Effects of Local Tumor Hyperthermia on the Growth of Solid Mouse Tumors

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ABSTRACT

The sensitivity to local tumor hyperthermia (43°, 60 min) of a spectrum of eight different solid mouse tumors (Lewis lung carcinoma, M5076 ovarian carcinoma, colon carcinoma 38, colon carcinoma 26, mammary adenocarcinoma C3HBA, mammary adenocarcinoma 16C, glioma 26, and B16 melanoma) was investigated. A microwave (2.45-GHz) apparatus produced localized heating of the tumors without generation of whole-body hyperthermia. The temperature at the center of the heated tumors was regulated to within ±0.1° while the temperature uniformity within the tumor was ±0.5°.

The local hyperthermia treatments reduced the size and retarded the growth of the treated tumors compared with control values for each of the tumors tested. The faster-growing Lewis lung carcinoma and B16 melanoma were the least responsive to treatment, while the slower-growing colon 38 and M5076 ovarian carcinomas were the most responsive. Multiple treatments resulted in longer growth delays and greater tumor growth inhibition than did single treatments. No consistent difference in life span between the control and treated groups was measured, and only five of 188 treated animals were cured.

INTRODUCTION

The use of local hyperthermia as a cancer treatment modality is not well established. Recent reviews have summarized the wide variety of local hyperthermia methods which have been applied to animal and human tumors. Differences in heating methods, treatment temperature, duration of heating, and treatment schedule make it difficult to compare the results of different investigators. In addition, only a few recent investigators have tested more than one tumor type under similar in vivo hyperthermia conditions. Three temperature ranges for LTH treatments (mild, 37–41°; moderate, 42–44°; marked, >45°) have been established. Mild LTH has been generally found to be ineffective in treating tumors and may in some cases stimulate metastasis. Moderate LTH is currently under intensive study due to the difference in temperature sensitivity between cancer cells and normal cells originating from intrinsic (14) and tumor environmental factors (24) such as acidity, hypoxia, and poor nutrition. Marked LTH has received little attention due to the problems of damage to surrounding normal tissues, but technological advances in local heating methods are improving treatment capabilities (9).

Previous investigators using moderate LTH alone against experimental animal tumors in vivo have demonstrated a wide range of tumor responses. In work spanning 50 years (5, 7, 8, 10, 18, 21–23, 25, 26, 30, 33), some animal tumors have been found to be relatively heat sensitive with 20 to 100% cures obtained using moderate LTH alone. An inverse relationship between the treatment temperature and the duration of heating required to cure tumors is evident from these studies. Above 42°, the effective treatment times decreased by a factor of 2 for each 1° increase in temperature (24). In contrast to these promising results, those of other researchers (32, 34–36) using similar moderate LTH conditions have demonstrated only TGI with few cures. There are also conflicting observations (7, 27, 35) that LTH stimulates the metastasis of tumors.

In an effort to assay tumor sensitivity to hyperthermia, a spectrum of 8 different solid mouse tumors was treated under carefully controlled conditions of moderate local hyperthermia. These tumors were selected for study because their response to drug therapy has been well established (1–3, 15), and future studies will be directed toward the combination of LTH with systemic drug treatments.

MATERIALS AND METHODS

Animals and Tumors. C3H/He, BALB/c × DBA/2F1 (hereafter called CD2F1), and C57BL/6 × DBA/2F1 (hereafter called B6D2F1) mice were obtained from the Mammalian Genetics and Animal Production Section, National Cancer Institute. Groups of 8 to 10 mice were housed in plastic cages and were given pelleted feed and water ad libitum.

Eight different mouse tumors were used in these experiments: Lewis lung carcinoma; B16 melanoma; glioma 26; M5076 ovarian carcinoma; colon adenocarcinoma 38; colon adenocarcinoma 26; mammary adenocarcinoma C3HBA; and mammary adenocarcinoma 16C. The tumors were obtained from the tumor repository of the Mammalian Genetics and Animal Production Section, National Cancer Institute. Standardized protocols of the Developmental Therapeutics Program, National Cancer Institute (12), were followed for continuous passage of the tumors in syngeneic mice.

Each tumor was maintained in the mouse strain of its origin and transplanted into C3H/He, CD2F1, or B6D2F1 mice for experimentation. The mammary and ovarian tumors were always transplanted into female mice, while the other tumors were transplanted into mice of either sex. Experimental tumors were implanted s.c. on the left flank just anterior to the rear leg with inocula of the following composition: Lewis lung, 10⁶ viable cells in 0.2 ml (B6D2F1); B16 melanoma, 0.25 ml of a 1:5 (w/v) tumor brei (B6D2F1); glioma 26, 0.2 ml of a 1:5 (w/v) tumor brei (B6D2F1); mammary 16C, 0.2 ml of a 1:5 (w/v) tumor brei (C3H/He); mammary C3HBA, 0.2 ml of a 1:5 (w/v) tumor brei (C3H/He); colon 26, 0.2 ml of a 1:25 (w/v) tumor brei (CD2F1);
colons, 0.2 ml of a 1:1 (w/v) tumor brei (B6D2F1); M5076 ovaries, 0.2 ml of a 1:1 (w/v) tumor brei (B6D2F1).

Microwave Hyperthermia Apparatus. A microwave system was utilized to provide controlled, localized hyperthermia in the tumors of the experimental animals. The system allowed the simultaneous treatment of 4 separate tumors. A complete description of the design and construction of this apparatus has been published (19), so only a brief characterization of the system and its operation will be presented. Chart 1 is a block diagram of the major components of the system, which consists of a 2.45-GHz microwave source, a 4-way power-dividing network and reflected power monitor, a temperature-controlled microwave power regulator, and small direct-contact microwave applicators. Adjustment of the temperature control results in elevated temperatures in the centers of tumors which could be maintained to within ±0.1° without the production of significant whole-body hyperthermia. The temperatures within the locally heated tumors (±3 mm from the center) were found to vary within ±0.5° of the center temperature. The temperature distribution measured in these tumors was: top, 43.5°; center, 43.0°; bottom and edges, 42.5°.

The microwaves were localized to the area of the tumor by using 3.2-cm-diameter microwave diathermy applicators (AT-502/7Z; ELMED Inc., Addison, Ill.). The applicator was efficiently coupled to the tumor by enclosing the tumor in a bolus of muscle-equivalent dielectric material (16). The encapsulation of the tumor in a bolus reduces the temperature gradients within the tumor and increases the efficiency of the applicator to deliver power to the tumor.

The intratumor temperature was measured using a copper-constantan thermocouple formed from 0.13-mm-diameter insulated wire. The thermocouple junction was placed in the tumor by passing a 25-gauge needle through the tumor and pushing the thermocouple wire through the lumen of the needle. The needle was removed, and the junction was positioned at the center of the tumor. The microwave applicator, tumor, and thermocouple were adjusted so that the maximum in the heating distribution from the applicator (20) corresponded with the center of the tumor. Previous studies (19) showed that this procedure reliably measures the temperature of the tumor. The rectal temperature was measured using a Teflon-coated copper-constantan thermometer probe (IT-1; Bailey Instruments Co., Inc., Saddle Brook, N. J.) which was inserted 1 cm. Both the intratumor and rectal thermocouples were connected to digital electronic thermometers (BAT8C; Bailey Instruments) which were periodically calibrated against an ASTM 91C liquid-in-glass thermometer.

Experimental Protocol. Two groups of 8 mice each were sorted by tumor size (50 to 250 cu mm) and body weight (20 to 25 g) approximately 1 week following tumor implantation. On treatment days, both groups were anesthetized with chloral hydrate (Matheson Coleman and Bell, Norwood, Ohio) at an i.p. dose of 525 mg/kg for B6D2F1, mice and 495 mg/kg for C3H/He mice. Both the control and the treatment groups of mice were kept under a 100-watt incandescent lamp to maintain their rectal temperatures in the normal 36°–38° range. The tumor of each control mouse was punctured with a sterile 25-gauge needle. Following anesthesia and before heating, thermocouples were implanted in each of the tumors of the mice in the treatment group, and a 6-g, 35- x 35- x 9-mm bolus was placed over each tumor. The treatment group was subjected to 60 min of LTH which was regulated at the center of the tumor to 43.0 ± 0.1°. In most experiments, the treatment group required an additional 150 mg/kg of chloral hydrate anesthetic half-way through the heating period. The rectal temperatures of both groups were measured before anesthesia and several times during the heating period to check for any whole-body hyperthermia. Both groups of mice were weighed, and their tumors were measured twice each week following treatment. The tumor size was estimated from caliper measurements of tumor length (L) and width (W) using the formula:

\[ Volume = \frac{\pi}{6} \times L \times W^2 \]

RESULTS

The mice tolerated the LTH treatments very well. The effect of the treatment on the body weights of both treated and control groups was minimal. The mice lost weight immediately following treatment (approximately 4% of total body weight) and then regained the lost weight over the next 2 days. No significant differences in this pattern of weight loss and recovery were observed between the treated and control groups.

Heated tumors appeared to be enlarged immediately following treatment. The local swelling was reduced by 24 to 48 hr following treatment. The treated tumors at these times were smaller and firmer than the control tumors. In a few mice, a small surface scab developed at the site of treatment, but in most cases the skin maintained a normal appearance following treatment.

Chart 2 shows the average rectal temperatures for heat and control groups before and after a local hyperthermia treatment. The initial rectal temperatures were elevated from the normal 37° due to handling. Both groups were administered chloral hydrate (525 mg/kg i.p.) at the initial time point. The rectal temperatures in both groups fell due to the anesthesia. During local heat treatment, the average rectal temperatures of the heated animals increased slightly. The control group which was kept under a heat lamp maintained a relatively constant average rectal temperature during the experiment. One hr following the treatment, both groups had similar average rectal temperatures. Table 1 lists the changes in average rectal temperature for both treatment and control groups in a series of 4 LTH treatments. The measurements indicate an average...
when the S.E.'s exceeded 0.1 °. Heated to 43° for 60 min (A). The average temperatures are shown; bars, S.E. were encapsulated in a 6-g bolus of tissue-equivalent dielectric material and during a LTH experiment. Both groups consisted of 8 B6D2F, female mice implanted with M5076 ovarian carcinoma. The tumors of the treated animals were regulated at 43.0°. Although slight increases in distribution in the tumors, the bottom edges were 42.5° when treatment was observed. In analyzing these results, it is important to recognize that, due to the nonuniform temperature distribution in the tumors, the bottom edges were 42.5° when the center was regulated at 43.0°. Although slight increases in life span were observed in several experiments, only 5 of a total of 188 treated animals were cured. None of the control tumors underwent spontaneous regression.

Table 2 is a summary of the results obtained from single and multiple local hyperthermia treatments of the 8 solid mouse tumors investigated. In general, local hyperthermia (43°, 60 min) reduced the size and retarded the growth of treated tumors compared with that of controls for each of the tumors tested. A difference in the sensitivity of the 8 tumors to the treatment was observed. In analyzing these results, it is important to recognize that, due to the nonuniform temperature distribution in the tumors, the bottom edges were 42.5° when the center was regulated at 43.0°. Although slight increases in life span were observed in several experiments, only 5 of a total of 188 treated animals were cured. None of the control tumors underwent spontaneous regression.

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The growth rates for tumors can be characterized by the tumor volume-doubling times. The number of days required for the control and treated tumors to grow from 250 to 500 cu mm and from 500 to 1000 cu mm was determined from their growth curves. For the control tumors, these doubling times indicate the growth rate of the tumor, while for the treated tumors they are an indication of the regrowth rate of the tumor following LTH. In order to compare the sensitivity of the tumors to heat with their growth rates, the doubling times for the tumors were averaged into 3 response groups based on the TGI and data of tumor regrowth was similar to the growth rate of the controls.

Tumor growth delay was defined as the extra number of days needed to reach an average tumor size of 1 cu cm. Using this parameter, the tumors again exhibit a wide range in their sensitivity to treatment. Table 2 lists the growth delay data for all the single and multiple treatment experiments for each of the tumors. The Lewis lung carcinoma and colon carcinoma 26 again appear to be the least responsive to single LTH treatments, with tumor growth delays of about 1 to 2 days, while the colon carcinoma 38 and M5076 ovarian carcinoma were delayed by 6 to 13 days. The other tumors were intermediate in response, with about a 3- to 7-day delay in tumor growth obtained. Multiple LTH treatments increased the response of all tumors.

The median day of death was recorded for both treated and control groups for each of the 8 tumors investigated. These data along with the percentage increase in life span are listed in Table 2. They show no consistent difference between the life spans of the treated and control groups for the early metastasizing colon carcinoma 26, Lewis lung carcinoma, B16 melanoma, and mammary adenocarcinomas 16C and C3HBA. However, the rarely metastasizing glioma 26, colon carcinoma 38, and M5076 ovarian carcinoma all showed an increase in life span of approximately 20% following multiple LTH treatments. With an early and rapidly metastasizing tumor, one would not expect a local treatment to prevent animal deaths from metastatic disease, but for rarely metastasizing tumors delays in the local tumor growth could be expected to translate into an increase in life span. Even though LTH did not significantly increase the life span of the treated animals, it is important to note that local hyperthermia did not consistently reduce life span for any of the tumors studied, as might be expected if tumor metastasis was stimulated.

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Table 1
Whole-body heating during LTH
Average rectal temperatures were measured before and after LTH (43°, 60 min). The experimental group consisted of 8 female B6D2F, mice with M5076 ovarian carcinoma tumors.

<table>
<thead>
<tr>
<th>Treatment day</th>
<th>Group</th>
<th>Before anesthesia</th>
<th>End of 60 min heating</th>
<th>Change in temperature</th>
</tr>
</thead>
<tbody>
<tr>
<td>11</td>
<td>Heat</td>
<td>38.1 ± 0.3*</td>
<td>37.3 ± 0.4*</td>
<td>−0.8*</td>
</tr>
<tr>
<td>Control</td>
<td></td>
<td>37.8 ± 0.3</td>
<td>36.7 ± 0.3</td>
<td>−1.1</td>
</tr>
<tr>
<td>13</td>
<td>Heat</td>
<td>38.5 ± 0.3</td>
<td>37.3 ± 0.5</td>
<td>−1.5</td>
</tr>
<tr>
<td>Control</td>
<td></td>
<td>38.1 ± 0.5</td>
<td>37.6 ± 0.3</td>
<td>−0.5</td>
</tr>
<tr>
<td>15</td>
<td>Heat</td>
<td>38.1 ± 0.5</td>
<td>38.1 ± 0.6</td>
<td>0.0</td>
</tr>
<tr>
<td>Control</td>
<td></td>
<td>38.3 ± 0.7</td>
<td>37.6 ± 0.2</td>
<td>−0.7</td>
</tr>
<tr>
<td>17</td>
<td>Heat</td>
<td>37.2 ± 0.4</td>
<td>37.9 ± 0.7</td>
<td>+0.7</td>
</tr>
<tr>
<td>Control</td>
<td></td>
<td>37.9 ± 0.5</td>
<td>36.2 ± 0.6</td>
<td>−1.7</td>
</tr>
</tbody>
</table>

* Average ± S.D.
Local Hyperthermia of Solid Mouse Tumors

Table 2

Tumor growth delay and mouse survival following LTH treatments

The growth characteristics of each tumor and the survival data were obtained from groups of 8 control and 8 locally heated mice. The mice were treated on the days listed in this table for 60 min at 43°.

<table>
<thead>
<tr>
<th>Experiment no.</th>
<th>Tumor type</th>
<th>Treatment day</th>
<th>Control (volumecu mm)</th>
<th>TGI* (%)</th>
<th>GDDb</th>
<th>T/Cc</th>
<th>% ILSd</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Colon 26</td>
<td>9</td>
<td>1102 ± 74</td>
<td>23</td>
<td>2</td>
<td>32/30</td>
<td>7</td>
</tr>
<tr>
<td>2</td>
<td>Colon 26</td>
<td>9</td>
<td>1485 ± 73</td>
<td>24</td>
<td>6</td>
<td>27/27</td>
<td>0</td>
</tr>
<tr>
<td>3</td>
<td>Lewis lung</td>
<td>8</td>
<td>1423 ± 131</td>
<td>6</td>
<td>1</td>
<td>23/28</td>
<td>-18</td>
</tr>
<tr>
<td>4</td>
<td>Lewis lung</td>
<td>8</td>
<td>1490 ± 138</td>
<td>28</td>
<td>1</td>
<td>27/25</td>
<td>-8</td>
</tr>
<tr>
<td>5</td>
<td>Lewis lung</td>
<td>4, 7, 10</td>
<td>1249 ± 145</td>
<td>46</td>
<td>3</td>
<td>28/26</td>
<td>8</td>
</tr>
<tr>
<td>6</td>
<td>Lewis lung</td>
<td>4, 7, 10</td>
<td>2245 ± 168</td>
<td>37</td>
<td>2</td>
<td>31/28</td>
<td>11</td>
</tr>
<tr>
<td>7</td>
<td>B16 melanoma</td>
<td>9</td>
<td>1213 ± 140</td>
<td>56</td>
<td>3</td>
<td>34/34</td>
<td>0</td>
</tr>
<tr>
<td>8</td>
<td>B16 melanoma</td>
<td>9</td>
<td>1427 ± 109</td>
<td>63</td>
<td>5</td>
<td>26/27</td>
<td>-4</td>
</tr>
<tr>
<td>9</td>
<td>Mammary C3HBA</td>
<td>9</td>
<td>1009 ± 97</td>
<td>38</td>
<td>5</td>
<td>34/41</td>
<td>-17</td>
</tr>
<tr>
<td>10</td>
<td>Mammary C3HBA</td>
<td>10</td>
<td>1296 ± 175</td>
<td>56</td>
<td>5</td>
<td>36/38</td>
<td>-5</td>
</tr>
<tr>
<td>11</td>
<td>Mammary C3HBA</td>
<td>14</td>
<td>1567 ± 281</td>
<td>74</td>
<td>7</td>
<td>48/38</td>
<td>26</td>
</tr>
<tr>
<td>12</td>
<td>Glioma 26</td>
<td>10</td>
<td>1577 ± 179</td>
<td>37</td>
<td>3</td>
<td>35/35</td>
<td>0</td>
</tr>
<tr>
<td>13</td>
<td>Glioma 26</td>
<td>11</td>
<td>1281 ± 208</td>
<td>90</td>
<td>14</td>
<td>56/41</td>
<td>22</td>
</tr>
<tr>
<td>14</td>
<td>Glioma 26</td>
<td>6, 13, 17, 21</td>
<td>1546 ± 162</td>
<td>55</td>
<td>6</td>
<td>31/31</td>
<td>0</td>
</tr>
<tr>
<td>15</td>
<td>Mammary 16C</td>
<td>9</td>
<td>2009 ± 176</td>
<td>55</td>
<td>6</td>
<td>31/31</td>
<td>0</td>
</tr>
<tr>
<td>16</td>
<td>Mammary 16C</td>
<td>9</td>
<td>1801 ± 243</td>
<td>70</td>
<td>5</td>
<td>31/28</td>
<td>11</td>
</tr>
<tr>
<td>17</td>
<td>Mammary 16C</td>
<td>10</td>
<td>1871 ± 178</td>
<td>68</td>
<td>5</td>
<td>42/44</td>
<td>-5</td>
</tr>
<tr>
<td>18</td>
<td>Ovary M5076</td>
<td>8</td>
<td>1143 ± 126</td>
<td>71</td>
<td>9</td>
<td>36/38</td>
<td>-5</td>
</tr>
<tr>
<td>19</td>
<td>Ovary M5076</td>
<td>10</td>
<td>1536 ± 82</td>
<td>62</td>
<td>6</td>
<td>65/47</td>
<td>38</td>
</tr>
<tr>
<td>20</td>
<td>Ovary M5076</td>
<td>11, 13, 15, 17</td>
<td>1249 ± 73</td>
<td>57</td>
<td>13</td>
<td>53/42</td>
<td>26</td>
</tr>
<tr>
<td>21</td>
<td>Colon 38</td>
<td>12</td>
<td>1187 ± 157</td>
<td>71</td>
<td>13</td>
<td>82/87</td>
<td>-6</td>
</tr>
<tr>
<td>22</td>
<td>Colon 38</td>
<td>8, 15</td>
<td>1102 ± 136</td>
<td>62</td>
<td>9</td>
<td>62/47</td>
<td>32</td>
</tr>
<tr>
<td>23</td>
<td>Colon 38</td>
<td>7, 10, 15</td>
<td>1027 ± 74</td>
<td>75</td>
<td>8</td>
<td>65/49</td>
<td>25</td>
</tr>
<tr>
<td>24</td>
<td>Colon 38</td>
<td>8, 17, 22</td>
<td>1298 ± 142</td>
<td>85</td>
<td>19</td>
<td>78/88</td>
<td>15</td>
</tr>
</tbody>
</table>

* Calculated using the formula:

\[
TGI = \left(1 - \frac{\text{Av. volume treated tumors}}{\text{Av. volume control tumors}}\right) \times 100
\]

b Growth delay days, difference between treated and control groups in tumor growth to 1 cu cm.

c Ratio of the median day of death of the treated group to the control group.

d Percentage increase in life span of the treated group over that of the control group from the median day of death (MDD), using the formula:

\[
% \text{ ILS} = \frac{\text{MDD}_c - \text{MDD}_t}{\text{MDD}_c} \times 100
\]

* Average ± S.E.

for growth delay days. These results are shown in Table 3. The poor-response group consists of the Lewis lung and colon 26 carcinomas. The moderate-response group consists of the B16 melanoma, mammary adenocarcinomas 16C and C3HBA, and glioma 26. The good-response group consists of the M5076 ovarian carcinoma and the colon carcinoma 38. In all 3 response groups, the doubling times of the control tumors increase with tumor size. The control tumors in the good-response group exhibited longer doubling times than those in the poor-response and moderate-response groups. Regrowth rates of the treated tumors were initially slower than those of the controls immediately following treatment, but no differences in growth rates were apparent during the next doubling period.

**DISCUSSION**

The solid tumors used in these experiments were selected to represent a range of tumor origin and behavior. The Lewis lung carcinoma and B16 melanoma are rapidly growing tumors which have been extensively studied in drug research and screening (15). The 2 mammary tumors (16C and C3HBA) are slower growing and represent just 2 of the many types of mammary tumors available for study (1, 3). The colon carcinomas 38 and 26 have recently been proposed as experimental models for human colon cancer (2). The colon carcinoma 38 is a slow-growing, rarely metastasizing, differentiated tumor, whereas the colon carcinoma 26 is a rapidly growing and metastasizing undifferentiated tumor. The glioma 26 and M5076 ovarian carcinoma were chosen because they are models of tumors for which local hyperthermia may have a potential clinical role.

The treatment conditions (43°, 60 min) selected were based on a review of previous in vitro and in vivo studies which pointed to this temperature as the middle of a critical range (42-44°), where the heat sensitivity of neoplastic tissue exceeds that of normal tissues (14, 24). At temperatures above 43°, thermal ablation of tumor cells, stroma, and vasculature may occur (11). Since future experiments will investigate the combination treatment of drugs with LTH, destruction of the tumor blood supply was not desired. The duration of hyperthermia of 60 min was based on results of other workers (24), on our desire to minimize duration of anesthesia, and to allow
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a reasonable number of animals to be treated daily.

The day of treatment varied due to our desire for the tumor sizes to be within the range of 50 to 250 cu mm on the first day of LTH. The multiple-treatment schedules were chosen to allow the animals at least a 2- or 3-day recovery period between treatments and to avoid induction of thermal tolerance.

Several other investigators using similar conditions of moderate hyperthermia have studied a few of the tumors included in this report. Yerushalmi (35) induced LTH at 42-43° for 20 min in Lewis lung tumors using a hot-air heating system without generation of whole-body hyperthermia. He reported no life span prolongation or slowing of tumor growth subsequent to treatment, but a delay in the appearance of pulmonary metastases in treated animals compared with the untreated tumor-bearing animals was noted. These results are in agreement with our findings using more extensive local hyperthermia conditions on this tumor.

Crile (4) reported that it was difficult to cure S91 melanoma by either heat (water bath) or radiation alone without using exposures that caused irreversible damage to normal tissues. However, in a later report, Crile (5) indicated that S91 melanoma could be cured using LTH conditions of 44° for 30 to 40 min, but he did not present any data to substantiate this claim. No reports of the sensitivity of the B16 melanoma to LTH were found in the literature for comparison with our results.

A number of different mammary tumors have been treated using moderate LTH with variable results. Mendecki (22) used a microwave system similar to ours and obtained 100% cures following four 45-min treatments at 43°, while Robinson (26) and Thrall (32) observed no cures following single treatments under similar exposure conditions. Marmor (21) observed a high percentage of cures after single treatments with moderate LTH on a mammary sarcoma and a less sensitive mammary carcinoma, while Overgaard (25) only obtained 25% cures using the same exposure conditions. Suit (29) observed a growth delay following LTH at 43° of mouse mammary carcinoma which increased with increases in either treatment temperature or duration. A correlation between the sensitivity of mammary tumors to cure by X-rays and to cure by hyperthermia was also noted by Suit, but was not seen in cell culture studies by Gerweck (13). However, the in vitro results that Gerweck obtained may not be pertinent to the in vivo response of tumors to LTH treatments. Dickson (6) showed that smaller tumors were more effectively cured by local hyperthermia than were larger tumors, possibly as a consequence of inadequate heating at the edges of the larger tumors. Alternately, as tumors grow larger, the central region usually becomes necrotic, the growth fraction is reduced, the pH decreases, and the percentage of hypoxic cells increases (24). These factors, as well as tumor antigenicity (28, 31), have been shown to affect the cure of tumors by LTH. Of the 2 types of mammary tumors studied in this research, the 16C was found to be slightly more sensitive to LTH than the C3HBA.

In a recent study of the effects of 27.12-MHz short-wave diathermy heating on 12 experimental mouse tumors, Overgaard (23) observed a variation in cures from 9 to 25% for a single standard heat dose (42.5° for 60 min). Local tumor control was obtained only after a minimum heat dose was attained, but larger heat doses did not improve the percentage of cures. Histological examination of the heated tumors showed destruction of the tumor core but not of all cells in the periphery of the tumor where tumor regrowth usually occurred. This nonuniform thermal destruction of the tumor was explained as a technical insufficiency of the hyperthermic treatments. An inverse relationship between the cure rate and the peripheral infiltrative growth of the different types of tumors was also noted.

The variation in response to LTH found in the spectrum of tumors included in this study is consistent with the results of previous investigators. Our results at minimum tumor temperatures of 42.5° for 60 min indicate few regressions or cures and variable tumor growth delay. They are not as encouraging as those of some previous workers (5, 21-23, 25, 26) who, using a similar extent and duration of hyperthermia, have given the impression that local control of many animal tumors can be achieved by moderate LTH. Our studies suggest that higher temperatures and/or longer durations of exposure are neces-
sary to effect local control of the tumors studied.

Although LTH alone may not be curative under these LTH conditions, when used against well-established solid tumors, it can still play an important role in cancer treatment as an adjuvant therapy. In addition to killing a large fraction of the cells in a tumor, LTH has the advantage of being relatively nontoxic in comparison with other anticancer modalities. It can be applied locally with no burns and minimal systemic effects. The question of the most effective combination of LTH with ionizing radiation and drug therapy is currently under investigation in many laboratories. However, before LTH can establish itself as a useful adjuvant tumor treatment modality, the uncertainty surrounding a potentially dangerous side effect, the stimulation of metastasis, must be resolved.

In the results described in this paper, no direct attempts were made to study the metastasis of the individual tumors following LTH. However, the data obtained on life span can be considered as a rough measure of metastasis. Since no significant decreases in life span were consistently obtained between the heat-treated and control groups, it was concluded that under the LTH conditions of these experiments metastasis was not stimulated. Our results are consistent with previous studies on tumor metastasis and hyperthermia, since the local heating was well confined to the treatment volume, and no significant whole-body hyperthermia was induced.

The results of the comparison between tumor-doubling times and response to LTH treatments indicate 2 general factors influencing the sensitivity of these tumors to heat. First, the slower-growing tumors were most responsive to the heat treatments as measured by TGI and growth delay days, and, second, the treated tumors regrew following recovery from the heat damage at growth rates similar to the controls.

In this research, the sensitivity to LTH (43°, 60 min) of a spectrum of 8 different solid mouse tumors was investigated. The local hyperthermia treatments reduced the size and retarded the growth of the treated tumors compared with control tumors for each of the tumors tested. The faster-growing Lewis lung carcinoma and B16 melanoma were the least responsive to treatment, while the slower-growing colon 38 and M5076 ovarian carcinomas were the most responsive. Multiple treatments resulted in longer growth delays and greater TGI than did single treatments. No consistent difference in life span between the control and treated groups was measured, and only 5 of 188 treated animals were cured.

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Effects of Local Tumor Hyperthermia on the Growth of Solid Mouse Tumors

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