer on irradiated area 7 days after 1,750 rad, single irradiation. Choi and Suh¹⁾ also reported similar results in 1,800 rad irradiated specimen of mice which was recovered completely in 12 days after irradiation.

Our result showed no significant ulcer even in 5,000 rad irradiated group, but investigation of submucosal edema and mucosal denuding in 3,000 rad irradiated group suggest that 5,000 rad which has been accepted as safe dose for colorectal complication, is not absolutely safe. Confirmation of the relationship between these findings and later chronic complication, such as stenosis or perforation should be demonstrated further experiment.

Rectal bleeding is the most common acute complication after abdominopelvic irradiation, because rectum is the commonest site of injury, even though small intestine and other part of colon may be damaged.⁸⁾ Not infrequently, considerable disparity exist between the severity of clinical manifestation and histopathological findings and radiation injuries of the colon and rectum are most often associated with small amounts of rectal bleeding with or without demonstrable ulceration after 4,500-6,000 rad irradiation.⁷⁾ Decosse et al,¹⁴⁾ Macmahon & Rowe¹⁷⁾ also emphasized that degree of rectal ulceration is not necessarily similar to histopathological finding and these rectal ulceration was seen in 4-12 month after irradiation. Our result showed no significant ulceration except mucosal denuding in 3,000 rad group, but occult blood test showed (+)~(++) in 100% of animals of 2,000 rad irradiated group, (+++)~(++++) in 100% of animals of 5,000 rad irradiated group. These result support that histopathological findings of colon after irradiation is not exactly same as the degree of occult blood test, but the tendency of increasing positive degrees, depending on total dose of radiation, cannot be denied.

Maximum tolerable radiation dose of colorectum has been reported to be 1,750~2,000 rad in single irradiation,^{1,2)} 4,500~5,500 rad in multiple fractionated dose.^{8,11,16)} But our result shows definite mucosal denuding and (+)~(+++) of occult blood test in 3,000 rad irradiated group, these means that we should be careful even below 3,000 rad level, even though the report of Choi and Suh,

¹⁾ which these acute reaction were recovered completely within 12 days especially in large field irradiation.

CONCLUSION

60 hybrid mice (30 male, 30 female) were irradiated with 2×3 cm field and 200 rad×5/wk fraction.

Analysis of histopathological findings at various total dose level are summerized as follows:

1) mucosal and submucosal edema were observed in 1,000 rad irradiated group, but the findings were evident in 2,000 rad irradiated group,

2) capillary congestion was evident in 2,000 rad group, and infiltration of inflammatory cells around vessels were observed in 3,000 rad irradiated group, and were evident in 4,000 rad group,

3) mucosal denuding was observed in 3,000 rad irradiated group,

4) occult blood test showed (+)~(+++) in 100% of animals of 3,000 rad irradiated group, (+++)~(+++++) in 100% of animals of 5,000 rad irradiated group.

These result suggest that occult blood test can not be a exact indicator of intestinal damage. But the tendency of increasing degree with total dose increment was evident, this may suggest the possibility that occult blood test can be used as a the relative scale of intestinal damage.

5) Mucosal denuding was observed in 3,000 rad irradiated group, but numerous mitotic figures were evident even in 5,000 rad irradiated group.

REFERENCES

1. Choi MS, Suh WS: The pathological changes of stomach in experimental rats following single irradiation of supervoltage. *J Korean Soc Ther Radiol.* 2, 25-32, 1984
2. Kieszel Z, Spiethoff A, Trott KR: Large bowel stenosis in rats after fractionated local irradiation. *Radiother Oncol.* 2, 247-254, 1984
3. Von der Masse H: Interactions of radiation & 5-FU, CY or Methotrexate on intestinal crypt cells. *Intl J Radiol Oncol Biol Phys.* 10, 77-86, 1984
4. Huh SJ, Park CI: Radiosensitivity and dose-survival characteristics of crypt cells of mouse jejunum. *J Korean Soc Ther Radiol.* 3, 9-12, 1985
5. Lee SH, Jho KH, Shin, Kim MS: Change of body weight and hematologic value with aging in hybrid mice. *J Korean Soc Ther Radiol.* 3, 169-173, 1985
6. Lee SH, Shin So, Kim MS: Radiation effect on body weight and hematological changes of hybrid mice by conventional fraction, large abdominal field irradiation. *J Korean Soc Ther Radiol.* 3, 153-157, 1985
7. Libschitz HI: Diagnostic roentgenology of radiotherapy change. Waverly Press. Baltimore, pp 85-99, 1979
8. Smith AN, Douglas M, Maclean M: Intestinal complications of pelvic irradiation for gynecologic cancer. *Surg Gynecol Obstet.* 127, 721-728, 1968
9. Logalio SA, Stone A, Friedman M: Surgical aspects of radiation enteritis. *Surg Gynecol Obstet.* 129: 1163-1172, 1969
10. Withers HR: The response of stem cells of intestinal mucosa to irradiation with 14 MeV Neutron. *Br. J. Radiol.* 43, 796-801, 1970
11. Weisbrot IM, Liber AF, Gordon BS: The effect of therapeutic radiation on colon mucosa. *Cancer.* 36, 931-940, 1975
12. Potish RA, Jones TK, Levitt S: Factors predisposing to radiation related small bowel damage. *Radiology.* 132, 479-482, 1979
13. Potish RA: Prediction of radiation related small bowel damage. *Radiology.* 135, 219-221, 1980
14. De Cosse JJ, Rhodes RS, Wentz WB: Nature of history and management of radiation induced injury of the G-I tract. *Ann Surg.* 170, 369-383, 1969
15. Maruyam Y, van Nagell JR, Utley J: Radiation and small bowel complications in cervical ca. treatment. *Radiology.* 112:699-703, 1974
16. Withers HR, Elkirnd MM: Radiosensitivity and fractionation response of crypt cells of mouse jejeunum. *Radiat Res.* 38, 598-613, 1969
17. MacMahon CE, Rowe JW: Rectal reaction following radiation therapy of cervical carcinoma. *Ann Surg.* 173, 264-269, 1971

국문초록 =

치료방사선 조사가 잠종 백색마우스의 대장에 미치는 병리조직학적 영향

영남대학교 의과대학 진단방사선학교실

조 길 호

치 료 방 사 선 학 교 실

김 명 세

암의 방사선 치료는 단독, 혹은 수술이나 화학요법과의 병합치료를 통하여 좋은 생존율을 보여주고 있으나 정상 조직의 손상으로 인한 후유증은 아직도 해결되지 못하고 있는 문제중의 하나이다. 그러므로 정상조직의 손상에 대한 연구는 암조직의 방사선에 대한 연구와 함께 필수적이며, 근래에 계속되고는 있으나 실제 임상에서 사용되고 있는 방법과 같은 분할조사에 대한 계통적인 연구는 매우 드물다. 이에 저자는 60마리의 백색 마우스를 사용하여 2×3 cm의 조사야로 고식적인 방법으로 분할 조사한후 전 조사량에 따른 병리조직학적인 변화를 분석하고 잠혈반응 검사와의 상관관계를 규명하여, 직장에 심한 후유증이 오지 않을 수 있는 가능한 최대 내선량을 추정하여 임상치료에 이용할 수있는 기본 자료로 삼고자 하였으며 그 결과는 다음과 같다.

점막 및 점막하 부종은 1,000 rad군에서부터 관찰되었다.

소혈관 울혈은 2,000 rad군에서 현저하였고 염증세포의 침윤은 3,000 rad군에서 현저하였다.

점막의 탈락은 3,000 rad군에서부터 관찰되기 시작하였다.

잠혈반응은 점막탈락, 혹은 괴양의 정확한 척도로 삼기에는 부적당 하였으나 총 조사량의 증가와 함께 잠혈반응의 양성도도 증가하는 추세를 보여 간접적인 지표로 삼을 수는 있으리라고 생각되었다.

소낭선세포의 분열상이 5,000 rad군에서도 관찰되는 것으로 미루어 5,000 rad의 조사에서도 재생능력이 남아 있음을 시사하였다.