The Effect of Radiation Therapy on the Healing Ability of Subsequent Surgical Wounds

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This study was undertaken to find the effect of radiation therapy on the healing ability of surgical wounds and on this basis, to find the proper time interval between the radiation therapy and surgery. Two hundred and fifty-two mice were used and a single dose of 2000 cGy was given in each instances to the hind limb of mice. Incisional wounds were produced after varying intervals in the previously irradiated areas and then they were followed up at regular intervals by the measurement of tensile strength.

The wounds which received surgery immediately, 1 or 2 weeks after irradiation revealed marked delay and the wounds which received surgery 12, 16 or 20 weeks after irradiation demonstrated slight delay in wound healing in terms of tensile strength measurement. But the wounds which received irradiation 4 or 8 weeks before surgery did not differ much in the wound healing process from that of the control group.

Histopathologic studies of the wounds demonstrated epithelization in most instances as quickly as in the control wounds. The appearance of fibroblasts and collagen fibers has delayed momently and appeared to have close correlation with the tensile strength healing curves.

Key Words: Preoperative radiotherapy, Wound healing, Tensile strength

INTRODUCTION

Radiation therapy plays an important role in the treatment of cancer. Seventy to eighty percent of cancer patients receive radiation therapy during the local course of their illness. The local control rate of curative purpose of radiotherpy reaches about 70% at present time[1].

In the locally advanced cancer, a combined irradiation and surgical resection increases greatly the local control of cancer[2,3]. But some of the earliest observations suggest that radiation delievered either before or after wounding the skin would interfere with reconstrucion of the structures and reepithelization[4]. Especially, the preoperative irradiation is known to affect the healing of the surgical wounds[5]. Therefore it is of great importance to study the effect of preoperative irradiation on the healing ability of a subsequent surgical wounds.

This study was undertaken to assess the healing ability of surgically incised wounds on previously irradiated skin with clinically significant radiation dose and to find the proper time interval for solid wound healing between radiation therapy and surgery.

MATERIALS AND METHODS

Two hundred fifty-two ICR mice, both sexes, weighing about 23±2 g, regardless of sexes were used in this experiment. These were subdivided into 8 groups including 1 control group and 7 test groups (Table 1).

The mice were anesthesized by ether inhalation and fixed to the wooden board on supine position. Radiation field size of $2\times2\,cm^2$ was marked with ink on the skin surface of the both ventral aspects of hind limb (Fig. 1). Irradiation was delivered with dose of 2000 cGy in single fraction from 4MV linear accelerator. 1.1 cm thick bolus material was applied to the skin to maximize the radiation dose to the skin surface.

Immediate, 1, 2, 4, 8, 12, 16 and 20 weeks after irradiation, skin was incised with surgical mess after careful preparation with 70% ethyl alcohol. The incised wound was approximated with silk suture material (Fig. 2). Bleeding was controlled by

Table 1. Experimental Animal Groups

Postincisional days / Postirradiation weeks	2	4	6	8	10	14	21	
Control	4	4	4	4	4	4	4	28
Immediate	4	4	4	4	4	4	4	28
1	4	4	4	4	4	4	4	28
2	4	4	4	4	4	4	4	28
4	4	4	4	4	4	4	4	28
8	4	4	4	4	4	4	4	28
10	4	4	4	4	4	4	4	28
12	4	4	4	4	4	4	4	28
16	4	4	4	4	4	4	4	28
20	4	4	4	4	4	4	4	28
Total	36	36	36	36	36	36	36	252

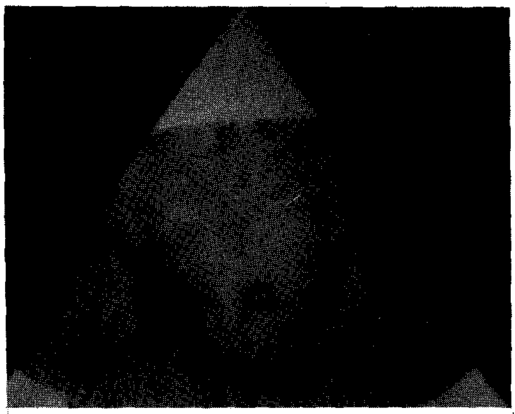

Fig. 1. Experimental mouse with radiation field marked on the hind limb.

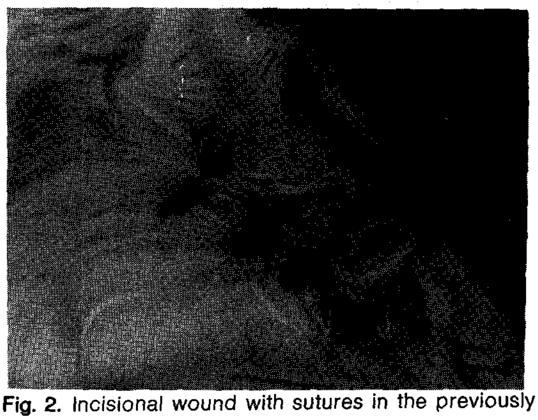

Fig. 2. Incisional wound with sutures in the previously irradiated field.

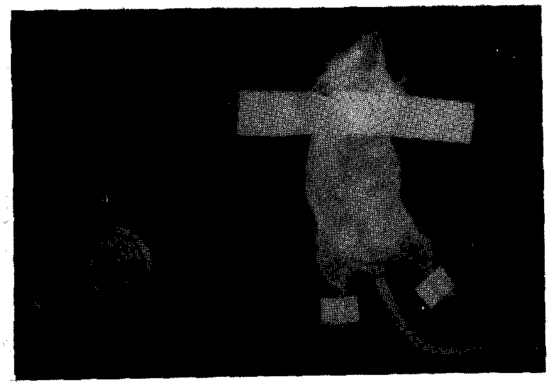

Fig. 3. For the measurement of tensile strength, skin along the wound was sutured and connected to the pulley which carried 20.4 g weight of container.

compression and the wound was coated with antibiotic solution to prevent the infection. Control group did not receive the irradiation, but the similar surgical incision was performed.

Immediate, 2, 4, 6, 8, 10, 14, 21 days after incision, 4 animals from each subgroups were randomly chosen and suture materials were carefully removed. Then the healing process was assessed by measurement of tensile strength, the method which was first described by Howes in 1929[6]. Tensile strength is defined as breaking strength per unit cross sectional areas. Since the incised areas were same in each experimental subgroups, the tensile strength could be expressed in grams (g). Only the skin layer was sutured with #3 silk suture material at 5 mm away from the incisional scar. The one side of suture string was fixed to the wooden board and the other side was connected to the pulley which carried 20.4 g weight of container (Fig. 3). The distilled water was dropped into the container with the speed of 20 cc per minute. It was read as tensile strength at the point when the skin began to be separated by the thread tester (Fig. 4). Tensile strength curves of the healing wounds in each group were constructed using the average strength measurements at varying time intervals.

Fig. 4. Tensile strength measurement at the point when the skin began to be separated by the thread tester.

At the same time, an animal was randomly sacrificed from each subgroup to obtain tissue sections for histopathologic studies.

RESULTS

1. Tensile Strength

The average values of tensile strength of the surgical wounds for both control and irradiated groups were recorded in terms of various time intervals following surgery (Table 2). The curves were constructed with average values of measured tensile strength at varying time intervals (Fig. 5-8).

Control group revealed 147 g at 2nd day of surgery and showed a rapid rise until the 10th day

Table 2. Averages and t-Values of Tensile Strength

Post surg. days / Post irrad. wks.	2	4	6	8	10	14	21
Control	147	182.9	384.4	592.5	769.4	515.7	543.2
Immediate	94.2 (2.071) *	115.9 (1.487)	179.4 (3.079) *	202.4 (2.955) *	244.4 (4.61) *	270.1 (5.732) *	349.4 (3.915) *
1	108.0 (1.595)	232.9 (−0.931)	234.1 (1.7)	251.1 (2.511) *	226.4 (4.836) *	201.8 (6.468) *	308.4 (0.073) *
2	50.6 (4.295) *	85.2 (1.843) *	115.8 (2.713) *	142.0 (4.05) *	196.9 (6.294) *	349.2 (3.181) *	553.9 (−0.102)
4	125.2 (0.77)	191.0 (−3.345) *	282.0 (1.086)	499.4 (−0.593)	319.9 (3.953) *	283.1 (2.795) *	357.2 (1.882)
8	118.8 (0.7)	160.3 (0.311)	335.8 (1.086)	357.4 (2.14) *	257.4 (2.536)	323.4 (1.959)	320.4 (1.662) *
12	66.9 (1.584) *	76.2 (1.179)	93.7 (3.556) *	162.4 (2.782) *	167.4 (2.972) *	252.7 (2.455) *	247.0 (1.712) *
16	86.2 (1.442)	50.3 (2.111) *	68.4 (3.059) *	129.2 (3.11) *	244.4 (2.586) *	312.0 (2.575) *	270.0 (1.554) *
20	43.6 (1.968) *	77.7 (2.187) *	88.3 (2.841) *	144.9 (4.203) *	240.5 (2.522) *	488.4 (0.166)	373.1 (1.2) *

* t-values which are signigicant.

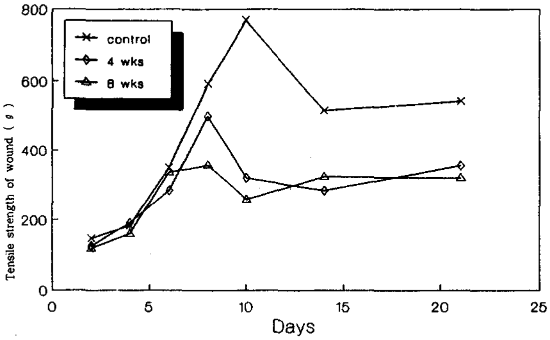

Fig. 5. Changes of tensile strength of control, 0, 1, and 2 wks postirradiation group.

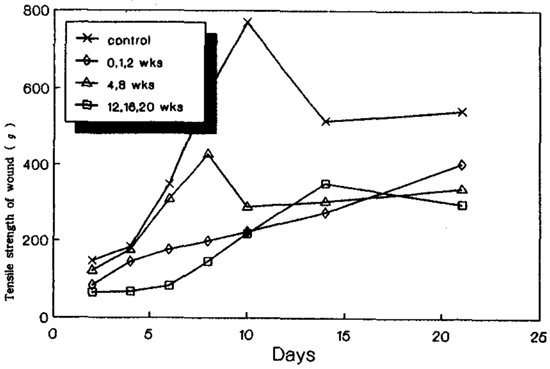

Fig. 6. Changes of tensile strength of control, 4, and 8 wks postirradiation group.

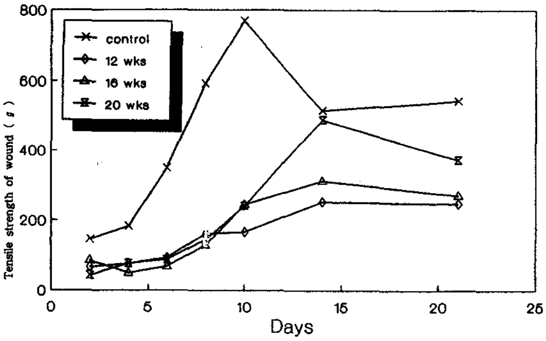

Fig. 7. Changes of tensile strength of control, 12, 16, and 20 wks postirradiation group.

with a value of 769.4 g. Afterwards it rather tended to be decreased. For immediate surgery group, tensile strength rose slowly up until the end of observation period and did not show peak values. For one week group, tensile strength rapidly rose from 2nd day, but it did not show a peak values

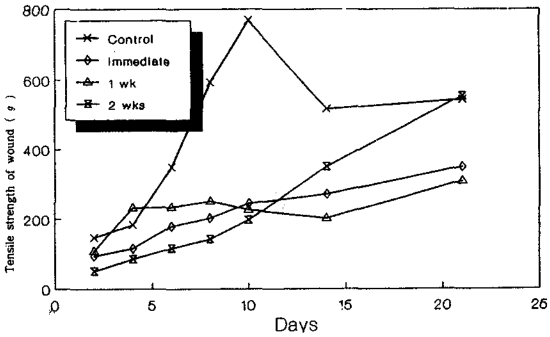

Fig. 8. Changes of average tensile strength of each groups.

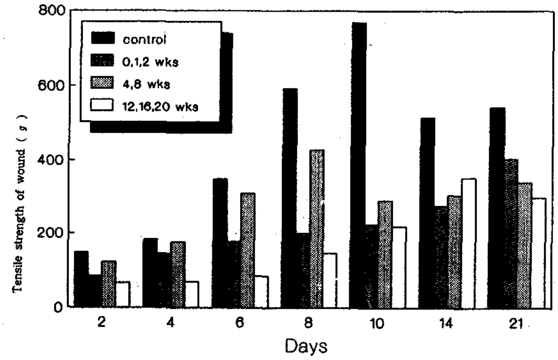

Fig. 9. Comparison of average tensile strength.

during the observation period. Two weeks group also did not reach its maximum value during 21 days of period. Thus it seemed that immediate, 1 and 2 weeks group did not obtain maximum tensile strength during the observation period.

For 4 weeks group, tensile strength rapidly rose from 6th day and it reached its peak value of 499.6 g at 8th day. Eight weeks group attained its maximum value of 357.4 g at 8th day. Therefore it was clear that the 4 and 8 weeks group revealed the similar features of the tensile strength curve to that of the control group.

Tensile strength of twelve weeks group revealed the slow rise from 4th day and reached its maximum at 14th observation day. For 16 weeks group, tensile strength revealed its peak value of 312.0 g at 14th day. Tensile strength of 20 weeks group revealed its peak value of 488.4 g at 14th day. Thus 12, 16 and 20 weeks group attaied their peak values at 14th day of observation, but their appearance appeared to be delayed compared to that of the control group.

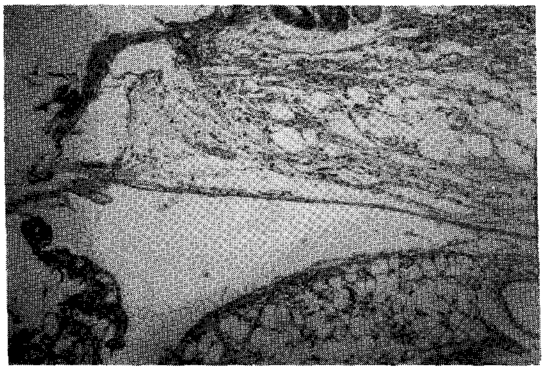

Fig. 10. Section of control wound 2 days after operation showing active epithelization and abundant amount of inflammatory exudate in dermis (hematoxylin and eosin, ×100).

Fig. 11. Section of control wound 6 days after operation showing active proliferation of fibroblasts and abundant newly formed connective tissue (hematoxylin and eosin, ×100).

Fig. 12. Section of control wound 14 days after operation showing a well-healed scar with mature connective tissue (hematoxylin and eosin stain, ×100).

Fig. 13. Section of 4 days old skin wounds from 4 weeks group showing immature but complete epithelization (hematoxylin and eosin, ×100).

All the test groups were categorized into three groups and their tensile strength values were compared to that of the control group (Fig. 9). The absolute tensile strength values of the irradiated groups were diminished at all the time intervals compared to that of the control group. Especially 12, 16 and 20 weeks group revealed markedly reduced values. In respect to the observation period, the absolute values of tensile strength was markedly diminished between 8th and 10th day.

T-test was utilized to find the significance of the observed values (Table 2). There was a significant difference in values from that of the control group at 2, 6, 8, 10, 14, 21st day for immediate group. One week group revealed difference at 2, 4, 6, 8, 10, 14 and 21st day. Therefore it was clear that immediate, 1 and 2 weeks group had a significantly different tensile strength changes from that of the control group for the most of the observation period. In contrast, 4 and 8 weeks group revealed difference only at three time intervals. Twelve, 16 and 20 weeks group had significantly different tensile strength values at six time intervals which indicated slight delay in wound healing process compared to that of the control group.

2. Histopathologic Studies

Histologic sections of the healing wounds of control group demonstrated the normal wound healing process. The surface epithelium covered the skin defects by its stretching and proliferation (Fig. 10). Complete epthelization was observed on 4th day. Six days after wound production, a sharp

Fig. 14. Section of 8 days old wound from one week postirradition group showing less pronounced fibroblastic proliferation compared to that of control group (hematoxylin and eosin, ×100).

Fig. 15. Section of 21 days old skin wounds from 12 weeks postirradiation group showing complete healing of wound with hyalinization of connective tissue (hematoxylin and eosin, ×100).

increase in quantity of fibroblasts in the underlying dermis was observed. Collagen fibers began to appear in the dermis on about 6~8th day (Fig. 11). The diameter of fibroblasts became thick on about 14~21st days and their arrangement appeared to be normal (Fig. 12).

Histologic studies of the irradiated wounds revealed similar reepithelization process to that of control group (Fig. 13). The appearance of fibroblasts was slightly delayed to occur on about 10~14th (Fig. 14). Fibroblasts were decreased in their quantity. The patterns of deposit of collagen was similar, but the the amount of it appeared to be increased, especially in 12, 16 and 20 weeks group (Fig. 15).

In view of above findings, it was clear that complete epithelization occured in the most of irradiated groups as quickly as in the control wounds. But there was a delay in appearance and decrease in its quantity of fibroblasts. Collagen deposit was also delayed slightly compared to the control group. There appeared to be an increased hyalinization in later days secondary to the effects of irradiation. In conclusion, the changes of tensile strength appeared to be more or less correlated with the appearance of fibroblasts and collagen deposit.

DISCUSSION

Radiation therapy can be given preoperatively, postoperatively or intraoperatively. They have their own advantages and disadvantages. Therefore it should be selected in the proper way for each cases.

The preoperative radiation has the advantages of sterilizing cells at the edges of resection, sterilizing cells that perhaps would be dislodged and seeded at the time of surgery and reducing the tumor volume sufficiently to allow resection[7]. But it also has the disadvantage of damaging the normal structures which may cause the delay of wound healing[5]. In these situations, the effect of radiation therapy on the healing ability of a subsequent surgical wound is of great importance. Previous literatures reported that significant wound complication occured in 1~20% of patients who underwent preoperative irradiation[8,9].

Retarding effects of irradiation was firstly mentioned as early as 1923 by Haberland in his experimental work[10]. Radiation depressed the fibroblastic growth and changed the fibroblastic developement with the production of abnormal cells[11].

Radiation dose and irradiation time appeared to be the important factors which affect the wound healing according to several reports[12~15].

Radiation effects was described both in low and high dose ranges. High radiation dose usually means above 1000 cGy given in a single fraction. The ra diation dose of 2000 cGy in a single exposure in our experiment which is radiobiologically equivalent to 5000 cGy at conventional fractionation signifies the clinicallly high radiation dose[16]. It was apparent from our experimental result that large radiation doses caused a considerable delay in wound healing. Similar findings were noticed in other studies where large doses in the order of from 1000 cGy in a single fraction to 4000 cGy at conventional fractions caused a healing disturbances in considerable degree[4]. Contrary, it was believed that small doses accelerated the healing of wounds by lessening the inflammatory exudate and depressing the proliferation of fibroblasts[12,17,18].

Intervals between surgery and irradiation believed to be an another important factor which influenced the wound healing. Though Powers' study did not demonstrate the optimal time delay between irradiation and surgery to permit complete and adequate wound healing, many other studies suggested that irradiation given immediately or 1 week before surgery interfere with rate of wound healing, but in cases of irradiation given 3~12 weeks prior to surgery did not impair the healing of surgical wounds[4,5,13]. Our study proved that the wounds received irradiation 4~8 weeks before surgery did not differ much in the wound healing process from that of the nonirradiated group, but the wounds received irradiation immediately, 1 and 2 weeks before surgery revealed most marked delay in wound healing.

Radiation given after surgery also affects the wound healing process. Nathanson observed that the acceleration of healing occurred in those which the wounds were irradiated immediately after incision, but the wounds received irradiation 24 hours after operation showed marked interference with healing[12]. Other study also proved that the irradiation given within 3 days after surgery delayed the scar formation[19]. On this basis, occasionally we irradiated the incisional scar within 24~48 hours after surgery to prevent the keloid formation.

Healing process of surface wounds can be evaluated by boh tensile and burst strength[6,13,20,21]. Tensile strength is breaking strength per unit cross sectional area. Tensile strength is a fundamental physiological property of tissue which can be considered a function of wound healing[4,18,22,23]. A simple quantitative method of investigating healing of wounds by measurement of tensile strength had been described by Howes, Sooy and Harvey[6]. Levenson et al described the change of tensile strength during the healing process of unirradiated tissue[20]. There was the initial lag period of 4~6 days during which the tensile strength did not rise. This phenomenon was explained as due to the formation of fibrin by the blood or blood clot[6]. Then the wound reached maximal strength at the end of 2 weeks. Histologists suggested that a correlation might exist between number of fibroblasts, quantity of collagen and tensile strength of a scar[20,24]. Our experimental result also supported these findings.

Through the histologic study of the sections, we could analyze the wound healing process and tried to find the correlation between tensile strength and histopathologic findings. It was noticed that the epithelial tissue was less affected than the deeper tissues by irradiation[22]. Thus reepithelization in the irradiated groups took place almost at the same time as that in the control group. But in the deeper tissue, the appearance of fibroblasts was delayed and the absolute amount of fibroblasts was decreased. The collagen deposit was delayed, but the amount was similar to that of the control group. In 12, 16 and 20 weeks groups, hyalinization was prominent due to the degenerative effects of previous irradiation. According to Ritchie's observation, there was a decreased amount of inflammatory cells and fibroblasts and delayed appearance of collagen in irradiated tissue[11]. The quantity and quality of collagen which could be observed in 10 days old surgical scar was not apparent in 20 days old irradiated scar tissue. Therefore, the delayed increment and reduced absolute values of tensile strength presumed to be caused by the changes of fibroblasts and collagen. But it was not sufficient to correlate the wound healing process with the histopathologic findings.

Wound healing process was also influenced by the nutritional status and the presence of foreign materials[5,22]. With the spontaneous removal of suture material, absolute values of tensile strength was relatively increased. There was also a tendency of reduction of tensile strength in later observation periods. which might have been caused by the poor nutritional status.

SUMMARY

The results of the study are summarized as follows:

1. The wounds which received surgery immediately, 1 or 2 weeks after irradiation revealed most marked delay in wound healing in terms of tensile strength measurement.

2. The wounds which received irradiation 4 or 8 weeks before surgery did not differ much in the wound healing process from that of the control group.

3. The groups which received surgery 12, 16 or 20 weeks after irradiation demonstrated slightly delayed healing of wound.

4. Histopathologic studies of the wounds

demonstrated epitelization in most instances as quickly as in the control wounds. The appearance of fibroblasts and collagen fibers has delayed momently and appeared to have close correlation with the tensile strength healing curves.

5. After reaching the peak, the values of tensile strength has shown a trend of decrease in absolute values as follow-up days passed by. It seemed to be affected by nutritional status.

In conclusion, high dose preoperative irradiation caused the delay in wound healing. Especially it could cause a serious delay in wound healing when operation was performed immediately, 1 or 2 weeks after irradiation. When the operation was performed 12, 16 or 20 weeks after irradiation, one could expect moderatte degree of delay in wound healing. But when the surgery was performed 4-8 weeks after irradiation, it could minimize the adverse effects of wound healing process.

REFERENCES

1. **Devita VT Jr:** Progress in cancer management. Cancer 51:2401~2409, 1983
2. **Fletcher GH:** Basic principles of radiotherapy, In Textbook of radiotherapy, 3rd ed, Fletcher GH, Philadelphia, Lea & Febiger, 1980, pp 219-224
3. **Perez CA, Brady LW:** Introduction, In Principles and practice of radiation oncology, Ist ed, Perez CA, Brady LW, Philadelphia, J B Lippincott. 1987, pp 35-38
4. **Powers WE, Ogura JH, Palmer LA:** Radiation therapy and wound healing delay. Radiology 89:112 -115, 1967
5. **Lawrence W, Nickson JJ, Warshaw LM:** Roentgen rays and wound healing, an experimental study. Surgery 33:376-383, 1953
6. **Howes EL, Sooy JW, Harvey SC:** The healing of wounds by their tensile strength. J A M A 90:42-45, 1929
7. **Hellman S:** Priciples of radiotherapy, In Cancer, Principles and practice of oncology, Vol 1, 3rd ed, DeVita VT, Hellman S, Rosenberg SA, Philadelphia, J B Lippincott, 1989, pp 272-273
8. **Deweerd JH, Colby MY:** Bladder carcinoma treated by irradiation and surgery. Interval report. J Urol 109:409-413, 1973
9. **Fortier GA, Krochak RJ, Kim JA, et al:** Dose response to preoperative irradiation in rectal cancer: Implications for local control and complications associated with sphincter sparing surgery and abdominoperineal resection. Int J Rad Onc Biol Phys 12:1559-1563, 1986
10. **Haberland HFO, cited by Lawrence W, Nickson JJ, Warshaw LM:** Roentgen rays and wound healing, an experimental study. Surgery 33:376-383, 1953
11. **Ritchie G:** Effect of roentgen irradition on healing of wounds. Archives of pathology 16:839-851, 1933
12. **Nathanson LT:** Effect on gamma-ray of radium on wound healing. Surg Gyneco & Obst 59:62-69, 1934
13. **Dobbs WGH:** A statistical study of the effect of roentgen rays on wound healing. Am J Roentgenol 41:625-632, 1939
14. **Howes EL, Harvey SC, Hewitt C:** Rate of fibroplasia and differential in the healing of cutaneous wounds in different species of animals. Arch Surg 38:934, 1939
15. **Pohle EA, Ritchie G, Moir WW:** Studies of the effect of roentgen rays on healing of wounds, III. Histologic changes in skin wounds in rats following postoperative irradiation with very small and moderate doses. Radiology 52:707-713, 1949
16. **Hall EJ:** Time, dose and fractionation in radiotherapy, In Radiobiology for the radiologist, 2nd ed, Hall EJ, Maryland, Harper & Row publishers, 1978, pp 275-280
17. **Fukase S, cited by Pohle EA, Ritchie, Moir WW:** Studies of the effect of roentgen rays on healing of wounds, III. Histologic changes in skin wounds in rats following postoperative irradiation with very small and moderate doses. Radiology 52:707-713, 1949
18. **Nathanson LT:** Effect on gamma-ray of radium on wound healing. Surg Gyneco & Obst 59:62-69, 1934
19. **Chung SH, Park CI, Han MC, et al:** The radiation effect on healing of surgical wounds on mouse skin. J of Korean Radiological Soc 18:193-199, 1982
20. **Levenson SM, Geever EF, Crowley LV, et al:** The healing of rat skin wounds. Annals of Surgery Feb: 293-308, 1965
21. **Madden JW, Andrew AJ:** Wound healing: Biologic and clinical features in Sabiston's textbook of surgery, 13th ed, 193-204, Philadelphia, Saunders Co, 1986
22. **Kissane JM:** Inflammation and healing in Anderson's pathology, 8th ed, 47-57, St. Louis, The C V Mosby Co, 1985
23. **Rothman S:** Physiology and biochemistry of the skin, Chapter 1, Chicago, University of Chicago Press, 1954
24. **Madden JW, Peacock EE:** Studies on the biology of collagen during wound healing, 1. Rate of collagen synthesis and deposition in cutaneous wounds of the rat. Surgery 64:288-294, 1968

＝국문초록＝

방사선조사가 피부의 창상치유에 미치는 실험적 연구

인제의대 서울백병원 치료방사선과학교실

서　현　숙

이화여자대학교 의과대학 방사선과학교실

이　정　식

수술전 방사선 조사는 불가능한 수술을 가능케 하며, 암의 크기를 축소시켜 수술범위를 감소시킬 수 있어 조직 및 장기의 보존과 기능을 유지할 수 있게 한다. 그러나 수술의 성과는 완전한 창상의 치유없이는 이루어질수가 없으며 수술전에 시행하는 방사선조사는 창상의 회복을 불완전하게 만들거나 지연시키는 것으로 알려져 있어 문제점으로 대두되고 있다.

본 연구는 252마리의 마우스 좌 우측 하지 상단 내측 피부에 일회 2000 cGy의 방사선조사를 실시한후 즉시, 1, 2, 4, 8, 12, 16, 20주후에 외과적 피부절개 및 봉합과 유사한 창상을 만들었고, 그후 일정한 간격으로 창상의 장력강도 측정과 병리학적 관찰을 하고 방사선조사와 수술이 시행될때까지의 시간적 간격과 창상 치유정도의 상관관계를 분석하여 다음과 같은 결론을 얻었다.

1. 방사선 조사직후와 1주, 2주내에 창상이 발생된 경우에는 장력강도의 최고치의 발현 및 상승속도가 지연되어 통계학적으로 의의가 있었다.

2. 방사선 조사후 4~8주내에 창상이 발생되었을 경우 장력강도의 변화는 대조군과 비교해서 별다른 차이가 없었다.

3. 방사선 조사후 12, 16, 20주내에 창상이 발생되면 장력강도의 최고치의 발현과 상승 속도가 약간 지연되었다.

4. 대조군에 비해 방사선조사군의 창상의 장력강도 평균치는 모든 관찰기간동안 감소 되었다.

5. 병리조직학적 소견상 상피세포는 방사선조사군에서도 정상적인 속도로 재생되었으며 섬유아세포 및 교원질의 출현시기는 대조군에 비해서 약간 지연되었으나 장력강도의 상승과 교원질량간에 상관관계가 존재함을 추정할 수 있었다.

6. 방사선 조사후 창상 발생의 관찰기간이 경과함에 따라 장력강도의 평균치가 오히려 감소됨은 영양과 전신상태에 관련있는 것으로 생각되었다.

그러므로 수술전 고선량의 방사선 조사를 시행하면, 창상치유는 지연시키며 특히 방사선조사 직후, 1주 및 2주후에 수술을 시행할 경우 창상치유를 심하게 지연시킬 수 있고, 또 12, 16, 20주후에도 역시 창상치유의 지연을 예상할 수 있겠다. 그러나 4~8주후에 수술을 시행하면 별부작용없이 창상치유가 가능하므로 가장 적절한 수술시기라고 생각한다.