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Interstitial Photoradiation Therapy for Primary Solid Tumors in Pet Cats and Dogs

Thomas J. Dougherty,[1] Richard E. Thoma, Donn G. Boyle, and Kenneth R. Weishaupt

Division of Radiation Biology, Roswell Park Memorial Institute, Buffalo, New York 14263 [T. J. D., D. G. B., K. R. W.], and Town and Country Animal Clinic, Cheektowaga, New York 14225 [R. E. T.]

ABSTRACT

Photoradiation therapy, a new method for treatment of solid malignant tumors, depends upon the tumor localization and retention of hematoporphyrin derivative, which is activated *in vivo* by light in the red region of the spectrum. As currently applied to cutaneous and s.c. lesions, the light dose is limited by both normal tissue reactions and the effective penetration of the light through the tissues. In this report, primary solid malignant lesions in pet cats and dogs have been treated by interstitial photoradiation therapy by applying the activating light from a laser [635 ± 5 (S. D.) nm] directly into the tumor masses through a 200-μm quartz fiber optic.

Twelve of 14 lesions (four osteosarcomas, two squamous cell carcinomas, two malignant melanomas, one mast cell sarcoma, one fibrosarcoma, one sebaceous gland sarcoma, and a metastatic prostatic carcinoma) responded to treatment, and three are currently considered permanently controlled at 1 year or more following treatment. This method has not only allowed photoradiation therapy to be applied to some remote lesions but has also nearly eliminated normal tissue effects, thus greatly extending the applicability of this treatment to a wide range of human tumors.

INTRODUCTION

Photoradiation therapy is currently under investigation as a new form of treatment for solid malignant tumors in humans. In its present form, the method involves *in vivo* photosensitization by HPD[2] following its relatively specific uptake and retention in malignant tissue (6–9). Such photosensitization apparently produces singlet oxygen as the cytotoxic agent (10), resulting in rapid tumor necrosis.

A tumor cure rate of approximately 50% has been demonstrated in isogenic rodent systems (2). In addition, a wide range of human tumors have been shown to be responsive to this modality (3, 4). In most cases, the tumors treated thus far by photoradiation have been cutaneous or s.c. metastatic lesions which had progressed or recurred following conventional therapy. Since the normal tissue damage can be minimized, even in areas in which lesions have recurred following tolerance ionizing radiation, this technique offers the possibility of a safe and effective treatment when other modalities either are no longer effective or cannot be repeated. As presently applied, however, the major limitations of this method are the response of normal tissue (skin in most cases examined to date) to the therapeutic light (600 to 700 nm) and the limited effective penetration of the light through tissue, estimated to be approximately 2 cm (3). This report demonstrates that both of these limitations can be eliminated by delivering the light through a quartz fiber optic, imbedded directly into the tumor mass. In order for this to be done effectively, a dye laser operating at 635 ± 5 (S. D.) nm is used as the light source. This technique, demonstrated in a variety of primary lesions in dogs and cats, greatly extends the applicability of photoradiation to remote and life-threatening lesions in humans.

MATERIALS AND METHODS

Animals and Tumors. Pet cats and dogs with primary tumors were obtained directly from owners following discussion of options for alternative treatment and the experimental nature of interstitial photoradiation. Animals were returned to the owners following treatment in most cases. Positive biopsies were obtained in every case from tissue sections sent to the Diagnostic Laboratory, New York State College of Veterinary Medicine at Cornell University. Where necessary, X-rays were obtained before and at various periods following treatment.

Tumor types and tumor sites are indicated in Table 1.

Drug. HPD was prepared by a modification of the method of Lipson (5), as described previously (3, 4).

Method. Two days prior to treatment, the animals were given injections of HPD (5.0 mg/kg i.v.) and kept out of sunlight to prevent photosensitivity reactions. Animals were anesthetized during treatment with halothane or xylazine and ketamine. For treatment, light was delivered through a Quartz Silice A-200 fiber optic guide, 200-μm step index (Quartz Products Corp., Watchung, N. J.). The ends of the fiber were cleaved optically flat by means of a sapphire blade (Math Associates, Great Neck, N. Y.) after the protective coating was stripped. Light was delivered into the fiber by coupling to the output beam of a Spectra-Physics Model 375 dye-laser (Spectra-Physics, Inc., Mountain View, Calif.) operating with rhodamine B dye and without the tuning wedge. The dye-laser was pumped with either a 4- or a 15-watt Spectra-Physics argon laser. The spectral output was centered at 635 nm and was approximately 10 nm wide. The maximum intensity available through the fiber was approximately 300 milliwatts using the 4-watt argon laser and 1500 to 2000 milliwatts using the 15-watt argon laser. The fiber was coupled to the dye-laser using a fiber holder assembly (Oriel Optic's Corp., Stamford, Conn.) fitted with ×10 microscope lens to allow focusing of the laser beam onto the fiber. Coupling efficiency (*i.e.*, output from the fiber relative to input to the fiber) exceeded 70% and was independent of fiber length within the range utilized (10 to 30 ft).

Prior to insertion, the fiber was soaked for 5 min in Cidex (Arbrook, Inc., Arlington, Texas) and cleaned with 70% ethanol. The fiber was then inserted through an 18-gauge needle which

[1] To whom requests for reprints should be addressed, at: Division of Radiation Biology, Roswell Park Memorial Institute, 666 Elm Street, Buffalo, N. Y. 14263.

[2] The abbreviation used is: HPD, hematoporphyrin derivative.

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Table 1

Response of primary tumors to interstitial photoradiation

All animals received HPD (5.0 mg/kg) 48 hr prior to treatment. Animals 1, 7, and 10 were pet cats; others were pet dogs. Light dose is the number of joules delivered over 20 to 30 min for each insertion of the fiber. In general, the fiber was inserted sequentially about 1 to 1.5 cm apart in lesions larger than 2 cm in diameter.

Animal	Tumor type	Location	Size (cm)	Tumor light dose/insertion (J)	Tumor response	Normal tissue effects	Follow-up[a]
1	Osteosarcoma	Mandible	2.5 x 2.5	360 (2)[b]	Complete[c]	None	34 mos.
2	Osteosarcoma	Sinus cavity-hard palate	3.0 x 3.5	360 (3)	Partial	None	1 mo.
			2.0 x 1.5[d]	720 (3)	Complete	None	12 mos.
3	Osteosarcoma	Tibia	7.5 x 6.5 x 3.5	300 (5)	None	None	1 mo.
4	Osteosarcoma	Tibia	5.0 x 2.0 x 2.0	300 (6)	None	None	1 mo.
			5.0 x 2.0 x 2.0[e]	960 (14)	None	None	2 mos.
5	Metastatic prostatic adenocarcinoma	Pelvis	4.0 x 4.0	288 (2)	Partial	Slight erythema (skin overlying lesion)	1 mo.
6	Malignant melanoma (amelanotic)	Hard palate	4.0 x 2.0	240 (2)	Partial	None	3 wk
			2.0 x 2.0[f]	240 (2)	Complete	None	12 mos.
	Malignant melanoma (pigmented)	Hard palate	3.0 x 2.0	240 (2)	None	None	3 wk
			3.0 x 2.0[f]	240 (2)	Partial	None	2 mos.
			0.5 x 0.5[g]	200 (1)			
				360 (1)[h]	Complete	None	12 mos.
7	Squamous cell carcinoma	Sinus cavity-mandible	2.0 x 1.0	360 (1)	None	None	1 mo.
			2.0 x 1.0[d]	525 (2)	Partial	None	3 mos.
			1.5 x 1.5[i]	270 (2)	Partial	None	4 mos.
				270 (2)	Partial	Some sloughing of soft tissue adjacent to lesion	5 mos.
8	Squamous cell carcinoma	Hard palate	3 x 4 x 2	300 (3)	Partial	None	3 wk
			3 x 3 x 1[f]	144 (7)	Partial	None	3 wk
			3 x 2 x 1[f]	187 (8)	Partial	None	3 wk
9	Mast cell sarcoma	Sternum-s.c.	2.5 x 2.5	900 (3)	Complete	Skin sloughing over lesion	8 mos.
10	Fibrosarcoma	Leg (3 lesions)	2.3 x 1.0	324 (2)	Partial	None	1 mo.
			3.5 x 2.5	480 (3)	Partial	None	1 mo.
			0.7 x 0.5	480 (1)	Partial	None	1 mo.
11	Sebaceous gland sarcoma	Face	3 x 3 x 2	900 (1)			
				1440 (1)	Partial	None	2 wk
			4 x 3.5 x 2[k]				
			4 x 3.5 x 2[k]	1619 (4)	Complete	None	3 mos.

[a] Time interval between the most recent treatment and examination.
[b] Numbers in parentheses, number of areas.
[c] Complete, no palpable or radiologically evident tumor at time of follow-up; partial, 50% or less of the original lesion remained at follow-up; none, more than 50% of original tumor volume remained.
[d] One month after first treatment.
[e] Six weeks after first treatment.
[f] Three weeks after first treatment.
[g] Two and one-half weeks after first treatment.
[h] A recurrent lesion, approximately 3 x 2 cm, was excised 2 weeks prior to the third photoradiation treatment.
[i] Three months after first treatment.
[j] New lesions grew adjacent to the treated areas.
[k] Five weeks after first treatment.

had been inserted into the desired location in the tumor mass. In most cases, no attempt was made to push the fiber beyond the point of resistance in the tissues. The needle was left in place to provide support for the fiber. In general, if bleeding occurred through the needle, the placement was repeated, since it was found that blood would frequently clot and occlude light from the fiber. If the tumor exceeded 2 cm in diameter, the fiber was implanted in several areas sequentially, placed approximately 1.5 to 2.0 cm apart. Treatment times varied from 20 to 30 min at 120 to 900 milliwatts. Therefore, the light doses delivered to the tumors ranged from 144 to 1620 J (Table 1). At the start of treatment, the power was turned up gradually, and the light emanating from the surface was observed to ensure that it increased proportionally. In cases where there was blood near the fiber tip, a sudden drop of light output was apparent as the applied power increased, indicating the necessity of repositioning the fiber. However, even in these cases, this did not occur below 200 to 250 milliwatts.

None of the animals received any cancer therapy following photoradiation, other than surgery, as indicated in one case.

In cases where the tumor was visible, moderate to intense HPD fluorescence was apparent when illuminated with a Blak-Ray lamp (Ultra-Violet Products, Inc., San Gabriel, Calif).

RESULTS

Table 1 summarizes results of 26 treatments of 7 histological types of malignant lesions in 11 animals (8 dogs and 3 cats). In all cases but one, the lesions were primary. Positive diagnoses in all cases were made histologically from biopsy sections. Of the 14 lesions treated (Animals 6 and 10 had 2 and 3 distinct lesions, respectively), only (2) (Animals 3 and 4), large osteogenic sarcoma tumors, were not responsive to photoradiation. In the 11 animals, there were 5 complete responses and 4 partial responses, and 2 were nonresponsive, with follow-up ranging from 2 to 34 months. Three of these animals can be considered to have been cured of local disease (1 year or more without local recurrence). Another has been 8 months without recurrence. One animal (Animal 6) died of metastatic melanoma to the lungs approximately 1 year after photoradiation treatment. There was no recurrent disease at the primary local site at autopsy.

The squamous cell lesions were found to be highly responsive to treatment (*i.e.*, rapid tumor necrosis and drainage) but progressed rapidly, initially in areas at the periphery of the treatment field and then engulfing the treatment field as well, within 2 to 3 weeks after treatment. While these lesions were retreated, the same pattern was observed in each case, eventually requiring the sacrifice of the animal. It was also noted that in both cases the squamous cell tumors were darkly pigmented, a situation making light penetration difficult, as we have observed previously (3), and this difficulty was also encountered in one of the melanoma lesions treated in Animal 6. While the nonpigmented lesion in Animal 6 was eradicated after the second treatment, the melanotic lesion recurred approximately 2 months after the second treatment. In this case, the lesion was excised as completely as possible, 2 weeks later, the residual lesion, now only 0.5 x 0.5 cm, was treated by photoradiation. Approximately 1 year after the last treatment, the animal was sacrificed due to metastatic lung lesions (malignant melanoma confirmed at autopsy). There was no residual disease at the sites treated by photoradiation.

Side effects of treatment were minimal. One animal experienced facial edema when it was exposed to sunlight shortly after receiving the HPD. The animal was treated with antihistamines, and the edema subsided in 3 to 4 hr.

There was essentially no normal tissue damage due to treatment. One animal had slight erythema of skin overlying the treated lesion, and another had skin sloughing over the lesion. Healing was uneventful. Aside from these minor effects, normal tissues were not affected. In the case of the osteosarcoma lesions which were eradicated, the bones remodeled over a period of 2 to 4 months after treatment, indicating that photoradiation did not destroy the normal bone-forming elements in the bone surrounding the lesions.

DISCUSSION

This report is the first indication that photoradiation therapy with HPD can permanently eradicate remote, primary, life-threatening malignant lesions. Of particular significance to the human situation is the permanent control of osteogenic sarcoma lesions. Further, it should be noted that the interstitial application of the activating light has all but eliminated normal tissue damage, the limiting factor in photoradiation as applied until now (3, 4). In the controlled osteogenic lesions, frequent X-ray examination revealed remodeling of the bones following resolution of the lesions. Therefore, we were able to completely eradicate these lesions with essentially no damage to normal bone-forming elements. This lack of normal tissue damage was seen in essentially all cases.

The ability to effectively deliver light through fiber optics directly to tumor masses not only largely eliminates normal tissue damage but also greatly extends the applicability of photoradiation to virtually any anatomical site reachable by either needles or endoscopes. The fiber can be easily threaded through the biopsy channel of endoscopes and subsequently inserted into various lesions, *e.g.*, in lung or bowel.

The nonresponsiveness of the osteogenic lesions in Animals 3 and 4 is attributed to the large volume at the time of treatment. Since our current system allows the coupling of only one fiber to the laser, it is necessary to treat areas sequentially. Based on the responsiveness of the various tumors (*e.g.*, Animals 1 and 11), it is estimated that 8 to 10 cu cm of tumor volume can be effectively treated from a single insertion of the fiber. Thus, currently very large lesions such as those in Animals 3 and 4 cannot be effectively treated, since the time for the numbers of insertions required would exceed the time that the animal could be safely kept under anesthesia. In order to solve this problem, we are examining a larger dose of HPD (*e.g.*, 10 mg/kg) to increase the effective treatment volume, as well as methods to effectively couple multiple fibers to the laser, thus allowing several areas to be treated concurrently.

The squamous cell lesions presented a difficult situation. Although they were responsive to treatment, new sites adjacent to the original treatment site occurred rapidly (2 to 3 weeks). Apparently, we were not successful in delineating the true extent of the lesions at the time of initial treatment. Also, in both cases the lesions were highly pigmented, thus reducing the effective penetration of the light, an effect also apparent in the pigmented melanoma lesion in Animal 6 and as we have observed earlier in patients (3).

It would be desirable to be able to assay for HPD levels in order to predict tumor responsiveness and more adequately plan the therapy. However, at present, there is no adequate assay for HPD *in vivo*, aside from the use of radioactively labeled material. The cost of this assay in large animals is prohibitive (*e.g.*, Animal 3, one of the animals with a nonresponsive tumor, was a 52-kg St. Bernard dog). While HPD fluorescence frequently has been used to indicate the presence of the drug in tissue, the method is neither quantitative nor accurate, as we have shown previously (5). Tissue pigmentation and blood are highly variable in tumors and other tissues, and both affect penetration of the light used for fluorescence activation (blue) as well as the fluorescent emission (red). We have noted recently a remarkable correlation between ^{67}Ga and HPD toward tissue distribution and retention in both normal and malignant tissues (1) and are investigating ^{67}Ga scanning as a predictor of HPD uptake and/or retention.

The techniques described in this report are currently being applied to various types of malignant lesions in humans *e.g.*,

large bulky lesions not amenable to excision or other therapy, lesions in the brain,[3] lung,[3,4] and vagina.[3]

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