CHRONIC IONIZING RADIATION EXPOSURE AS A TUMOR PROMOTER IN MOUSE SKIN

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ABSTRACT

We have tested a chronic exposure to ^{90}Y beta-radiation as a tumor promoter in mouse skin previously exposed to a chemical tumor initiator. Three different tests of radiation as a stage I tumor promoter, in skin subsequently given chemical stage II promotion, all indicated that the beta-radiation acted as a weak stage I skin tumor promoter. It showed no action as either a stage II or complete tumor promoter.

INTRODUCTION

Tests of ionizing radiation for its action at the promotion step in the multi-step process of carcinogenesis have usually indicated a lack of tumor promotion by single or multiple doses of radiation (1,2). One test of γ-radiation as a stage I promoter, given as two doses of 1.0 Gy each to DMBA initiated mouse skin, and followed by a chemical stage II promoter also failed to produce tumors (3). In spite of these negative results, exposure of cells to ionizing radiation results in expression of protein kinase C, C-*jun* and C-*fos* (4,5), molecular changes associated with tumor promotion. *In vivo* tumor promotion by ionizing radiation may therefore occur only at certain doses or dose rates. In this paper we present the results of our tests of chronic exposure to ^{90}Y beta-radiation as a stage I, stage II or complete tumor promoter in mouse skin.

EXPERIMENTAL MEASUREMENTS

Groups of female 7-8 week old SENCAR mice had their dorsal skins initiated by a single topical application of 10 nmol of the carcinogen 7,12, dimethylbenz(a)anthracene (DMBA, in acetone). Some groups subsequently received the complete chemical tumor promoter 12-0-tetradecanoylphorbol-13-acetate (TPA, 2 μg in acetone, twice/week) for 13-20 weeks, after which time multiple skin tumors appeared on the mice. For some groups chemical promotion was broken into two steps, stages I and II. Those groups received TPA stage I promotion, as above for 2 weeks only, followed by mezerein (4 μg) applied in the same protocol for an additional 13 weeks. Multiple skin tumors also appeared after this treatment. Some groups had their dorsal skins irradiated with a ^{90}Y source at a surface dose rate of 13.4 mGy/s, twice/week for varying periods (chronic exposure).

Table 1 shows the results of two tests of multiple 0.5 Gy β-radiation exposures as a complete promoter in DMBA initiated skin. Used alone this radiation exposure produced no tumors, and in conjunction with the complete chemical promoter significantly reduced ($p<0.001$) the tumor

frequency. The reduction in tumor frequency may be attributable to radiation induced death of initiated cells.

Table 1: Chronic β-Radiation Exposure as a Complete Tumor Promoter.

Test No.	Treatment[a]	No. Animals	Tumors/Animal ± SD
1	DMBA → No promotion	23	0
	DMBA → TPA (13 weeks)	25	9.72 ± 0.45
	DMBA → 0.5 Gy (13 weeks)	100	0
2	DMBA → TPA (15 weeks)	25	14.21 ± 0.30
	DMBA → TPA + 0.5 Gy (15 weeks)	22	9.88 ± 0.54

[a] TPA treatment twice per week
0.5 Gy exposure twice per week.

Table 2 shows the tests of chronic exposure to β-radiation used as a stage II promoter in a two stage promotion protocol. DMBA initiated skin, stage I promoted with TPA (2 weeks) followed by 13 weeks of chemical stage II promotion by mezerein produced multiple tumors/animal. Substituting β-radiation exposure (0.5 Gy, twice/week, 13 weeks) for mezerein resulted in essentially no tumors indicating that this radiation exposure did not act as a stage II promoter. The reduced tumor frequency seen when chronic radiation exposure was given prior to or with the chemical stage II promoter, again may be attributable to radiation induced cell death.

Three protocols were used to test chronic radiation exposure as a stage I tumor promoter (Table 3). In the first, initiated skin was subsequently exposed to radiation alone as a stage I promoter followed by chemical stage II promotion. For the second protocol, initiated skin was subsequently treated with a combination of radiation and a chemical stage I promoter (TPA), followed by chemical stage II promotion. The third protocol gave the radiation exposure entirely preceding the initiation, which was then followed by chemical promotion. In all three cases the radiation significantly ($p<0.05$) increased the tumor frequency, in spite of the concurrent, radiation induced loss of initiated cells shown in Tables 1 and 2.

At the doses and dose rates tested the action of β-radiation as a stage I tumor promoter was weak in comparison to the chemical promoters, consistent with its relatively weak action as a tumor initiator (6) or progressing agent (1). However, its promotion action was offset by its apparent lethal action on initiated cells. At lower dose rates, lethal effects may diminish and enhance the effectiveness of radiation as a promoter.

Table 2: Chronic β-Radiation Exposure as a Stage II Tumor Promoter.

TREATMENT[a]	NO. ANIMALS	TUMORS/ANIMALS ± SD
DMBA → TPA → Mezerein (2 wks) (13 wks)	24	6.05 ± 0.29
DMBA → TPA → no treatment (2 wks) (13 wks)	25	0
DMBA → TPA → 0.5 Gy (2 wks)(13 wks)	25	0.14 ± 0.03
DMBA → TPA → 0.5 Gy + Mezerein (2 wks) (13 wks)	25	4.79 ± 0.26
DMBA → TPA → 0.5 Gy → Mezerein (2 wks)(13 wks) (13 wks)	25	4.53 ± 0.28

[a] TPA, Mezerein and/or radiation (0.5 Gy) were all applied twice per week for the indicated time.

Since stage I promotion can occur prior to the DNA damaging initiating event in cells (7), it is likely to depend on an inducible biological process which subsequently acts on the initiated cell to produce the change necessary for this step in carcinogenesis. We have other (unpublished) evidence that tumor promotion is a process which occurs naturally, in the absence of stimulation by chemical or physical agents. We suggest that the chronic radiation exposure prior to or after tumor initiation stimulates this natural process, such that when an initiating event occurs, there is an increased probability that stage I promotion will also occur and tumor frequency will consequently rise.

Table 3: Chronic β-Radiation Exposure as a Stage I Tumor Promoter.

Test No.	Treatment[a]	No. Animals	Tumors/Animal ± SD
1	DMBA → No treatment → Mezerein (13 wks) (13 wks)	24	3.46 ± 0.16
	DMBA → 0.5 Gy → Mezerein (13 wks) (13 wks)	25	3.99 ± 0.23
2	DMBA → TPA → Mezerein (2 wks) (13 wks)	24	6.05 ± 0.29
	DMBA → TPA + 0.5 Gy → Mezerein	25	6.95 ± 0.43
3	MNNG → TPA (20 wks)	25	4.73 ± 0.23
	0.5 Gy → MNNG → TPA (4 wks) (20 wks)	23	5.47 ± 0.37

[a] Chemical tumor promotion or radiation exposure (0.5 Gy) was twice per week.

CONCLUSIONS

Chronic exposure to ^{90}Y beta-radiation does not act as a complete tumor promoter or a stage II tumor promoter in mouse skin previously exposed to a tumor initiator. However, it does act as a stage I skin tumor promoter possibly by stimulating or inducing a naturally occurring stage I tumor promotion process in cells. This result suggests that chronic exposure to radiation may increase the risk of tumor formation in persons exposed to a previous or subsequent tumor initiating dose.

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