ELSEVIER

Int. J. Radiation Oncology Biol. Phys., Vol. 34, No. 4, pp. 905–910, 1996

0360-3016/96 $15.00 + .00

0360-3016(95)02182-5

● *Brief Communication*

QUANTITATIVE COMPARISONS OF CONTINUOUS AND PULSED LOW DOSE RATE REGIMENS IN A MODEL LATE-EFFECT SYSTEM

DAVID J. BRENNER, PH.D.,* ERIC J. HALL, D.SC.,* GERHARD RANDERS-PEHRSON, PH.D.,* YOUPING HUANG, PH.D.,* GARY W. JOHNSON, A.A.S,* REGINALD W. MILLER, D.V.M,† BIN WU, M.D.,‡ MARCELO E. VAZQUEZ, M.D., PH.D.,‡ CECILY MEDVEDOVSKY, M.D.‡ AND BASIL V. WORGUL, PH.D.‡

*Center for Radiological Research, †Institute of Comparative Medicine, and ‡Eye Radiation and Environmental Research Laboratory, Columbia University, 630 West 168th Street, New York, NY

Purpose: There is increasing interest and usage of pulsed low dose rate (PDR) brachytherapy, in which a single source is shuttled through the catheters of an implant, typically for about 10 min each hour. This study was designed to compare the late effects produced in various PDR regimens with those from the corresponding continuous low dose rate (CLDR) regimens.

Methods and Materials: A model late-responding system was used, namely, cataract induction in the rat lens. This system has the advantage of being highly quantifiable. The rats eyes were exposed to a total dose of 15 Gy either continuously over 24 h, or with three different PDR regimens, all with the same total dose and overall time. We addressed three questions: (a) are late effects increased when a CLDR regimen is replaced with 10-min pulses repeated every hour? (b) Are late effects increased if hourly 10-min pulses are replaced with 10-min pulses repeated every 4 h? (c) Are late effects increased if 10-min pulses are replaced with 100-s pulses?

Results: We found that the four regimens under test, continuous, 10-min pulses each hour, 10-min pulses every 4 h, and 100-s pulses every hour, showed no significant differences in cataractogenic potential, as estimated with the Wilcoxon-Gehan test. Power tests indicated that the experimental design was adequate to detect relatively small differences in cataractogenicity between regimens.

Conclusions: The equality of late effects from CLDR and PDR in these experiments must imply that sublethal damage repair is quite slow in this model late-responding system, in agreement with trends observed in the clinic for sublethal damage repair of late sequelae. Such trends would suggest that PDR is unlikely to produce significantly worse late effects than the corresponding CLDR regimen, which is in agreement with early clinical data using PDR. Caution, however, is strongly recommended.

Brachytherapy, Pulsed dose rate, Late effects.

INTRODUCTION

Because of its practicality, use of pulsed dose rate (PDR) brachytherapy is increasing (1, 16, 18, 20, 21, 25, 27). In PDR, a continuous low dose rate (CLDR) brachytherapy regimen is replaced with one involving a series of high dose rate pulses, typically (though not always) taking about 10 min each hour and typically (though not always) with the same overall dose and time as the corresponding CLDR regimen.

Pulsed dose rate is achieved with a remote afterloader containing a single high-activity source that is stepped through the catheters of an implant, with dwell positions and times adjusted under computer control to achieve the required dose distribution.

The advantages of PDR have been discussed elsewhere (3, 14). Essentially, they involve a considerably increased level of convenience, both for the patient and for the clinical staff. The patient has much more mobility—during the off periods—than in a conventional CLDR regimen, during which nursing and visiting can be safely accomplished. There are two clinical advantages: (a) by varying the dwell times and locations of the source as it shuttles through the tumor, the dose distribution can be optimized for the actual locations of the implanted catheters relative to the tumor and normal tissues; (b) the overall dose rate can be maintained even as the source decays, by increasing the length of individual pulses. Finally, from a practical viewpoint, the use of a single source

Reprint requests to: David J. Brenner, Ph.D., Center for Radiological Research, Columbia University, 630 West 168th Street, New York, NY 10032.

Acknowledgements—This work was supported by NCI Grant CA-24232, and by Research to Prevent Blindness, Inc.

Accepted for publication 26 October 1995.

is significantly more convenient and safe than the usual inventory of sources.

The key radiobiological question for PDR revolves around the question of equivalence between the results of CLDR and those of a corresponding PDR regimen. Initial calculations (3, 10), based on data from *in vitro* systems, suggested that as long as the time between 10-min pulses was not increased much beyond 1 h, early responding normal tissues would not show significant differences in response between CLDR and PDR (for the same overall dose and time). Subsequent *in vitro* experimental results (2, 6) have corroborated this conclusion, as have *in vivo* studies with an early responding end point (19). The limited clinical experience with PDR reported to date also suggests that early response is not markedly different from CLDR (18, 25).

Several authors, however, have pointed to the need for caution with regard to late effects (3, 8). Essentially this is because of the fact that late-responding tissues are more sensitive than early responding tissues to changes in fractionation patterns (13). These authors pointed out that changes in late effects when moving from CLDR to PDR are essentially determined by the rate of repair of sublethal late-responding damage, and that these repair rates are simply not well known. Essentially, the trend, schematically illustrated in Fig. 1, is that rapid repair rates in late-responding tissues would lead to increased late effects in PDR compared with CLDR (5). On the other hand, slow repair rates would imply that PDR might well produce less late effects than the corresponding CLDR regimen (5). Again, the fact is that these repair rates are not well known (9).

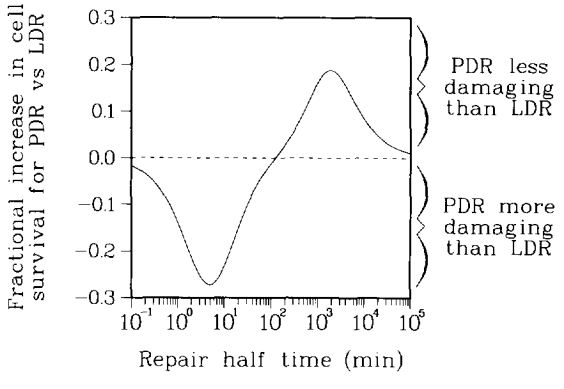

Fig. 1. Calculated fractional change in cell survival for PDR compared with CLDR as a function of the assumed half time for sublethal damage repair. Both treatments consist of 60 Gy delivered in exactly 120 h, either continuously (CLDR) or in 120 equally spaced 10-min 0.5 Gy pulses (PDR). The calculated quantity is $(S_{PDR}-S_{CLDR})/S_{CLDR}$; here the survival ($S$) is calculated, using the linear-quadratic formalism (17), as $S = \exp(-\alpha D - G\beta D^2)$, where D is the total dose, α and β are the linear-quadratic formalism parameters, and G is the quantity describing sublethal damage repair (3, 17), which depends on the half-time of sublethal-damage repair, $T_{1/2}$. Thus, the quantity calculated is actually $\exp[-(G_{PDR} - G_{CLDR})\beta D^2] - 1$. In the calculation, we have assumed that $\beta = 0.025$ Gy^{-2} (26), though similar conclusions are obtained for other values of β.

Experimentally, changes in late-responding sequelae are hard to quantify, particularly when these changes may be relatively small. This is true both in the clinic and in the laboratory. In the clinical PDR series with the largest reported patient size (n = 111) and the longest reported follow-up (treatments from August 1990 to May 1993), no significant difference was seen in late-effect rates between PDR and CLDR (18). Laboratory studies are ongoing, using as an end point rectal toxicity in the rat (28), but no quantitative results are as yet available. In summary, the body of experimental information about late effects from PDR is very limited.

In this work we discuss *in vivo* comparisons between continuous and pulsed exposures for a model late-responding sequela, namely, opacification of the rat lens. This effect, which typically occurs weeks to month after radiation exposure, is not, of course, of direct clinical relevance; cataractogenesis is, however, an experimentally tractable and highly quantifiable model system for the late effects caused by radiation in humans (24).

In this work we seek to address three questions: (a) are late effects increased when a CLDR regimen is replaced with 10-min pulses repeated every hour (keeping the overall dose and time fixed)? (b) Are late effects increased if hourly 10-min pulses are replaced with 10-min pulses repeated every 4 h (again, keeping the overall dose and time the same)? (c) Are late effects increased if 10-min pulses are replaced with 100-s pulses?

This last question relates to the fact that as the single source shuttles through the tumor over, say, a 10-min pulse, even within this pulse individual cells will be subject to considerable variations in instantaneous dose rate.

METHODS AND MATERIALS

Columbia-Sherman female albino rats weighing 260 ± 10 g, were lightly anesthetized (i.p. injection of 25 mg/kg ketamine and 5 mg/kg xylazine) and restrained in specially designed flexible jackets that were attached to a slowly or intermittently rotating turntable accommodating up to 12 rats. Irradiations were performed with 240-kVdc x-ray machine, operating at maximum voltage and with a Thoraeus filter. Rotating the animals corrected for slight radial inhomogeneties in the dose distribution from the x-ray machine. By shielding the rats' bodies with lead, only their heads were exposed to the radiation. Changes in instantaneous dose rate, ranging from 0.62 to 21.6 Gy/h, were accomplished by raising or lowering the x-ray machine and/or changing the current.

All animals were exposed to a total dose of 15 Gy and for an overall time of 24 h, during which time the rats had continuous access to food and water. In our first experiment (Table 1), the heads of 12 animals (24 eyes) were exposed continuously for 24 h, followed immediately by another group of 12 animals who were exposed to a 10-min pulse every hour for 24 h. In a second experiment in which all exposures again totaled 15 Gy in 24 h

Table 1. Details of CLDR and pulsed regimens used

Experiment No.	No. of animals	Total dose (Gy)	Overall time (h)	Pulse Period (h)	Pulse width (min)	Instantaneous dose rate (Gy/h)
Ia	12	15	24		1440	0.62
Ib	12	15	24	1	10	3.6
IIa	12	15	24	1	10	3.6
IIb	10	15	24	4	10	12.9
IIc	9*	15	24	1	1.66	21.6

* While there were originally 10 animals in this group, the harness of the remaining animal became partially loosened, resulting in her head being shielded by the lead block serving as a body shield.

(Table 1), 12 animals were exposed to a 10-min pulse every hour (identical protocol to the first experiment, used as a check of internal consistency), 10 animals were exposed to a 10-min pulse every 4 h, and 10 animals were exposed to a 100-s pulse every hour.

All surviving animals were examined every 2–4 weeks, within a period of 3 days, up to a postirradiation time of 36 weeks. Cataract development was monitored by slit-lamp biomicroscopy and analyzed using a modified version of the Merriam-Focht scoring method (22). The scoring, a semiquantitative technique using a 0–4 range, depends upon the fact that radiation cataracts develop in a characteristically sequential fashion. The earliest changes consist of vacuoles or diffuse opacities around the central suture in the posterior subcapsular region, and are scored as Stage 1. When vacuoles are present but number fewer than 4, a 0.5 cataract is scored. Progression of the posterior subcapsular region and the early involvement of the anterior subcapsular region defines Stage 2. If fewer than four vacuoles or opacities are observed anteriorly a Stage 1.5 cataract is scored. A Stage 3 is noted when the anterior opacities progress and the density of the cataract posteriorly does not allow assessment of the vitreous beyond. If the entire posterior cortex is involved, yet the capsule can still be discerned, a Stage 2.5 cataract is scored. Stage 4 is one with complete anterior opacification preventing visualization of the remainder of the lens. If the opacity has not become severe enough to prevent passage of the beam to the posterior region, but has made detailed visualization impossible, a Stage 3.5 is scored. Further details of the scoring system used are described by Worgul *et al.* (30).

In terms of data analysis, our observations are right censored in the sense that if a cataract appears, we know (within reasonable limits) when it appeared; however, if an animal died from any cause before showing any or complete lens opacification, we do not know what the subsequent history of the lens would have been. Our aim is to estimate the prevalence, $P(t)$, which is the probability that an animal will develop a cataract of a particular stage by a given time, t. For our right-censored data, Kaplan-Meier or product-limit estimation (15) was used to obtain nonparametric maximum likelihood (NPML) estimates of the stage-specific cataract prevalence as a function of time after irradiation. Using this technique, we estimate $1 - P(t)$, which is the probability that the rat does not have a given grade of cataract at time t

$$1 - \hat{P}(t) = \prod_{t_i<t} (1 - C_i/N_i), \qquad \text{(Eq. 1)}$$

where N_i are the number of eyes at risk just prior to time t_i and C_i are the number of eyes that have developed a given stage of cataract by time t_i. Use of Eq. 1 presupposes that cataract development is independent of death. Estimates of the standard error and variance were made using Greenwood's formula (12):

$$\text{var}[\hat{P}(t)] = [1 - \hat{P}(t)]^2 \sum_{t_i<t} C_i/N_i(N_i - C_i)]. \qquad \text{(Eq. 2)}$$

RESULTS

Figure 2 shows NPML estimates of the prevalence of different grades of cataracts for different regimens; typical standard errors are also shown. Estimated prevalences do not necessarily reach 100% if the last observation is censored for any reason.

Direct comparisons between the cataractogenic effects of the different regimens can be made with the Wilcoxon-Gehan test (11, 29). This test uses all available information at a given time postirradiation to test the null hypothesis that the grades of induced cataracts in two groups (e.g., one group exposed to CLDR and one exposed to 10-min pulses, scored at the same time postirradiation) have the same distribution. We calculate a standard score, Z, associated with the Wilcoxon-Gehan test, which is approximately normally distributed with unit variance and zero mean. If the absolute value of Z is greater than 1.64, the null hypothesis (that the treatment of the two groups produced indistinguishable effects) can be rejected at the 10% level of significance; the corresponding value for 5% level of significance is 1.96.

The results are shown in Fig. 3. Values of Z between -1.64 and 1.64 imply statistically indistinguishable effects at the 10% level of significance. It is clear that none of the four regimens used here, all delivering 15 Gy in a 24 h period, induced different late effects from one another, based on any reasonable level of confidence.

To reduce systematic errors, comparisons are shown in

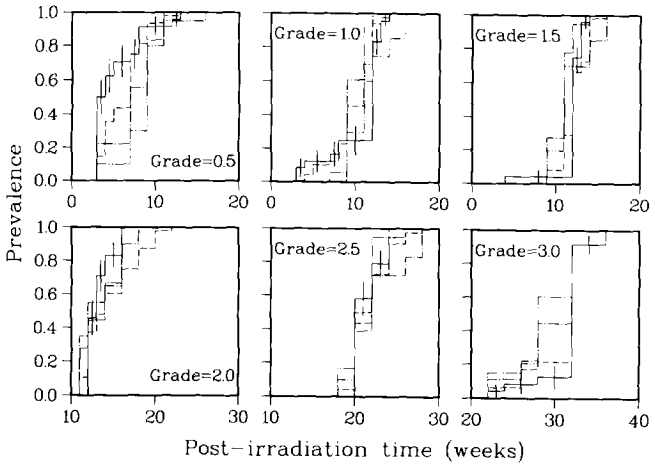

Fig. 2. Prevalence estimates for cataract induction as a function of time postirradiation, for cataracts of various grades induced by 15 Gy of x-rays delivered over 24 h, either continuously or with various pulsed regimens (Table 1). The data for 10-min pulses each hour are combined from two individual experiments, the results of which were indistinguishable using the Wilcoxon-Gehan test. For clarity, standard errors are shown only on the CLDR data set: ——— CLDR; – – – 10-min pulses every hour; – · – · –; 100-s pulses every hour; – · · – · · – 10-min pulses every 4 h.

Fig. 3 only within each of the two experiments. However, comparisons across experiments (e.g., 10-min pulses every 4 h vs. CLDR, and 100-s pulses every hour vs. CLDR) also showed no significant differences.

Statistical power

The results of these experiments are that all four of the regimens under consideration produce results that are not statistically significant from each other. Before we can draw conclusions about these results, it is important to investigate the statistical power of the experimental design; in other words, if there were a difference between two regimens, would the current experiments detect it?

In order to address this question, we have examined the power of the experiments to detect small changes either in the time of onset of cataractogenesis or the severity of cataractogenesis. These investigations were performed using the technique of artificial data simulation.

To assess the power of the current experiments to detect small changes in the time of onset of cataractogenesis, we took the actual experimental data set for the hourly 10-min pulses, and from this generated an artificial data set that was exactly the same except that all the observation times were shifted by a given time, such as 2 weeks. The actual and the artificial data sets were then compared using exactly the same statistical techniques (described above) that were used to compare the four actual data sets. The results indicated that the experimental design has sufficient power to detect fairly small changes in onset time, down to changes of two weeks, though a difference of less than this would not have been detectable.

In further simulations, we addressed the power of the experimental design to detect small changes in the severity of cataractogenesis. Here, we took the actual experimental data set for the hourly 10-min pulses, and from this generated artificial data sets in which the measured grade of unhealthy (nonzero grade) eyes in some fraction

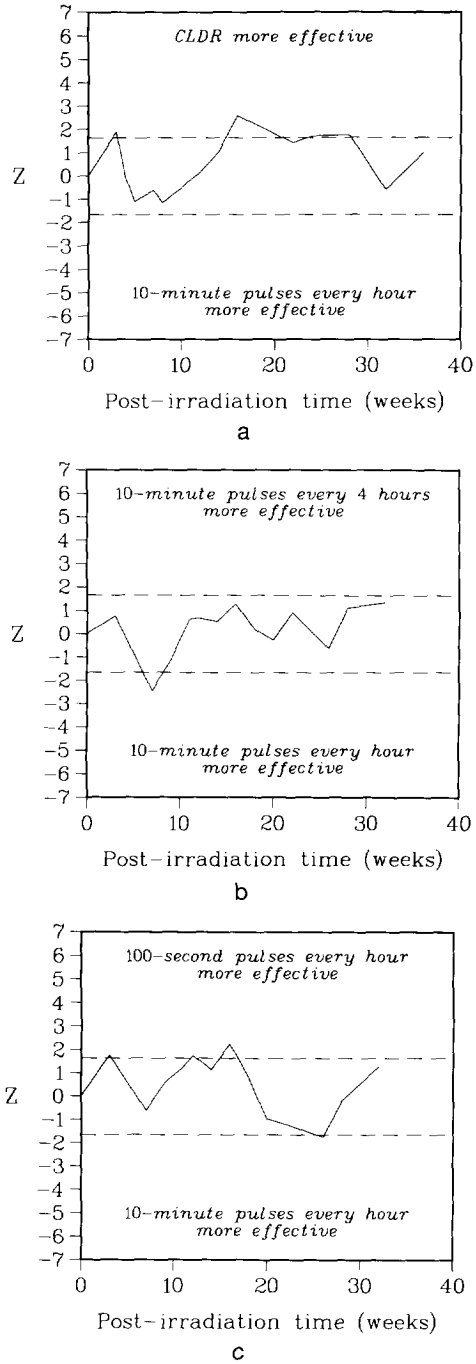

Fig. 3. Standard normal score, Z, associated with the Gehan-Wilcoxon statistic, used to compare the late effects of two different treatment regimens. Z-values between the upper and lower dashed lines indicate that the late effects from the two regimens under comparison are not significantly different at the 90% level of significance: (a) CLDR compared with 10-min pulses each hour; (b) 10 min pulses every hour compared with 10-min pulses every 4 h; (c) 10-min pulses every hour compared with 100-s pulses every hour.

of the animals was increased by one increment (0.5). Again, the actual and the artificial data sets were compared using the same statistical techniques that were used to compare the actual data sets. There were 24 eyes in the actual hourly 10-min pulse study; if five or more of the eyes had their nonzero grades of cataract increased by one increment (0.5), then the experimental design had sufficient power to detect this difference, at a reasonable level of significance.

DISCUSSION

The results from these experiments show that for the model system under consideration here, changing from a continuous to a pulsed regimen does not result in any significant change in late effects, even when pulse intervals as long as 4 h were used. Power studies indicate that the experimental design was sufficiently sensitive to detect relatively small changes in effectiveness. The results suggest that there is some latitude in terms of pulse duration and pulse frequency, relative to the commonly used regimen of a 10-min pulse each hour, within which significant excess late effects are unlikely.

Such results are, *prima facie*, surprising, in that a less continuous regimen, such as the pulsed regimens under consideration here, would be expected to produce significantly greater late effects than CLDR at the same dose. Given that our power studies suggest that the differences in effectiveness between these regimens are genuinely small, and following the logic described in the Introduction, a likely explanation lies in a relatively slow rate of repair for sublethal damage for late-responding end points.

Of course, it is important to emphasize that these results are for a late effect in a model animal system, and extrapolations to late effects of relevance to the clinic must be made with extreme caution. However, there is, as discussed elsewhere (4), good evidence from the clinic that sublethal damage responsible for late sequelae also repairs relatively slowly. The most direct evidence comes from Cox *et al.* (7), who compared the results of hyperfractionated radiotherapy when the interfraction interval was either > 4.5 h or ≤ 4.5 h. Cox *et al.* (7) showed that, while there was no difference in local control, late sequelae were significantly increased for the ≤ 4.5 h interfraction arm. This suggests half times of repair of at least $\sim$200 min for late-responding tissues, and less than $\sim$100 min for early-responding tissues. While this and other evidence (4) suggest that late-responding tissues have slow repair times, implying that the patterns found in this investigation might also hold in the clinic, it is important to emphasize that repair rates are not well known (9); therefore, caution is strongly recommended.

The results here give credence to the notion that practical PDR regimens can be designed that are biologically equivalent to CLDR, not only in terms of early responses such as tumor control, but also in terms of late sequelae. It is not known how long the time between pulses can reasonably be extended while still maintaining biological equivalence—the results discussed here were based on regimens with a maximum of 4 h between pulses. Further laboratory and cautious clinical studies are clearly needed.

REFERENCES

1. Adler, G. F.; Karolis, C.; Kramer, W.; Sun, E. C; Yuile, P. G. Initial PDR brachytherapy experience at Sydney, Australia. Activity (Int. Nucletron Radiother. J.) Report #5:11–14; 1994.
2. Armour, E.; Wang, Z. H.; Corry, P.; Martinez, A. Equivalence of continuous and pulse simulated low dose rate irradiation in 9L glioblastoma cells at 37 degrees and 41 degrees C. Int. J. Radiat. Oncol. Biol. Phys. 22:109–114; 1992.
3. Brenner, D. J.; Hall, E. J. Conditions for the equivalence of continuous to pulsed low dose rate brachytherapy. Int. J. Radiat. Oncol. Biol. Phys. 20:181–190; 1991.
4. Brenner, D. J.; Hall, E. J.; Huang, Y.; Sachs, R. K. Optimizing the time course of brachytherapy and other accelerated radiotherapeutic regimes. Int. J. Radiat. Oncol. Biol. Phys. 29:893–901; 1994.
5. Brenner, D. J.; Hall, E. J.; Huang, Y.; Sachs, R. K. Potential reduced late effects for pulsed brachytherapy compared with conventional LDR. Int. J. Radiat. Oncol. Biol. Phys. 31:201–202; 1995.
6. Chen, C.-Z.; Huang, Y.; Brenner, D. J.; Hall, E. J. Pulsed brachytherapy as a substitute for continuous low dose rate: An *in vitro* study with human cancer cells. Int. J. Radiat. Oncol. Biol. Phys. (submitted).
7. Cox, J. D.; Pajak, T. F.; Marcial, V. A.; Coia, L.; Mohiuddin, M.; Fu, K. K.; Selim, H.; Rubin, P.; Ortiz, H. Interfraction interval is a major determinant of late effects, with hyperfractionated radiation therapy of carcinomas of upper respiratory and digestive tracts: Results from Radiation Therapy Oncology Group protocol 8313. Int. J. Radiat. Oncol. Biol. Phys. 20:1191–1195; 1991.
8. Fowler, J. F. Why shorter half-times of repair lead to greater damage in pulsed brachytherapy. Int. J. Radiat. Oncol. Biol. Phys. 26:353–356; 1993.
9. Fowler, J. F. Are half-times of repair reliably shorter for tumors than for late normal-tissue effects? Int. J. Radiat. Oncol. Biol. Phys. 31:189–190; 1995.
10. Fowler, J. F.; Mount, M. Pulsed brachytherapy: The conditions for no significant loss of therapeutic ratio compared with traditional low dose rate brachytherapy. Int. J. Radiat. Oncol. Biol. Phys. 23:661–669; 1992.
11. Gehan, E. A. A generalized Wilcoxon test for comparing arbitrarily singly censored samples. Biometrika 52:203–223; 1965.
12. Greenwood, M. The natural duration of cancer. Rep. Public Health Med. Sub. 33:1–26; 1926.
13. Hall, E. J. Radiobiology for the radiologist, 4th ed. Philadelphia, PA: J. B. Lippincott Co.; 1994.
14. Hall, E. J.; Brenner, D. J. The dose rate effect in interstitial brachytherapy—A controversy resolved. Br. J. Radiol. 65:242–247; 1992.
15. Kaplan, E. L.; Meier, P. Nonparametric estimation from incomplete observations. J. Am. Stat. Assoc. 53:457–481; 1957.

16. Kovacs, G.; Kohr, P.; Zimmerman, J.; Negel, C.; Kimmig, B. N. Initial PDR brachytherapy experience at Kiel, Germany. Activity (Int. Nucletron Radiother. J.) Report #5:15–21; 1994.
17. Lea, D. E.; Catcheside, D. G. The mechanism of the induction by radiation of chromosome aberrations in Tradescantia. J. Genet. 44:216–245; 1942.
18. Levendag, P.; Visser, A.; Jansen, P. Fractionated brachytherapy in cancer of the head and neck (Abstr.) Radiother. Oncol. 31(Suppl. 1):S35; 1994.
19. Mason, K. A.; Thames, H. D.; Ochran, T.; Janjan, N. Comparison of continuous and pulsed low-dose rate brachytherapy: Biological equivalence *in vivo*. Int. J. Radiat. Oncol. Biol. Phys. 28:667–671; 1994.
20. Mazeron, J. J.; Boisserie, G.; Gokarn, N.; Baillet, F. Pulsed LDR brachytherapy, current clinical status, Paris, France. Activity (Int. Nucletron Radiother. J.) Report #5:24–28; 1994.
21. McLean, M. Initial PDR brachytherapy experience at Toronto, Canada. Activity (Int. Nucletron Radiother. J.) Report #5:6–10; 1994.
22. Merriam, G. R., Jr.; Focht, E. F. A clinical and experimental study of the effect of single and divided doses of radiation on cataract production. Trans. Am. Ophthalmol. Soc. 60:35–52; 1962.
23. Merriam, G. R., Jr.; Worgul, B. V.; Medvedovsky, C.; Zaider, M.; Rossi, H. H. Accelerated heavy particles and the lens. I. Cataractogenic potential. Radiat. Res. 98:129–140; 1984.
24. Schenken, L. L.; Hagemann, R. F. Time/dose relationships in experimental radiation cataractogenesis. Radiology 117:193–198; 1975.
25. Swift, P. S.; Fu, K. K.; Phillips, T. L.; Roberts, L. W.; Weaver, K. A. Pulsed LDR interstitial and intracavitary therapy, clinical experience at UCSF, San Francisco, USA. Activity (Int. Nucletron Radiother. J.) Report #5:29–31; 1994.
26. Turesson, I.; Thames, H. D. Repair capacity and kinetics of human skin during fractionated radiotherapy: Erythema, desquamation, and telangiectasia after 3 and 5 year's follow-up. Radiother. Oncol. 15:169–188; 1989.
27. van Eijkeren, M. Initial PDR brachytherapy experience at Ghent, Belgium. Activity (Int. Nucletron Radiother. J.) Report #5:6–10; 1994.
28. White, J.; Bluj, N.; DeWitt, C.; Armour, E.; Corry, P.; Martinez, A. Animal models for evaluating rectal toxicity from two methods of low dose rate brachytherapy. In Abstracts, Forty First Annual Meeting of the Radiation Research Society, Dallas; 1993.
29. Wilcoxon, F. Individual comparison by ranking methods. Biometrics 2:80–83; 1945.
30. Worgul, B. V.; Brenner, D. J.; Medvedovsky, C.; Merriam, G. J., Jr.; Huang, Y. Accelerated heavy particles and the lens VII. The cataractogenic potential of 450 MeV/amu iron ions. Invest. Opthal. Vis. Sci. 34:184–193; 1993.