Interstitial Hypertension in Superficial Metastatic Melanomas in Humans

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ABSTRACT

Since 1950, several investigators have demonstrated that interstitial hypertension is a pathophysiological characteristic of experimental solid tumors. To date, interstitial fluid pressure (IFP) has not been measured in human tumors in situ. In this study we measured with the wick-in-needle technique the interstitial fluid pressure in superficial melanoma metastases (n = 12) in patients (n = 10) before and during systemic therapy. In the majority of tumors the pressure was found to be almost uniform, while in others it varied several-fold. The large variations in IFP in some tumors may be due to technical or biological factors. With the data obtained before and during therapy grouped, the mean IFP in melanoma lesions varied between 2 and 41 mm Hg with an overall mean of 14.3 ± 12.5 (SD). IFP was found to be significantly higher (P < 0.01) in large (22.8 ± 13 mm Hg; n = 6) than in small (5.8 ± 2 mm Hg; n = 6) lesions. This study demonstrates that IFP can be measured in human tumors using the wick-in-needle technique and that the pressure in some of the large melanomas exceeds the values measured to date in rodent tumors or human tumor xenografts. The latter result suggests that caution must be exercised in extrapolating values of pathophysiological parameters from transplanted tumors to human tumors.

INTRODUCTION

Since the original study of Young et al. (1) in 1950, several investigators have demonstrated that interstitial hypertension is a pathophysiological characteristic of experimental solid tumors (2, 3). These studies have shown that in general (a) IFP increases with size, (b) an increase in IFP is associated with a decrease in blood flow, and (c) pressure decreases from the center to the periphery of tumors. IFP is a critically important parameter in the delivery of blood-borne agents in solid tumors (2–5). We have recently initiated IFP studies in metastatic melanoma (present study), cervical carcinoma (6), breast cancer (7), colorectal cancer (7), and head and neck tumors (6) in patients. We report here the first measurements of IFP in superficial metastatic melanomas in patients prior to and during systemic therapy. These studies were carried out using the wick-in-needle technique to answer the following questions: (a) Is IFP in human tumors comparable to that in experimental tumors? (b) Is IFP tumor size dependent? (c) Does IFP change during therapy?

MATERIALS AND METHODS

Patient Selection. The protocol to measure IFP in patients with dermal and s.c. metastatic melanomas was approved by the Institutional Review Board of the Presbyterian University Hospital and of the University of Pittsburgh. Patients selected for this study were recruited from 3 different therapy protocols approved for the treatment of metastatic melanoma: (a) Phase IB study of monoclonal antibody R24; (b) Phase IB study of intralymphatic and s.c. interleukin 2; and (c) Phase II trial using carboplatin, dacarbazine, and (±)-tamoxifen. The age of the patients varied between 18 and 61 years old.

IFP Measurement. IFP was measured with the wick-in-needle technique developed by Fadnes et al. (8). In brief, 23-gauge needles with a 2–3-mm side-hole at 4–5 mm from the tip were filled with five surgical sutures (6-0 ethilon). The nylon sutures increase the contact area which should favor a better fluid communication between the interstitial space and the needle. In 3 patients with small tumors 26-gauge needles without a side hole and filled with 2 surgical sutures (6-0 ethilon) were used to measure pressure. In a separate study in experimental tumors no difference in the magnitude of IFP measurements was found between 23- and 26-gauge needles. The needles were connected to a pressure transducer (model P23XL; Spectramed Inc., Oxnard, CA) by polyethylene tubing filled with sterile, heparinized (70 units/ml) saline (Fig. 1A). The pressure transducer was connected to a preamplifier (model 11-4113-01; Gould Inc., Cleveland, OH) and the amplified signal was sent to a dual-channel chart recorder (model 30-V7202-11; Gould Inc.).

Depending upon the location of the melanoma lesion on the body, the patients were either sitting in bed, lying supine, or in the lateral decubitus position during the IFP measurements. The level of the heart (right atrium) was taken to be 7 cm below the manubriosternal angle in the sitting patients, at the midaxillary line in patients lying supine, and at midline in patients in the lateral decubitus position. The hydrostatic load between heart and tumor was measured with the pressure transducer and WIN setup, by placing the needle at heart level and then at tumor level.

As feasible, 1 or 2 superficial tumor lesions per patient were selected for pressure measurement. The pressure was measured in the same lesions before therapy and once during therapy between days 4 and 7. Under sterile conditions, the needle was placed at tumor level and the balance knob on the preamplifier was used to place the pen of the chart recorder at the zero reference point. Then the needle was inserted into the central regions of the tumor and left in place without external fixation. Two needles were inserted in different regions of a tumor. The number of measurements was determined by the tumor size; small lesions (0.1–4.0 cm3, n = 6) a maximum of 2 measurements was obtained, whereas in some of the larger lesions (8.0–106 cm3, n = 6) once the first pressure recording was completed, the needle was advanced deeper in the tumor, giving a total of 4 measurements with the 2 needles. For all the measurements the fluid communication between the pressure transducer and the tumor was checked by compressing and decompressing the tubing with a screw clamp (Fig. 1A). A measurement was accepted as valid when the pressure following compression and decompression did not differ by more than 15%. The mean IFP of the measurement was determined from the stable values after compression and decompression. The majority of the pressure values given in Table 1 represent the mean of 2–4 measurements; in 3 cases, only 1 measurement was valid or obtained.

Systemic blood pressure, tumor response to therapy, and tumor size were also recorded. The tumor volume was estimated from the formula for an ellipsoid:

\[
V = \frac{1}{2} A \times B^2
\]

where A and B represent, respectively, the maximum and minimum perpendicular diameters. Statistical analysis between means was evaluated with the Kruskal-Wallis test.

To determine if the hydrostatic load between heart and tumor level...
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Fig. 1. A, the wick-in-needle and pressure transducer setup used for the measurement of IFP. A 23-gauge needle is built with a 2-3-mm-long side hole at 4-5 mm from the tip and filled with five nylon sutures (6-0 ethilon). The wick-in-needle is connected via polyethylene tubing to a 23-gauge needle adapter attached to the dome of the pressure transducer. The screw clamp is used to compress and decompress the tubing in order to test the fluid communication between the transducer and the tumor. B, an actual pressure recording in a melanoma lesion. Following IFP stabilization at 0 mm Hg and needle insertion, the pressure rises rapidly and reaches a stable value of 36 mm Hg. After pressure stabilization, the tubing connecting the wick-in-needle to the transducer is compressed which results in a sharp increase in pressure, then an exponential decrease, followed by another period of stabilization before decompressing the tubing which produces a rapid decrease and an exponential increase. The mean IFP is calculated from the stable pressure values after compression and decompression.

Influenced the IFP measured, the posture of 2 patients in bed was modified during the measurements. The pressure transducer was adjusted to tumor level before inserting the needle into the tumor and after the posture modification. The needle was left in place during posture modification in order to obtain a continuous pressure measurement from one location in the tumor. Determinations of IFP without posture modification were faster, taking less than 30 min to complete. However, the measurements during posture modification took an extended period of 60 min or more, which accounts for our ability to examine the effect of hydrostatic load in a limited number of patients.

RESULTS

Eleven patients with metastatic melanomas consented to participate in the study. Procedural complications were limited to pain during the insertion of the needle which was managed with oral analgesics. In a few cases, following withdrawal of the needle, bleeding occurred which was self-limited. The IFP data of one patient were not accepted because the pressure did not reach a stable value. In some cases individual measurements were rejected because of poor fluid communication. After the introduction of a needle in a tumor, a sharp rise in pressure was generally observed followed by a stable pressure (Fig. 1B). The time constant of a rise varied between 10 and 60 s for the majority of measurements; however, it was as high as 1–2 min in a limited number of measurements. To check the fluid communication between the transducer and the tissue the compression and decompression test was done (Fig. 1B). When the fluid communication was good, response time after compression and decompression varied from 2 to 40 s in most cases. In a few cases the pressure reached a stable value after 1–4 min.

Individual IFP measurements in normal skin varied between −1 and +3 mm Hg (0.4 ± 1.7 mm Hg; n = 5), whereas in metastatic melanomas the pressures varied between 2 and 48 mm Hg. In the majority of tumors, pressures varied by less than 30% between the measurement sites; in the remaining lesions, the differences in IFP were severalfold. The mean IFP in melanoma lesions varied between 2 and 41 mm Hg (Table 1). With the data obtained before and during therapy grouped, the overall mean was 14.3 ± 12.5 mm Hg (n = 12). In order to further characterize the data the tumors were divided in small (0.1–4.0 cm²) and large (8–106 cm²) sizes. The mean pressures in large and small metastatic melanomas are given in Table 2. IFP magnitudes did not change significantly during therapy in 5 patients treated with R24 antibody. Of the ten patients included in the study none responded to therapy.

The hydrostatic load between heart and tumor level varied between +3 and +17 mm Hg (Table 1). In order to evaluate what percentage of the hydrostatic load was transmitted to the tumor, IFP was measured before and after posture modification in 2 patients. In one patient with an initial hydrostatic load of +5 mm Hg at tumor level (right lower quadrant of the abdomen) when supine and a hydrostatic load of −19 mm Hg when sitting, the IFP remained constant at 9 mm Hg. In the same patient with a melanoma lesion on the left lateral abdomen, the hydrostatic load was +4 when supine and +10 mm Hg when lying in the right lateral decubitus position; the pressure increased by 5 mm Hg in the lateral decubitus position. In a second patient with a melanoma lesion on the right lower quadrant of the abdomen, the hydrostatic load was +4 mm Hg when supine and −13 mm Hg in the sitting position; IFP decreased by 5 mm Hg when the patient was supine which represents 29% of the hydrostatic load transmitted to the tumor. In these two patients, IFP in the skin was also measured during posture modification; the pressures were not modified by variations of the hydrostatic load.

DISCUSSION

The present study demonstrates that IFP is greater in melanomas than in normal tissues and that IFP increases with tumor volume. The pressures measured in experimental tumors (2, 3) as well as in human tumors [cervical carcinomas during radiotherapy (6), head and neck tumors, breast and colorectal carcinomas (7) before surgical resection] varied between 2 and 33 mm Hg. In the present study some of the larger melanomas had pressures as high as 48 mm Hg. While the basis for the differences in the pressure magnitudes between large human melanomas and other types of human and experimental solid tumors is not clear, these pressure differences must be kept in mind while extrapolating values for pathophysiological parameters from one tumor type to another.

We recently demonstrated that IFP was uniform from the
central regions to the periphery of experimental tumors, with a sharp drop at the tumor-normal tissue interface (3). In most melanoma metastases IFP magnitude was almost similar; however, in some melanoma lesions the differences in IFP from one region to another were severalfold. It was not possible to determine if these differences in pressure represent different molecules which is believed to occur predominantly by convection (13). The presence of high uptake occur adjacent to regions of low uptake within individual tumor lesions. While it is possible that this poor accumulation results from the poor and heterogenous blood supply of melanomas, the high IFP could also be an important barrier (13). The presence of high pressures in melanomas would reduce the pressure gradients from the vascular to the interstitial space, thus reducing the extravasation of macromolecules which is believed to occur predominantly by convection (14). Therefore, novel strategies are now needed to overcome these physiological barriers in tumors.

Table 1 Interstitial fluid pressure in metastatic melanoma

<table>
<thead>
<tr>
<th>Age/sex</th>
<th>Tumor diameters (cm)</th>
<th>Tumor volume (cm³)</th>
<th>Hydrostatic load (mm Hg)</th>
<th>Before therapy</th>
<th>During therapy</th>
</tr>
</thead>
<tbody>
<tr>
<td>51/M</td>
<td>8.5, 5</td>
<td>106</td>
<td>+7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>18/M</td>
<td>4.4</td>
<td>32</td>
<td>+11</td>
<td>8 (120/88)</td>
<td>22 (116/80)</td>
</tr>
<tr>
<td>55/M</td>
<td>3.5, 3.5</td>
<td>21</td>
<td>+3</td>
<td>11 (132/80)</td>
<td>3 (160/82)</td>
</tr>
<tr>
<td>2, 1.5</td>
<td>2</td>
<td>2</td>
<td>+5</td>
<td>9</td>
<td></td>
</tr>
<tr>
<td>42/F</td>
<td>3.3</td>
<td>14</td>
<td>+12</td>
<td>37 (130/90)</td>
<td>34 (126/88)</td>
</tr>
<tr>
<td>49/M</td>
<td>4.2</td>
<td>8</td>
<td>+17</td>
<td>19</td>
<td></td>
</tr>
<tr>
<td>53/M</td>
<td>2.1, 5</td>
<td>2</td>
<td>+10</td>
<td>20 (110/70)</td>
<td>21 (110/70)</td>
</tr>
<tr>
<td>54/M</td>
<td>0.8, 0.5</td>
<td>0.1</td>
<td>+12</td>
<td>5 (118/72)</td>
<td>2 (120/74)</td>
</tr>
<tr>
<td>33/F</td>
<td>2.2</td>
<td>4</td>
<td>+5</td>
<td>7 (112/70)</td>
<td></td>
</tr>
<tr>
<td>45/M</td>
<td>0.7, 0.5</td>
<td>0.1</td>
<td>+6</td>
<td>5</td>
<td></td>
</tr>
<tr>
<td>61/F</td>
<td>0.9, 0.7</td>
<td>0.2</td>
<td>+5</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Maximum and minimum perpendicular tumor diameters.
* The height of the water column between heart and tumor measured with the pressure transducer. The positive value indicates that the tumor was higher than heart level.
* Blood pressure (mm Hg).
* Patient not available.

In a separate study, in 4 patients with complete regression of cervical carcinomas, IFP decreased from values between 10 and 24 mm Hg (mean, 17 mm Hg) to values between 0 and 7 mm Hg (mean, 4 mm Hg) during the course of fractionated radiotherapy. In 3 patients with a partial or no regression, IFP remained unchanged or increased (6). The relationship between IFP changes and tumor response has, however, not been established in the present study since no clinical responses to therapy were observed. Pressure measurements obtained before and during therapy revealed that IFP was not modified significantly. Additional studies are needed in human tumors to determine whether changes in IFP may be of predictive value for clinical antitumor response.

Table 2 Mean interstitial fluid pressure in small and large metastatic melanomas

<table>
<thead>
<tr>
<th>Tumor volume (cm³)</th>
<th>Before therapy (mean ± SD)</th>
<th>During therapy</th>
<th>Pooled*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Large (8–106 cm³)</td>
<td>19.0 ± 11.3 (5)</td>
<td>22.7 ± 13.5 (6)</td>
<td>22.8 ± 13.0 (6)</td>
</tr>
<tr>
<td>Small (0.1–4 cm³)</td>
<td>6.5 ± 2.0 (4)</td>
<td>4.0 ± 2.6 (3)</td>
<td>5.8 ± 2.2 (6)</td>
</tr>
<tr>
<td>P</td>
<td>0.027</td>
<td>0.052</td>
<td>0.001</td>
</tr>
</tbody>
</table>

* To calculate the pooled mean, the data obtained before and during therapy was grouped for each tumor.
* Numbers in parentheses, number of tumors.

REFERENCES

1. Young, J. S., Lumsden, C. E., and Stalker, A. L. The significance of the tissue pressure of normal testicular and of neoplastic (Brown-Pearce carci...
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