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γ-Irradiation facilitates the expression of adoptive immunity against established tumors by eliminating suppressor T cells*

Robert J. North

Trudeau Institute, Inc., P.O. Box 59, Saranac Lake, NY 12983, USA

Summary. *It was found that sublethal (550 rad) whole-body γ-irradiation of mice bearing established immunogenic tumors enabled tumor-sensitized spleen cells infused intravenously 1 h later to cause complete tumor regression in all mice. In contrast, γ-irradiation alone caused only a temporary halt in tumor growth, and immune cells gave practically no therapeutic effect at all. This result was obtained with the SA1 sarcoma, Meth A fibrosarcoma, P815 mastocytoma, and P388 lymphoma. Additional experiments with the Meth A fibrosarcoma revealed that the spleen cells from tumor-immune donors that caused tumor regression in γ-irradiated recipients were T cells, as evidenced by their functional elimination by treatment with anti-Thy-1.2 antibody and complement. It was shown next that adoptive T-cell-mediated regression of tumors in γ-irradiated recipients was inhibited by an intravenous infusion of spleen cells from donors with established tumors, but not by spleen cells from normal donors. The spleen cells that suppressed the expression of adoptive immunity were functionally eliminated by treatment with anti-Thy-1.2 antibody and complement. Moreover, they were destroyed by exposing the tumor-bearing donors to 500 rad of γ-radiation 24 h before harvesting their spleen cells. The results are consistent with the interpretation that γ-radiation facilitates the expression of adoptive T-cell-mediated immunity against established tumors by eliminating a population of tumor-induced suppressor T cells from the tumor-bearing recipient.*

Introduction

Previous publications from this laboratory have shown [1, 2] that it is relatively easy to cause the regression of established, immunogenic murine tumors by the passive transfer of tumor-sensitized T cells, provided the tumors are growing in recipient mice that have been made T-cell-deficient by thymectomy and lethal irradiation, and restored with bone marrow (TXB mice). The need for this type of recipient to demonstrate successful adoptive immunotherapy of established tumors suggested the existence in immunocompetent tumor-bearing recipients of a tumor-induced, T-cell-dependent mechanism that prevents passively transferred, tumor-sensitized T cells from expressing their antitumor function. Evidence that this obstacle to adoptive immunotherapy is a T-cell-mediated mechanism of immunosuppression was supplied by the results of experiments which showed that intravenous infusion of splenic T cells from immunocompetent mice with established tumors can inhibit the ability of passively transferred, tumor-sensitized T cells to cause tumor regression in TXB recipients. On the basis of this and other evidence [12], it was hypothesized that progressive growth of an immunogenic tumor results in the generation of a mechanism of T-cell-mediated immunosuppression that functions to "down regulate" a preceding concomitant antitumor immune response before this response reaches sufficient magnitude to destroy the tumor. It was predicted that any treatment that causes the elimination of suppressor T cells from a tumor-bearing recipient will facilitate the antitumor function of passively transferred T cells and result in tumor regression. It was shown more recently [11], in support of this hypothesis, that cyclophosphamide treatment of tumor-bearing recipients facilitates adoptive immunotherapy of their established tumors by eliminating a cyclophosphamide-sensitive population of suppressor T cells.

The purpose of this paper is to show that sublethal, whole-body γ-radiation facilitaties the expression of passively transferred, T-cell-mediated immunity against an established tumor for the same reason.

Materials and methods

Mice. Specific, pathogen-free $AB6F_1$ (A/J × C57BL/6), $CB6F_1$ (BALB/c × C57BL/6) and $B6D2F_1$ (C57BL/6 × DBA/2) mice, as well as parenteral A/J, C57BL/6 and BALB/c mice, were supplied by the Trudeau Institute Animal Breeding Facility. All mice were free of common viral pathogens as determined by routine testing by the Animal Diagnostic Testing Service of Microbiological Associates, Bethesda, MD.

Tumors. The P815 mastocytoma (DBA/2), P388 lymphoma (DBA/2), Meth A fibrosarcoma (BALB/c) and SA1 sarcoma (A/J) were employed. All tumors were grown for several weeks in vitro in Fisher's medium (Grand Island Biological Co., Grand Island, NY) containing 10% fetal calf serum (FCS) to "cure" them of possible contamination with lactic-dehydrogenase virus. They were then screened for and proven free of viral pathogens. To obtain stock suspensions, the tumors

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Reprint requests should be addressed to R. J. North

were grown as ascites in the peritoneal cavities of syngeneic mice and harvested in phosphate-buffered saline (PBS) containing 5% FCS and 10 U/ml of heparin. They were washed in PBS, counted, dispensed in small vials in Fisher's medium containing 20% FCS and 10% dimethylsulfoxide, and stored over liquid nitrogen. For experiments a vial was thawed, the cells washed with PBS, and $1-2 \times 10^6$ of them used to induce a peritoneal ascites. Ascites tumor cells were harvested 6–8 days later in heparinized PBS-FCS, washed in PBS, counted and resuspended in an appropriate volume of PBS for initiating experimental tumors. Tumors were initiated by implanting 10^6 tumor cells either in a right-hand footpad, or intradermally in the belly region. Tumor growth was followed by measuring changes against time in the mean diameter (mean of two diameters at right angles) of intradermal tumors, or the thickness of footpad tumors with dial calipers [1].

Adoptive immunization. Syngeneic mice were immunized against each of the four tumors by a procedure already described [4] for immunizing against the Meth A and P815 tumors. It involved injecting syngeneic mice intradermally with an admixture of 10^6 tumor cells and 100 g of *Propionibacterium acnes* (*C. parvum* from Burroughs Wellcome Co., Research Triangle Park, NC). This results in a 9–10 day period of tumor growth, followed by complete tumor regression. Tumor regression is associated with the acquisition of long-lived immunity to growth of a tumor implant [4]. The mice were used as donors 1–3 weeks after complete regression of their tumors. Their spleens were finely diced, and forced through a 50-mesh stainless screen into PBS-FCS, triturated to break up clumps, and passed through six layers of surgical gauze to remove debris. They were then washed twice in PBS and resuspended to an appropriate concentration in PBS for intravenous infusion into recipients bearing established intradermal or intrafootpad tumors.

Gamma-Irradiation. Recipient or donor mice were exposed to varying doses of γ-radiation in a ^{137}Cs irradiator that delivered a mid-phantom dose of 30 rad/min.

T-cell-deficient mice. Mice were made T-cell-deficient (TXB) by thymectomy at 4 weeks of age followed 1 week later by lethal whole-body γ-radiation (850 rad). The mice were infused intravenously with 10^6 syngeneic bone marrow cells 1 h after radiation, and employed in experiments no sooner than 6 weeks later.

Anti-Thy-1.2 treatment. Monoclonal IgM anti-Thy-1.2 antibody was produced by a hybridoma (30-H12) from the Salk Institute, La Jolla, CA) growing in vitro in RPMI (Grand Island Biological Co., Grand Island, NY) medium containing 10% FCS. The hybridoma was grown to 10^6 cells/ml, and the culture medium centrifuged to remove cells and debris before it was diluted 1 in 5 in RPMI-FCS. Rabbit serum was employed as a source of complement. It was obtained from rabbits bred at the Trudeau Institute and selected on the basis of minimum toxicity for murine thymocytes. Spleen cells were depleted of T cells by incubating them at 10^7/ml in the 1 : 5 dilution of anti-Thy-1.2 antibody solution for 30 min at 10° C. They were then washed in FBS-FCS and incubated at the same concentration in a 1 : 5 dilution of rabbit serum for 20 min at 37° C. They were washed once in PBS and resuspended in PBS for intravenous infusion.

Results

Gamma-radiation facillitated adoptive immunotherapy of four different immunogenic tumors

It was anticipated, on the basis of results obtained with TXB [1, 2] or cyclophosphamide-treated tumor-bearing recipients [11], that whole-body γ-radiation of tumor-bearing recipients would facilitate the expression of passively transferred, T-cell-mediated immunity against an established tumor by eliminating a tumor-induced population of suppressor T cells. This possibility was tested with the Meth A fibrosarcoma, SA1 sarcoma, P815 mastocytoma and P388 lymphoma growing from an intradermal implant. It can be seen in Fig. 1, that with all four tumors, 500 rad of γ-radiation on day 4 of tumor growth followed 1 h later by intravenous infusion of one organ equivalent (1.5×10^8) of spleen cells from tumor-immune donors resulted, after about a 4–6 day delay, in complete regression of tumors in all mice. In contrast, γ-radiation alone caused a temporary halt in progression of the Meth A and P815 tumors, whereas immune cells alone gave either a partial effect, or no therapeutic effect at all. Because of the delay after the passive transfer of tumorsensitized T cells before adoptive immunity was expressed in γ-irradiated recipients, the tumors had time to grow to a relatively large size before they underwent regression. The results indicate that γ-radiation eliminates a mechanism from the tumor-bearing recipients which functions to prevent passively transferred, tumour-sensitized spleen cells from expressing their antitumor function.

The dose of γ-radiation required to facilitate the expression of passively transferred immunity was between 250 and 500 rad as shown in Fig. 2. This experiment employed recipients bearing a Meth A tumor in the right-hind footpad instead of intradermally in the belly region as in Fig. 1. It will be noted that the tumor growing at this site was not sensitive to γ-radiation alone. This difference in radiosensitivity between intradermal and intrafootpad tumors has been repeatedly observed in this laboratory.

The identity of the spleen cells that passively transfer immunity against the Meth A fibrosarcoma in γ-irradiated recipients is shown in Fig. 3, where it can be seen that the responsible cells were T cells, as evidenced by their functional elimination by treatment with anti-Thy-1.2 antibody and complement.

γ-Radiation does not directly affect tumor growth

It was shown in the preceding section that γ-radiation alone caused a significant, though temporary, halt in the Meth A fibrosarcoma and P815 mastocytoma growth. It might be argued, therefore, that γ-irradiation facilitates the expression of passively transferred immunity against these tumors by directly destroying some tumor cells and damaging others. That this is not the case, however, is shown by the results of an experiment designed to determine whether γ-irradiation alone would cause a reduced rate of growth of the Meth A tumor in TXB mice. The results in Fig. 4 show that whereas 500 rad of γ-radiation caused, after about a 4-day delay, a significant reduction in the rate of growth of the Meth A tumor growing in immunocompetent mice, the same dose of radiation had no effect at all on the growth of the same sized tumor growing in TXB mice. It is apparent, therefore, that the antitumor effect of γ-irradiation is based on its ability to facilitate the expression of a T-cell-dependent, antitumor mechanism in the host.

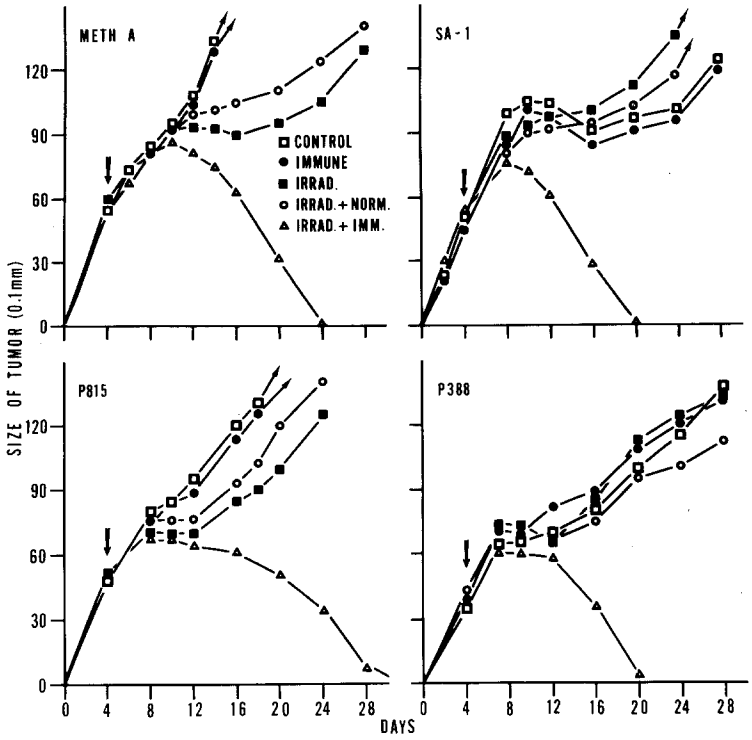

Fig. 1. Demonstration that combination therapy consisting of 500 rad of whole-body γ-radiation, followed 1 h later by intravenous infusion of one-organ equivalent (1.5×10^8) of spleen cells from immune donors (IRRAD + IMM) on day 4 of tumor growth (*arrow*) is capable, after about a 4-day delay, of causing complete regression of four different immunogenic tumors: the Meth A fibrosarcoma (BALB/c), SA1 sarcoma (A/J), P815 mastocytoma (DBA/2), and P388 lymphoma (DBA/2). Immune spleen cells alone (IMM) had no effect on tumor growth, whereas γ-radiation alone caused a significant, though temporary delay in tumor growth that was not modified by infusion of 1.5×10^8 normal spleen cells (IRRAD + NORM). In this experiment tumors were initiated intradermally in the belly region. Means of five mice per group

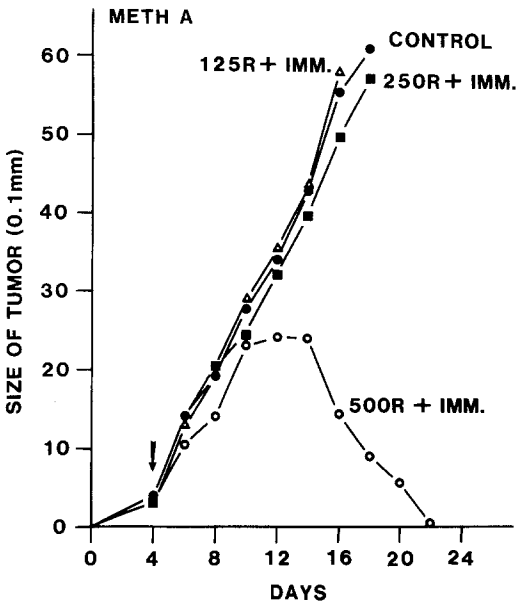

Fig. 2. Evidence that the dose of γ-radiation required to facilitate the expression of passively transferred immunity was between 250 to 500 rad. In this experiment the recipients were bearing a Meth A tumor in the right-hind footpad. γ-radiation was given on day 4 (*arrow*) and 1.5×10^8 immune spleen cells were infused intravenously 1 h later. Means of five mice per group

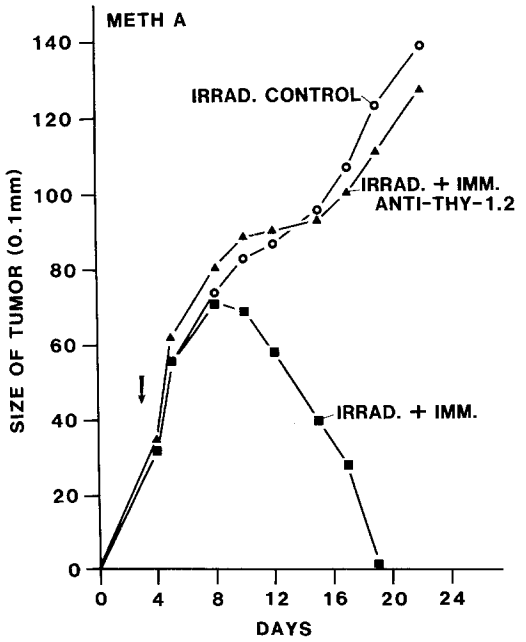

Fig. 3. Spleen cells from immunized mice which, on passive transfer, were capable of causing tumor regression in γ-irradiated recipients were T cells, as evidenced by their functional elimination by treatment with anti-Thy-1.2 monoclonal antibody and complement (IRRAD + IMM ANTI-THY-1.2). This experiment was performed with recipients bearing a Meth A tumor that was initiated intradermally in the belly region 4 days earlier. Means of five mice per group

γ-irradiation-facilitated adoptive immunotherapy is inhibited by passive transfer of T cells from tumor-bearing donors

The results of previous studies that employed TXB [1, 2] or cyclophosphamide-treated [11], tumor-bearing recipients, showed that tumor regression caused by the passive transfer of tumor-sensitized T cells could be inhibited by passive transfer of splenic T cells from mice with established tumors. It was anticipated that γ-irradiation-facilitated adoptive immunotherapy of established tumors also would be inhibited by spleen cells from tumor-bearing donors. This was tested by exposing mice bearing a 4 day Meth A tumor to 500 rad of γ-radiation, infusing them intravenously 1 h later with one organ equivalent (1.5×10^8) of spleen cells from immune donors, and infusing them after a further 3 h with one organ equivalent (1.5×10^8) of spleen cells from donors bearing a 14-day tumor.

It can be seen in Fig. 5 that tumor regression caused by combination therapy with γ-radiation and immune spleen cells was inhibited by passive transfer of spleen cells from donors with established tumors. In contrast, intravenous infusion of the same number of spleen cells from normal mice had no inhibitory effect on the expression of passively transferred immunity. This result was obtained with the Meth A fibrosarcoma and P815 mastocytoma.

Evidence that the spleen cells from tumor-bearing donors which suppress the expression of adoptive immunity in γ-irradiated recipients were T cells is shown by the results with the Meth A tumor in Fig. 6, where it can be seen that suppressor activity was eliminated by treating the spleen cells with anti-Thy-1.2 antibody and complement.

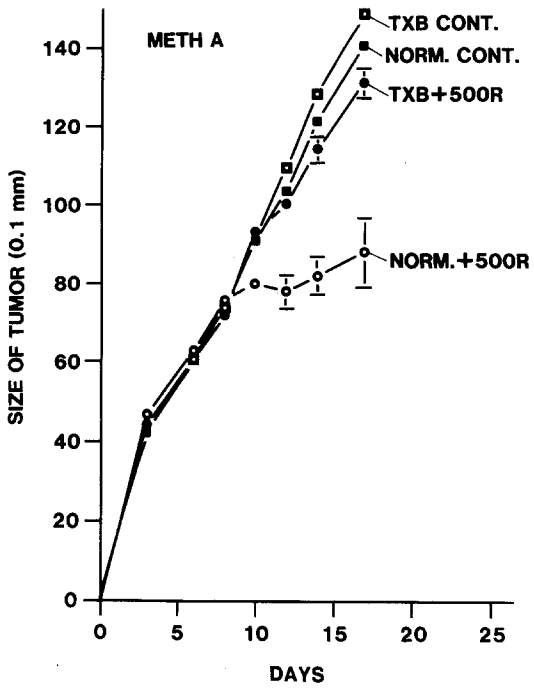

Fig. 4. Evidence that the antitumor effect of 500 rad of γ-radiation alone is not direct, but depends on an intact T-cell system. Shown are the results of giving 500 rad of γ-radiation to tumor-bearing normal mice (NORM + 500 R), and tumor-bearers that had been made T-cell-deficient by thymectomy and irradiation (TXB + 500 R). γ-radiation caused a significant antitumor effect in normal tumor bearers, but not in TXB tumor bearers. The experiment was performed with the Meth A fibrosarcoma initiated intradermally in the belly region. γ-radition was given on day 4 of tumor growth. Means of five per group ± SE

Radiosensitivity of immune cells and suppressor cells

The foregoing results show that passive transfer of tumor-sensitized T cells results in the complete regression of tumors growing in γ-irradiated recipients, but not in normal recipients. They show, in addition, that tumor regression caused by passively transferred immune T cells can be inhibited by an infusion of T cells from donors with established tumors. These results indicate that γ-radiation facilitates the expression of adoptive immunity against an established tumor by eliminating a population of suppressor T cells from the tumor-bearing recipient. If so, it would follow that suppressor T cells should be eliminated from tumor-bearing donors if the donors are given 500 rad of γ-radiation. This was investigated by an experiment that measured the radiosensitivity of the T cells from immune donors which passively transfer immunity to γ-irradiated tumor-bearing recipients, and the suppressor T cells from tumor-bearing donors that suppress the expression of passively transferred immunity. This experiment employed the Meth A fibrosarcoma growing in a hind footpad.

It can be seen in Fig. 7 (panel A), in keeping with the preceding results, that when mice bearing an intrafootpad tumor were given 500 rad of γ-irradiation and infused with 1.5×10^8 immune spleen cells 1 h later, complete tumor regression occurred in all mice. However, tumor regression failed to occur if the immune spleen cells were harvested from mice that were exposed to 500 or 800 rad of γ-radiation 24 h before. On the other hand, exposing immune mice to 250 rad caused only partial elimination of immune T cells, whereas 125 rad of γ-radiation was without effect. It will be noted again that the Meth A tumor growing in the footpad was not sensitive to γ-radiation alone.

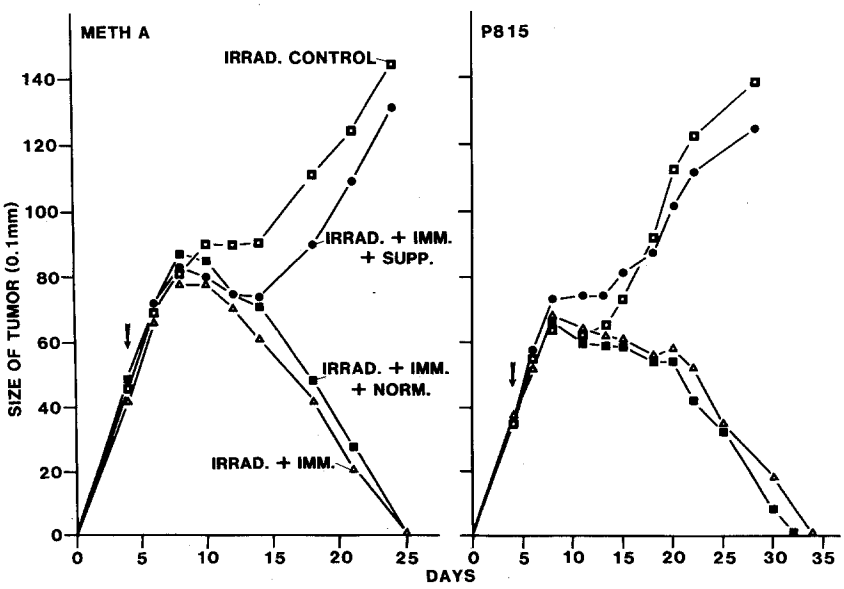

Fig. 5. Tumor regression caused by combination therapy consisting of 500 rad γ-radiation on day 4 followed 1 h later by infusion of 1.5×10^8 immune spleen cells (IRRAD + IMM) was inhibited by infusion after a further 3 h of 1.5×10^8 spleen cells from mice bearing a 14-day tumor (IRRAD + IMM + SUPP), but not by the same number of normal spleen cells (IRRAD + IMM + NORM). The Meth A and P815 tumors were initiated intradermally in the belly region. Means of five mice per group

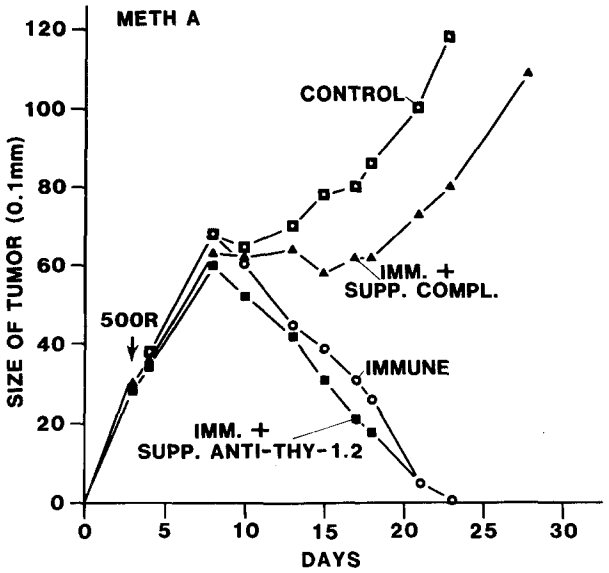

Fig. 6. The spleen cells from donors with a 14-day tumor which were capable, on passive transfer, of suppressing the expression of immunity by passively transferred immune T cells (IMMUNE), as shown in Fig. 5, were T cells, as evidenced by the total elimination of suppressor function by treatment with anti-Thy-1.2 monoclonal plus complement (IMM + SUPP ANTI-THY-1.2). Complement alone (IMM + SUPP.COMPL) was without effect on suppressor cells. All groups were irradiated on day 3, 1 h before receiving donor spleen cells. Means of five mice per group

Figure 7 (panel B) shows, in addition, that suppressor T cells also were functionally eliminated by γ-irradiation. It can be seen that, whereas spleen cells from donors with 14-day tumors inhibited the capacity of immune spleen cells to cause tumor regression in γ-irradiated recipients, spleen cells from 14-day tumor-bearing donors given 500 or 800 rad of γ-radiation 24 h before cell harvest failed to inhibit the expression of adoptive immunity. Suppressor T cells were not eliminated by exposing the donors to 250 or 125 rads of γ-radiation. These results leave little doubt that the T cells in tumor-bearing donors that can passively transfer suppression are eliminated by a dose of γ-radiation that needs to be given to tumor-bearing recipients in order to facilitate the regression of their tumors by passively transferred immune T cells.

Discussion

The literature shows [13] that, until recently, most attempts to adoptively immunize against established, immunogenic tumors have been unsuccessful. Recent experiments in this laboratory [1, 2, 11] have provided evidence consistent with the hypothesis [12] that the failure of passively transferred, tumor-sensitized T cells to mediate the regression of established tumor is caused by the presence in the tumor-bearing recipients of a tumor-induced population of suppressor T cells that functions to "down regulate" a preceding concomitant immune response before it reaches a sufficent magnitude to

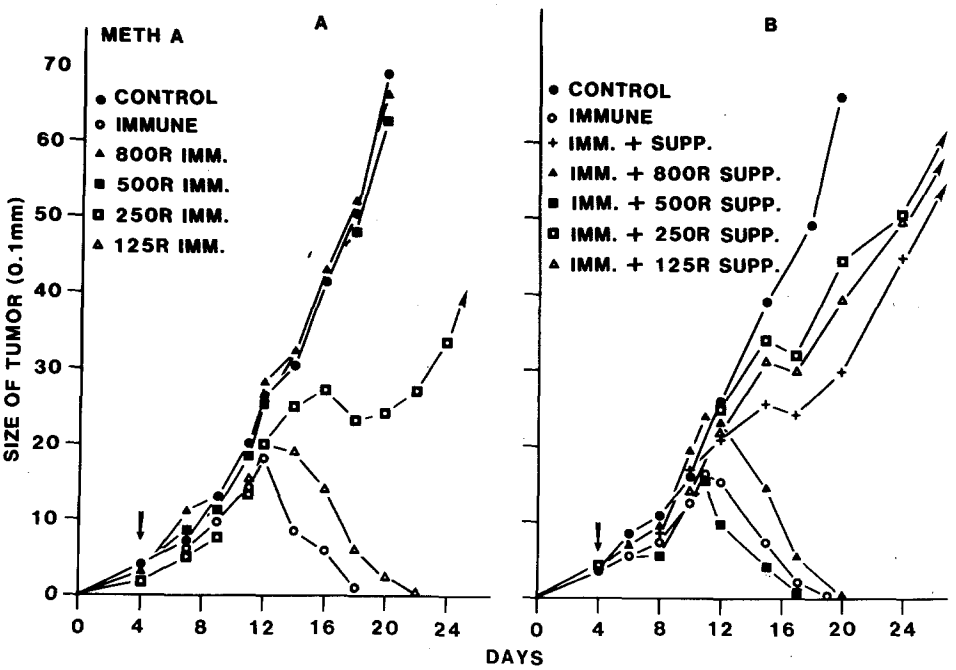

Fig. 7. Effect of graded doses of γ-radiation on T cells that transfer immunity and T cells that transfer suppression. In this experiment all test recipients, except controls, were given 500 rad of γ-radiation on day 4 of growth of a footpad tumor (*arrows*). After a further 1 h the irradiated recipients were infused with 1.5×10^8 spleen cells from immune donors that had received 800, 500, 250, or 125 rad of γ-radiation 24 h before harvesting their spleen cells (*left-hand panel*). It can be seen that it required between 250 and 500 rad of radiation to destroy T cells capable of transferring immunity. The *right-hand panel* shows that regression of recipient tumor caused by infusion of immune cells (IMMUNE) was inhibited by an infusion of suppressor spleen cells from mice with a 14-day tumor, (IMM + SUPP), but was not inhibited by spleen cells from tumor bearers given 500 or 800 rad of γ-radiation 24 h before spleen-cell harvest. In this experiment the recipient and donor tumors were growing in the right-hind footpad. Means of five mice per group

cause tumor regression. Evidence for the generation of this mechanism of T-cell-mediated suppression was supplied by the demonstration that (a) the passive transfer of tumor-sensitized T cells can cause the regression of established tumors T-cell-deficient recipients, but not in immunocompetent recipients, and (b) this adoptive T-cell-mediated regression of a tumor in T-cell-deficient recipients can be inhibited by passive transfer of splenic T cells from immunocompetent donors with established tumors [1, 2]. It was shown more recently [11] that cyclophosphamide treatment of tumor-bearing donors facilitates adoptive T-cell-mediated regression of their tumors, and that this tumor regression is also inhibited by passive transfer of cyclophosphamide-sensitive suppressor T cells from donors with established tumors. More recent evidence shows [3] that the T cells that passively transfer immunity against an established tumor, and the T cells from tumor-bearing donors which suppress this immunity are specific for the tumor that evokes their generation.

In keeping with the results obtained with cyclophosphamide, the results presented here indicate that γ-radiation of tumor-bearers facilitates the expression of adoptive immunity against their tumors by eliminating a radiosensitive population of suppressor T cells. They show that complete regression of an established tumor can result from combination therapy consisting of 500 rad of γ-radiation followed 1 h later by intravenous infusion of immune cells, and that tumor regression can be inhibited if the recipients are also infused with T cells from donors with established tumors, but not with spleen cells from normal donors. The finding that suppressor T cells can be eliminated from the tumor-bearing donors by the dose of γ-radiation that needs to be given to tumor-bearing recipients, in order for passively transferred immune T cells to cause tumor regression is convincing evidence that γ-radiation facilitates adoptive immunotherapy of established tumors by eliminating a tumor-induced population of suppressor T cells. The finding that combination therapy with γ-irradiation and immune spleen cells caused the regression of two leukemias and two sarcomas suggests that this result is likely to be obtained with many other immunogenic tumors. It has been shown [7] that adoptive immunotherapy of syngeneic rat tumors with tumor-sensitized T cells generated in vitro requires that the tumor-bearing recipients be sublethally irradiated.

There was no evidence that suppressor T cells were selectively eliminated by γ-irradiation. On the contrary, the results show that suppressor T cells were no more radiosensitive than the immune T cells that passively transfer immunity. It should be pointed out, in this connection, that the immune T cells that are employed routinely in this laboratory to adoptively immunize against established tumors are not cytolytic effector T cells, but memory or helper T cells with no cytolytic activity of their own [12]. This must be taken into consideration when attempting to explain why tumor regression does not commence immediately after the passive transfer of immune T cells, but commences only after about a 6 to 8-day delay. It was shown recently with TXB tumor-bearing recipients [10] that this appreciable delay, which allows the tumors to grow to a relatively large size before they undergo regression, represents the time needed for passively transferred memory T cells to give rise to an adoptive, cytolytic T cell response in the recipients. It was demonstrated, in addition [10], that passive transfer of suppressor T cells from tumor-bearing donors greatly inhibits this adoptive cytologic T cell response in the recipients. It is apparent, therefore, that suppressor T cells function in these models of adoptive immunotherapy to prevent adoptively immunized recipients from generating a sufficient number of effector T cells to cause the regression of their tumors. In support of this interpretation is published in vitro evidence [8, 14, 15] showing that suppressor T cells function to inhibit the generation, rather than the function, of tumor-sensitized cytolytic T cells.

However, previous studies of immunosuppression in this laboratory were not performed with γ-irradiated recipients, but with TXB recipients that were incapable themselves of generating cytolytic T cells. It cannot be assumed that γ-irradiated recipients also are incapable of generating effector T cells. On the contrary, ongoing research in this laboratory (North, to be published) shows that exposing mice bearing 4-day tumors to 500 rad of γ-radiation does not suppress the capacity of these mice to generate concomitant immunity, as measured by immunity of tumor-bearers to growth of a tumor implant. If anything, concomitant immunity is enhanced by this dose of γ-radiation, and this could explain the significant, though temporary, reduction in the rate of tumor growth which occurs 6 days after giving γ-radiation alone to mice bearing a 4-day Meth-A tumor: an explanation that is supported by the additional finding that the antitumor effect of γ-radiation is not expressed in T-cell-deficient tumor bearers. Indeed, this explanation is in keeping with the findings of Hellström et al. [9] who showed that irradiation of mice bearing as well-established immunogenic fibrosarcoma resulted, after an appreciable delay, in complete regression of the tumor in some mice and partial tumor regression in others. The additional finding by these authors that this antitumor effect of irradiation could be inhibited by intravenous infusion of T cells from the normal mice, was interpreted as meaning that irradiation causes tumor regression by eliminating a population of precursor suppressor T cells, thereby enabling the host to generate an antitumor immune response. Similar results were later reported from another laboratory [6]. However, attempts in this laboratory to use γ-radiation alone to induce regression of the Meth A fibrosarcoma, P815 mastocytoma and P388 lymphoma have failed, although as discussed above, a greatly reduced rate of tumor growth can occur. In the case of the SA1 sarcoma, however, γ-radiation can result, after a 4–6-day delay, in complete regression of this tumor in most mice, provided the mice are γ-irradiated after the tumor has been growing for at least 6 days (North, to be published). It may well be formally shown, therefore, that tumor regression caused by sublethal doses of ionizing radiation represents a convincing example of tumor immunotherapy.

The purpose of discussing these antitumor effects of irradiation alone is to draw attention to the likely possibility that a γ-irradiated tumor-bearing recipient is not imunosuppressed, but capable of generating normal or higher than normal levels of concomitant immunity. If so, then the antitumor function of passively transferred, sensitized T cells can be overestimated, in that the recipient may well be generating almost enough immunity of its own to cause the regression of its tumor. This possibility would need to be considered in interpreting the results of experiments that employ cytolytic T cells [5], or helper T cells [7] generated in vitro to cause the regression of tumors in irradiated recipients. Even so, the evidence presented here shows that the success of attempts to adopticely immunize against an established tumor in an irradiated recipient depends ultimately on the ability of irradiation to delay the onset of expression of tumor-induced, T-cell-mediated immunosuppression for a long enough period

to allow passively transferred, tumor-sensitized T cells to express their antitumor function.

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