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Microwave Hyperthermia for Choroidal Melanoma in Rabbits

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Radiation has provided excellent local control rates in choroidal melanoma, but significant impairment in visual acuity has occurred in 30–60% of patients due in part to the development of radiation vasculopathy in the fovea and optic disc. Hyperthermia has been shown to have a synergistic effect when added to radiation therapy in human malignancies. The use of hyperthermia in ocular melanoma may allow a reduction in the total radiation dose necessary to achieve local control. A 2450-MHz microwave plaque applicator with integral surface cooling was used to deliver hyperthermia treatments to rabbit eyes containing choroidal melanomas. Extensive thermal mapping was done in acute eyes. In 18 survival animals, a single 23-G needle thermocouple probe with three sensors was inserted into the tumor. Target temperatures of 41.0–46.0°C were maintained for 1 hour. All tumor-bearing eyes were followed for 1 month after treatment, or until tumor growth was noted, with serial ultrasound measurements and visual examinations. A 92% response rate was obtained in tumors treated at temperatures greater than 43.0°C for 1 hour with no significant toxicity. Heat alone has significant tumoricidal properties in this animal model. Invest Ophthalmol Vis Sci 31:1754–1760, 1990

Uveal melanoma is the most common primary intraocular malignancy.[1,2] Radiation has produced excellent local control and survival, but there may be a subsequent development of many ocular complications.[1,3–8] Melanomas have been treated with 50–90 Gy to the tumor apex. Since as many as 50% of uveal melanomas were located within 3 mm of the optic nerve or fovea,[1,8] brachytherapy and heavy-ion approaches may deliver substantial radiation doses to these visually critical structures.[3,5–7,9–11] Visual deterioration, variously defined as decline to less than 20/400 or a decline of two lines or greater on the Snellen visual acuity chart,[1,7,8] is seen in 33–60% of patients followed at least 5 years after treatment.[1] Doses to these structures have declined through the use of ^{125}I with gold rim plaques for shielding,[10–13] but visual deterioration continues to be seen in 38% of patients with posteriorly located tumors treated with radioactive iodine and followed for a mean duration of 45–48 months.[7,8] The amount of visual loss was partially dependent on the foveal and optic nerve dose with secondary development of radiation vasculopathy in these structures.[1,7,8]

Hyperthermia has been shown to have a synergistic effect with radiation in the treatment of human tumors.[14–17] It is possible that hyperthermia combined with a lower radiation dose will preserve useful vision without compromising overall survival or local tumor control.[14,18–21] Microwave radiation, with its limited penetration, is ideally suited for uveal melanoma, because of the tumor size and location immediately beneath the avascular scleral surface. A 2450-MHz microwave antenna with a built-in surface cooling system has been designed at the University of California at San Francisco and incorporated in the plaques currently used to deliver radiation treatment with ^{125}I. This plaque has been used in the treatment of Greene melanoma implanted suprachoroidally in rabbit eyes to study heating uniformity, to examine acute and delayed histologic effects of heat on both tumor and normal tissues, and to determine the efficacy of heat alone in the eradication of melanoma in this animal model.

Materials and Methods

Antenna Design

A 2450-MHz microwave plaque applicator with built-in surface cooling system was used for all hyperthermia treatments (Fig. 1). The antenna was a loop of copper tubing soldered to the inner conductor of a

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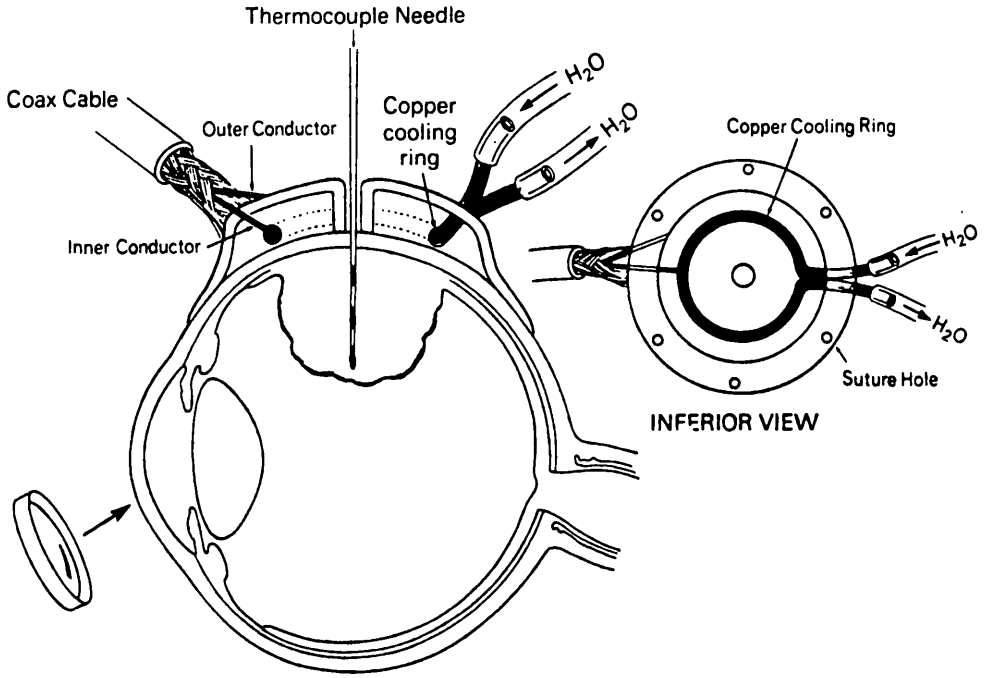

Fig. 1. Inferior view of microwave plaque antenna and cross-sectional view of plaque in place over tumor in the eye. Single thermocouple is depicted passing through the plaque and into the tumor.

coaxial cable. The outer conductor of the cable was grounded to the back of a copper plaque similar to the dummy carrier plaques used in ^{125}I therapy for uveal melanomas in humans. The loop was embedded in a silicone resin 1 mm equidistant from the interior of the plaque bowl. Temperature-controlled water was circulated through the copper loop to cool the scleral surface. A 2450-MHz microwave generator supplied power to the antenna, and a double-stub tuner was used to optimize impedance matching between the antenna and the tissue surface.[22]

Animal Model

We used 3-kg, male albino New Zealand rabbits in the experiments, after approval of the protocol by the University of California, San Francisco, Animal Care Committee. The study was conducted in accordance with the ARVO Resolution on the Use of Animals in Research. Greene melanoma cells were implanted suprachoroidally in the equatorial region of the eye and allowed to grow for 3–4 weeks to 4.5–9 mm in maximum base diameter and 3–8 mm in height, as measured by ultrasound and indirect ophthalmoscopy.

Heating Method

At the beginning of each heat treatment, a conjunctival flap was elevated from the sclera, and the thermometry system was implanted as described. The eyelids were initially kept out of the treatment field using a metal speculum. In later experiments sutures were used instead to minimize thermal lid damage. The microwave plaque antenna was sutured over the tumor after localization with direct visualization and transillumination. Baseline temperatures were recorded, and the power and cooling fluid pump were turned on simultaneously. Once the desired temperature was achieved (5–7 min), it was maintained for 1 hr by varying the power level (2–7 Watts).

Thermometry Techniques

Extensive thermal dosimetry trials were done in acute animals to define the heating pattern achieved in tumor-bearing eyes using the antenna. In four such tumor-bearing eyes, three 19-G teflon catheters were inserted through the tumors at depths of 1, 3–4, and 6–7 mm beneath the plaque–sclera interface. A fourth catheter was inserted perpendicular to the previous three, through the plaque and into the tumor and vitreous to a depth of 12 mm. Single-sensor fiberoptic temperature probes were inserted into these catheters and temperatures monitored during treatment. Prior experiments using this plaque and phantom material did not reveal the presence of hot or cold spots beneath the antenna. Orientation of the catheters beneath the plaque did not affect the temperature distribution. At 10-min intervals during heating, the sensors were moved at 2-mm increments within the catheters and readings recorded. The rabbits were euthanized at the completion of each heating while still under anesthesia.

In the survival animals, a single 23-G needle thermocouple was inserted through the back of the

plaque and into the eye. The distal sensor of the triple-sensor thermocouple was located 2 mm from the tip of the needle. The needle was inserted so that the distal sensor was finally located 3 mm from the tumor apex. This depth was chosen to avoid penetration of the needle beyond the tumor into the vitreous. All temperature measurements were made using a power-off method to minimize artifacts arising from the use of a metal thermocouple in an electromagnetic field.[23] The power and cooling liquid were shut off simultaneously and the temperature recorded 2 sec later. Cooling fluid was maintained at 18–20°C using a temperature-controlled reservoir.

Acute Studies

The effect of water cooling was studied in eight eyes. Two tumor-bearing eyes were heated with microwave, with circulating water temperatures of 0°, 18°, and 23.5°C, versus no water cooling. Six extensive mapping trials were then done, one per animal, using 18–23°C water for surface cooling plus microwave. Comparison trials using thermocouples and fiberoptic probes were described in a previous report.[23]

Survival Studies

Eighteen tumor-bearing eyes were treated with 1 hr of hyperthermia using a triple-sensor thermocouple for thermometry. Temperatures were recorded at 5-min intervals. The rabbits were examined for evidence of damage and tumor response immediately after the hyperthermia, and at weekly intervals for 1 month or until tumor progression was seen, whichever was shorter. All eyes were then enucleated and examined histologically.

Complete regression (CR) of the tumor was defined as visible evidence of scar only, with no viable cells identified histologically at the time of enucleation. A partial response (PR) was defined as reduction in tumor size to less than or equal to one-half the original tumor dimensions, with some viable cells appearing on histologic examination. No response (NR) was defined as a less than 50% reduction in size of tumor or progression of growth.

Results

Temperature-Distribution Studies

The comparison of heating patterns measured along the central tumor axis using different cooling temperatures during microwave heating is illustrated in Figure 2. When no coolant was circulated through the loop, temperatures diminished by 1.5°C/mm from the surface. A 10°C gradient existed between the surface and the tissue at a depth of 7 mm. Coolant temperatures approaching 0°C resulted in excessive surface cooling and necessitated increasing power levels to achieve temperatures greater than 42.0° at 5 mm depth. Coolant temperatures of 18–20°C resulted in temperatures in the 42.5–45°C range from 1–9 mm depth. This coolant temperature was therefore chosen for the survival animal trials.

Figure 3 is a composite of the thermal distributions obtained in four separate tumor-bearing eyes under

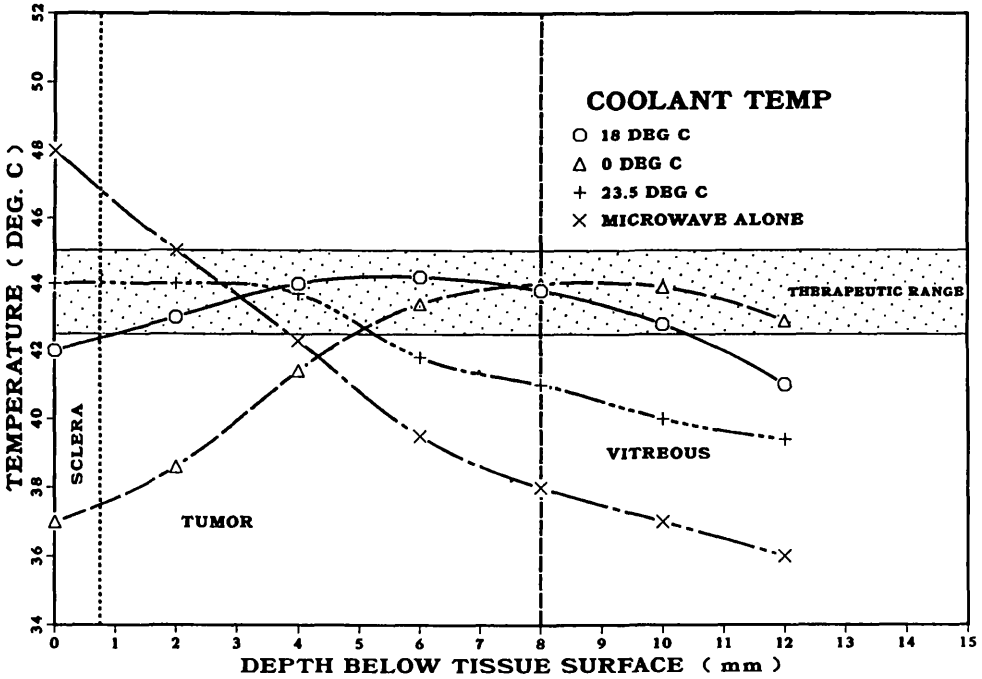

Fig. 2. Microwave heating temperature distribution in rabbit eye tumor measured along the central tumor axis, with different circulating coolant temperatures.

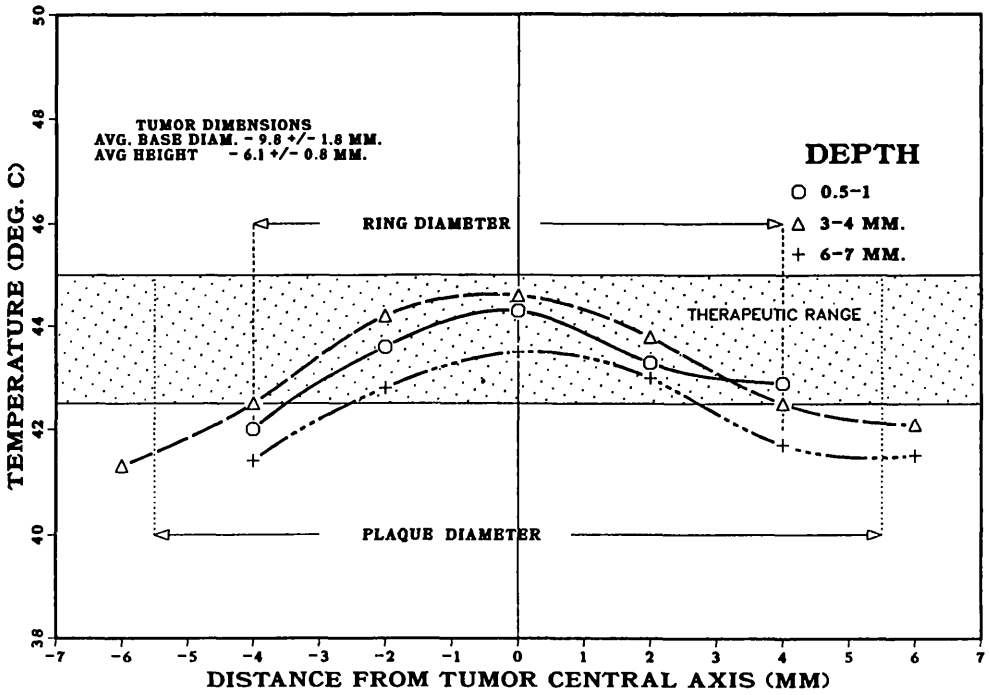

Fig. 3. Composite temperature distributions of four eyes treated under similar conditions of microwave power and coolant temperatures. Readings were recorded with fiberoptic sensors at 2 mm increments along the paths of three catheters parallel to the plaque-sclera interface.

conditions of similar applied power and circulating coolant. Tumor dimensions were closely matched, with base diameters of 8–11 mm and tumor heights of 5–7 mm. Assuming symmetry of heating due in part to uniform blood perfusion of the intraocular tumor volume, a conical volume with base diameter of 8 mm, reducing to a diameter of 5 mm at a depth of 6–7 mm was entirely within the therapeutic range of 42.5–45°C, with no measured temperatures exceeding 45°C in the tumor, sclera, or vitreous. Additional probes placed adjacent to the posterior capsule of the lens in three of the trials revealed peak temperatures below 40.5°C.

Survival Animal Treatments

Tumor response to treatment with temperatures at or below 43°C versus temperatures above 43°C (measured at a reference point 3 mm beneath the tumor apex on the central tumor axis) for all animals is displayed in Figure 4. Response as a function of maximum base diameter is shown in Figure 5 and as a function of temperature, in all tumors ≤ 8 mm in height, in Figure 6. Overall, 10 of 18 eyes showed a CR, 3 of 18 had a PR, and 5 had NR. Three of the NR tumors were treated with temperatures ≤ 42°C. Temperatures ≥ 43.5°C resulted in CR plus PR in 11 of

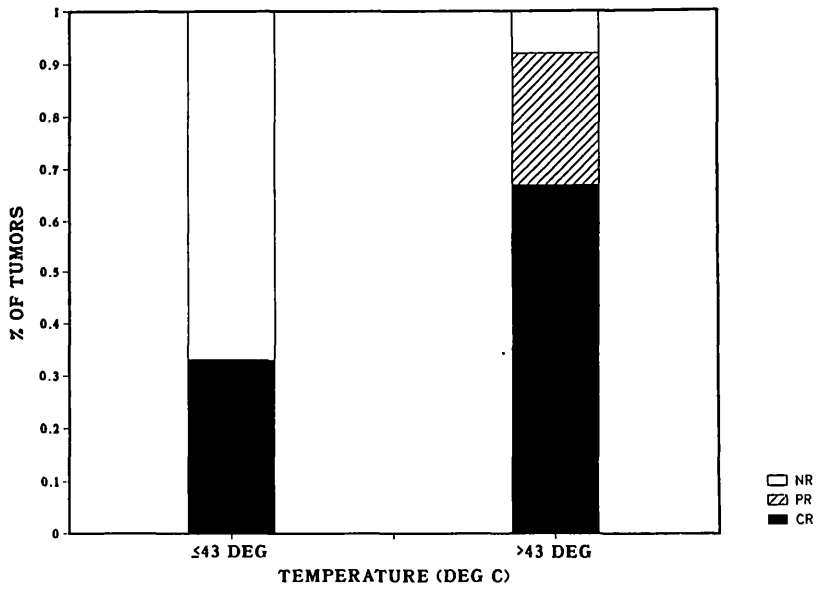

Fig. 4. Tumor response versus temperature in all 18 survival eyes.

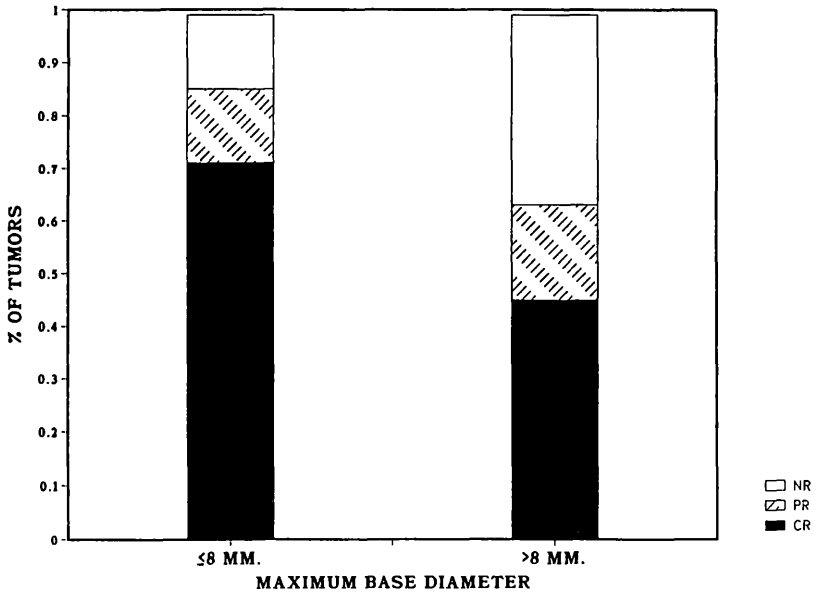

Fig. 5. Tumor response versus maximum base diameter of tumor.

12 tumors. Temperatures ≤ 43°C resulted in NR in four of six animals.

A CR plus PR was achieved in six of seven tumors with a maximum base diameter < 8 mm. Seven of 11 tumors ≥ 8 mm base in diameter had a CR or PR. Tumors ≤ 8 mm in height achieved a CR or PR in 11 of 12 eyes heated at > 43°.

Acute toxicity was minimal. In the first two rabbits studied, thermal burns occurred in the upper eyelid, due to the use of the metal speculum. In subsequent animals, the metal speculum was no longer used, and no further burns were seen. No scleral or corneal damage or cataract formation were noted. Histologic examination revealed no evidence of retinal detachment, scleral thinning, or vitreous hemorrhage.

Discussion

Although excellent uveal-melanoma control rates occur with current radiotherapeutic approaches, significant ocular complications occur.[1,3,6–8] Radiation vasculopathy, a dose-dependent, gradually progressive occlusive disorder of the microvasculature, is a major cause of visual loss.[1] The radiation dose currently used to obtain local control is 60–90 Gy to the tumor apex, although minimal dose-response relationship has been defined for this tumor. Given the exponential fall-off of dose rate with distance from the brachytherapy sources, doses as high as 400 Gy may be delivered to the tumor base of large tumors and up to 300 Gy to the adjacent retina. The use of

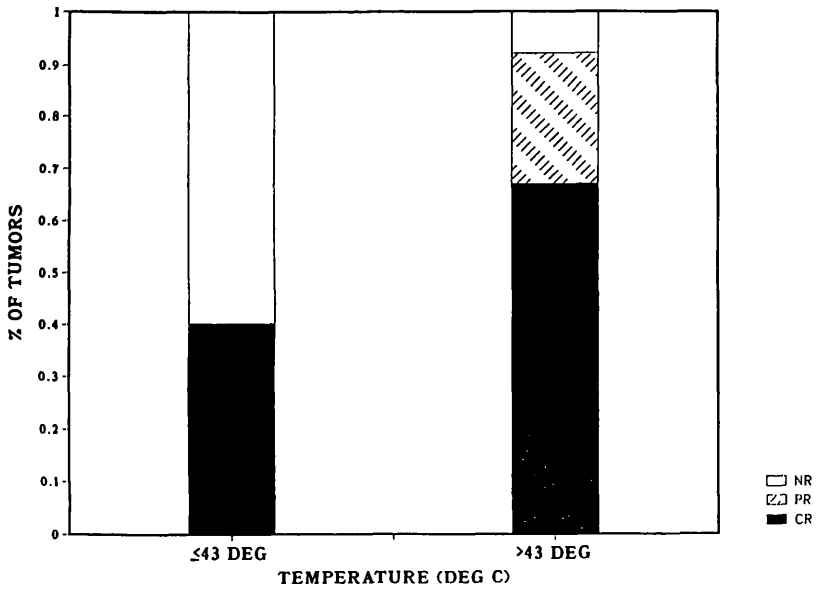

Fig. 6. Response in tumors less than 8 mm height versus temperature.

^{125}I has allowed for a significant reduction in dose delivered to portions of the eye not directly beneath the plaque due to the improved collimation and shielding provided by the gold-rim plaque and the low energy of the photon submitted by the ^{125}I compared with ^{60}Co.[10-13] Garretson et al[7] reported a 55% incidence of visual deterioration in 18 tumors located within three disc diameters of the fovea or optic nerve. Bosworth et al[8] reported a 39% incidence of decreased vision. In both studies patients were followed for an average of only 4 years. As follow-up lengthens, these rates may also increase. Stallard[24] reported on the treatment of 107 choroidal melanomas with ^{60}Co plaques, and visual acuity remained better than 20/60 in only 34 of 107 patients treated with doses of 80–100 Gy to the tumor apices.[9,24] Lommsatzsch,[5] using the Ru/Rh-106 applicator to deliver similar doses, reported that only 26% retained good visual acuity after treatment. Cruess et al[3] demonstrated that a dose in excess of 50 Gy to the fovea or disc would result in visual acuity worse than 20/200 in 35–48% of patients. Charged-particle irradiation has the advantage of a uniform dose delivery to the entire tumor, but decreased visual acuity is still seen in a significant proportion of patients with posteriorly located tumors.[2,4,6]

To reduce the radiation dose without compromising local control rates, hyperthermia is being tested as an adjuvant therapy in uveal melanoma by several groups.[8,19,21] Heat augments the effectiveness of radiation in many human cancer histologies including melanoma.[14,15-17] The mechanisms of heat-induced activity include direct cytocidal damage at the molecular and cellular levels,[25,26] preferential disruption of tumor vasculature,[27,28] and radiosensitization through interference with sublethal damage repair.[25,29] Given the proximity of choroidal melanoma to the scleral surface and fragile vasculature, it is ideal for adjuvant therapy with superficial hyperthermia.

Proton irradiation combined with ultrasound hyperthermia was used by Riedel et al[21] in the treatment of rabbit eye tumors measuring up to 4.5 × 4.5 × 4 mm in size. Temperatures and radiation doses, each shown to be subtherapeutic when employed singly, were combined to give excellent local control rates. Burgess et al[18] reported on the use of ultrasound hyperthermia, heating intraocular tumors to 43–65°C for various time intervals, to obtain a response in 15 of 23 rabbits. Finger and co-workers[19] used microwave hyperthermia and ^{125}I radiotherapy, alone and in combination, in 11 rabbit eye tumors measuring up to 6 × 6 × 5 mm. Surface temperatures ≥ 48°C were necessary to achieve temperatures of 43°C at 5 mm depth due to the characteristic drop-off energy deposition seen with microwave use. In a separate report from the same institution on the use of a combined thermoradiotherapy plaque in six humans, no toxicity was reported.[20]

The microwave antenna with a built-in cooling system described in this report maintains surface temperatures at levels compatible with minimal toxicity and elevates the entire tumor (up to 8 mm maximum base diameter and 9 mm in height) to therapeutic temperatures. This latter point is crucial, because the rate and duration of tumor response to heat, when used with radiation, is directly proportional to the minimum tumor temperature.[30]

One of the major drawbacks in current hyperthermia technology is the need for invasive thermometry. Due to the diversity in vascularity of tumors and their physiologic environs, temperatures throughout a tumor can vary greatly. Surface temperatures are poor predictors of intratumoral hot and cold spots. Concern over possible metastases, autoimmunization, trauma, and infection make it imperative to minimize invasive thermometry. Although fiberoptic monitoring is the most accurate thermometry system available for use in microwave heating, the larger gauge of these probes makes them unsuitable for use in the eye. Thermocouples can be built into 29-G needles. There is no evidence of tumor seeding after fine-needle aspiration in any tumors with a 24-G or finer needle.[31] The lack of evidence for tumor seeding with narrow-gauge needles and the low morbidity seen with fine-needle aspiration in human subjects with choroidal melanoma leads us to believe that intratumoral temperatures can be safely monitored using 29-G thermocouples.

This study demonstrates that microwave-induced heating alone, delivered with a 2450-MHz frequency applicator with a built-in cooling system, can have a significant impact on the growth of Greene melanoma in rabbit eyes. We report a 92% CR + PR rate in tumors up to 8 × 8 × 8 mm treated with reference temperatures above 43°C maintained for 60 minutes without the development of significant short-term toxicity. Based on the experience of the use of heat and radiation in nonocular melanoma,[14] it is reasonable to expect that use of adjuvant intraocular hyperthermia may allow us to reduce the dose of radiation needed to provide local control rates comparable to those attainable with radiation alone.

Key words: uveal melanoma, hyperthermia, microwave, choroidal tumor

References

1. Char DH: Therapeutic options in uveal melanoma. Am J Ophthalmol 98:796, 1984.
2. Gragoudas ES and Seddon JM: Management of choroidal melanomas. Int Ophthalmol Clin 26:145, 1986.

3. Cruess AF, Augsburger JJ, Shields JA, Donoso LA, and Amsel J: Visual results following cobalt plaque radiotherapy for posterior uveal melanomas. Ophthalmology 91:131, 1984.
4. Gragoudas ES, Goitein M, Verhey L, Munzenreider J, Urie M, Suit H, and Koehler A: Proton beam irradiation of uveal melanomas: Results of 5½-year study. Arch Ophthalmol 100:928, 1982.
5. Lommsatzsch PK: Beta-irradiation of choroidal melanoma with $^{106}Ru/^{106}Rh$ applicators. Arch Ophthalmol 101:713, 1983.
6. Seddon JM, Gragoudas ES, Egan KM, Glynn RJ, Munzenrider JE, Austin-Seymour M, Goitein M, Verhey L, Urie M, and Koehler A: Uveal melanomas near the optic disc or fovea: Visual results after proton beam irradiation. Ophthalmology 94:354, 1987.
7. Garretson BR, Robertson DM, and Earle JD: Choroidal melanoma treatment with iodine 125 brachytherapy. Arch Ophthalmol 105:1394, 1987.
8. Bosworth JL, Packer S, Rotman M, Ho T, and Finger P: Choroidal melanoma: I-125 plaque therapy. Radiology 169:249, 1988.
9. Char DH, Lonn LI, and Margolis LW: Complications of cobalt plaque therapy of choroidal melanomas. Am J Ophthalmol 84:536, 1977.
10. Chiu-Tsao ST, O'Brien K, Sanna R, Tsao HS, Vialotti C, Chang YS, Rotman M, and Packer S: Monte Carlo dosimetry for ^{125}I and ^{60}Co in eye plaque therapy. Med Phys 13:678, 1986.
11. Wu A, Sternick ES, and Muise DJ: Effect of gold shielding on the dosimetry of an ^{125}I seed at close range. Med Phys 15:627, 1988.
12. Packer S, Fairchild RG, and Salanitro P: New techniques for iodine-125 radiotherapy of intraocular tumors. Ann Ophthalmol 19:26, 1987.
13. Earle J, Kline RW, and Roberston DM: Selection of iodine-125 for the collaborative ocular melanoma study. Arch Ophthalmol 105:763, 1987.
14. Overgaard J: The role of radiotherapy in recurrent and metastatic malignant melanoma: A clinical radiobiologic study. Int J Radiat Oncol Biol Phys 12:867, 1986.
15. Arcangeli G, Cividalli A, Nervi C, Creton G, Lovisolo G, and Mauro F: Tumor control and therapeutic gain with different schedules of combined radiotherapy and local external hyperthermia in human cancer. Int J Radiat Oncol Biol Phys 9:1125, 1983.
16. Kim JH, Hahn EW, and Ahmed SA: Combination hyperthermia and radiation therapy for malignant melanoma. Cancer 50:478, 1982.
17. Perez CA, Nussbaum G, Emami B, and VonGerichten D: Clinical results of irradiation combined with local hyperthermia. Cancer 52:1597, 1983.
18. Burgess SEP, Chang S, Svitra P, Driller J, Lizzi FL, and Coleman J: Effect of hyperthermia on experimental choroidal melanoma. Br J Ophthalmol 69:854, 1985.
19. Finger PT, Packer S, Svitra P, Paglione RW, Anderson LL, Kim JH, and Jakobiec FA: Thermoradiotherapy for intraocular tumors. Arch Ophthalmol 103:1574, 1985.
20. Finger PT, Packer S, Paglione RW, Cratz JF, Ho TK, and Bosworth JL: An interstitial thermoradiotherapy plaque technique for treatment of malignant melanoma of the choroid. Int J Radiat Oncol Biol Phys 13(Suppl):139, 1987.
21. Riedel KG, Svitra PP, Seddon JM, Albert DM, Gragoudas ES, Koehler AM, Coleman DJ, Torpey J, Lizzi FL, and Driller J: Proton beam irradiation and hyperthermia: Effects on experimental choroidal melanoma. Arch Ophthalmol 103:1862, 1985.
22. Stauffer PR, Swift PS, Sneed PK, Char DH, and Phillips TL: Temperature controlled microwave ring radiator for hyperthermia therapy. Proc IEEE Engineering in Medicine and Biology Society Meeting, New Orleans, Louisiana Nov. 1988, pp. 1273–1274.
23. Swift PS, Stauffer PR, Fries PD, Sneed PK, Phillips TL, and Char DH: A 2450 MHz microwave antenna plaque used in choroidal melanoma model in rabbits. J Endocurie Therapy/Hyperthermia 1988 (in press).
24. Stallard HB: Malignant melanoblastoma of the choroid. Mod Probl Ophthalmol 7:16, 1968.
25. Harisidias L, Sung D 2d, Kessaris N, Hall EJ, and Phil D: Hyperthermia: Biological studies at the cellular level. Radiology 129:195, 1978.
26. Overgaard J: Histopathologic effects of hyperthermia. *In* Hyperthermia in Cancer Therapy, Storm FK, editor. Boston, GK Hall, 1983, pp. 163–185.
27. Eddy HA: Alterations in tumor vasculature during hyperthermia. Radiology 137:515, 1980.
28. Song C: Blood flow in tumors and normal tissues. *In* Hyperthermia in Cancer Therapy, Storm FK, editor. Boston, GK Hall, 1983, pp. 187–206.
29. Ben-Hur E, Elkind MM, and Bronk BV: Thermally enhanced radioresponse of cultured Chinese hamster ovary cells: Inhibition of repair of sublethal damage and enhancement of lethal damage. Radiat Res 58:38, 1974.
30. Dewhirst MW and Sim DA: The utility of thermal dose as a predictor of tumor and normal tissue responses to combined radiation and hyperthermia. Cancer Res 44(Suppl 10):4772s, 1984.
31. Char DH, Miller TR, Ljung B-M, Howes EL Jr, and Stoloff A: Fine needle aspiration biopsy in uveal melanoma. Acta Cytol 33:599, 1989.