Predictors of Thrombosis in Hepatic Vasculature during Microwave Tumor Ablation of an In Vivo Porcine Model

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ABSTRACT

Purpose: To evaluate and model the risk of in vivo thrombosis in each hepatic vessel type during hepatic microwave ablation as a function of vessel diameter, velocity, and vessel-antenna spacing.

Materials and Methods: A single microwave ablation antenna was inserted into a single porcine lobe (n = 15 total) adjacent to a hepatic artery, hepatic vein, or portal vein branch. Conventional ultrasound and Doppler ultrasound were used to measure the vessel diameter, blood flow velocity, and vessel-antenna spacing. A microwave ablation zone was created at 100 W for 5 minutes. Thrombus formation was evaluated on ultrasound performed immediately after the procedure. Logistic regression was used to evaluate the predictive value of vessel diameter, blood flow velocity, and vessel-antenna spacing on vascular thrombosis.

Results: Thrombosis was identified in 53% of portal veins, 13% of hepatic veins, and 0% of hepatic arteries. The average peak blood flow rate of the hepatic artery was significantly greater than the average peak blood flow rate of the hepatic vein and portal vein. Peak blood flow velocity < 12.45 cm/s, vessel diameter < 5.10 mm, and vessel-antenna spacing < 3.75 mm were strong predictors of hepatic vein thrombosis. However, these individual factors were not predictive of the more common portal vein thrombosis.

Conclusions: Hepatic arteries do not appear to be at risk for thrombosis during microwave ablation procedures. Portal vein thrombosis was more common than hepatic vein thrombosis during microwave ablation treatments but was not as predictable based on vessel diameter, flow velocity, or vessel-antenna spacing alone.

ABBREVIATION

ROC = receiver operator characteristic

During a thermal ablation procedure, image guidance with computed tomography or ultrasound (US) is used to guide the applicator into the vicinity of the tumor. The process of planning applicator placement takes into account the effects of thermal damage on nearby vasculature. Larger blood vessels can act as a heat sink and limit tumor cell death in perivascular regions. Such vascular heat sinks have been particularly detrimental to the slower heating techniques associated with radiofrequency ablation, preventing the heating zone from extending to necessary margins for adequate coverage of larger tumors (1,2).

Microwave ablation has been shown to create faster heating and a larger ablation zone, which more effectively overcome nearby heat sinks. Although this heating advantage can be associated with improved performance when treating perivascular tumors, the large heat deposition into the vessel may have unintended consequences (3,4). Excess thermal damage to portal veins can cause acute thrombosis, leading to liver decompensation and lobar infarcts—devastating consequences for patients with diminished liver reserve (5,6). In case reports of
thrombus formation during microwave ablations, patients have required treatment with anticoagulation therapy (7,8).

Regardless of whether thrombosis is a desired effect of the treatment or an unintended consequence, the mechanism by which microwave ablations may produce vascular thrombosis in portal veins, hepatic veins, and hepatic arteries is poorly understood. With increased adoption of microwave ablation technologies worldwide, better understanding of the incidence, risk factors, and potential implications of microwave ablation–induced vascular thrombosis is needed to help guide clinical decision making and treatment planning. The goal of this study was to evaluate the risk of thrombosis in hepatic blood vessels during microwave ablation as a function of vessel type, diameter, velocity, and vessel-antenna spacing.

**MATERIALS AND METHODS**

All studies were performed under approval from our institutional animal care and use committee and complied with National Research Council guidelines (9). Female domestic swine (n = 5, mean weight = 70 kg; Arlington Farms, Arlington, Wisconsin) were sedated with 7 mg/kg tiletamine hydrochloride and zolazepam hydrochloride (Telazol; Fort Dodge Animal Health, Fort Dodge, Iowa) and 2.2 mg/kg xylazine hydrochloride (Xyla-Ject; Phoenix Pharmaceutical, St Joseph, Missouri) administered by intramuscular injection. Anesthesia was maintained with inhaled 1.0%–2.0% isoflurane (Halocarbon Laboratories, River Edge, New Jersey). An ear vein was cannulated with a 20-gauge angiocatheter for administration of intravenous fluids.

Microwave ablations (n = 15) were performed under US guidance with a single microwave antenna (LK-15; Neuwave Medical, Inc, Madison, Wisconsin) at 100 W for 5 minutes. Conventional and Doppler US (ACUSON Antares; Siemens Healthcare, Issaquah, Washington) imaging was performed immediately before and after the ablations to determine the diameter; blood flow velocity (peak and temporal pattern of flow); and vessel-antenna spacing of the nearest hepatic artery, hepatic vein, and portal vein with a 5-MHz center-frequency linear transducer (VFX 13-5; Siemens Healthcare) (Fig 1a–c). A single ablation was created in each liver lobe. Thrombosis of a vessel was identified through a loss of Doppler signal within the vessel lumen and was generally associated with hypoechoic thrombus identified on B-mode imaging. Thrombus formation

![Figure 1](image-url). Sample measurements taken in experimental setup. (a) US guidance of microwave antenna (arrowhead) positioned in the proximity of the portal veins and hepatic veins. (b) Portal vein walls (top), comprising loosely packed connective tissue, appear hyperechoic on US imaging. Hepatic vein walls (bottom), comprising tightly packed collagen fibers, are characterized by the absence of echogenic artifacts. (c) On Doppler imaging, portal vein (top) blood moves anterograde toward the hepatic sinusoids, greatly dampening the flow. Conversely, hepatic vein (bottom) blood flow is more pulsatile owing to anterograde-retrograde pressure/flow variations from the cardiac cycle.
was defined as a binary event, categorized as either present (Fig 2a) or absent (Fig 2b) for statistical analysis.

After ablation, animals were sacrificed with an intravenous injection of Beuthanasia-D (390 mg/mL pentobarbital sodium and 50 mg/mL phenytoin sodium at 0.2 mL/kg; Schering-Plough, Kenilworth, New Jersey). The liver was removed and sectioned along the axis of each antenna to confirm the location of the ablation zone near the targeted vessels and the visual presence of thrombus formation (Fig 2c). The cross-section of the vessel, ablation zone, and thrombus formation on gross pathology was used to validate the US images taken during the study.

**Statistical Analysis**

Differences in mean diameter, peak velocity, and vessel-antenna spacing were evaluated between hepatic arteries, hepatic veins, and portal veins by using a paired Student t test. Partial and full thrombus formation was considered a positive event, and the absence of thrombus formation was considered a negative event. With thrombus formation coded as a binary event, univariate logistic regression was used to model the rate of thrombosis as a function of the predictive variables (vessel diameter, blood flow velocity, and vessel-antenna spacing) in each type of vessel. Multivariate logistic regression was also performed to account for the simultaneous effect of two or three of the variables against thrombosis formation. Receiver operator characteristic (ROC) curves and areas under the curve were obtained for each model. Differences in areas under the curve were assessed with DeLong and DeLong tests (10). Two-tailed P values < .05 were considered significant. Statistical analysis was performed using MedCalc v7.4 (MedCalc Software, Inc, Mariakerke, Belgium) and R Statistical Software v3.01 (R Foundation for Statistical Computing, Vienna, Austria).

**RESULTS**

**Vessel Characteristics**

A sample size of n = 15 was collected for each vessel type. The mean hepatic artery diameter was 2.2 mm (range, 1.1–3.7 mm), peak blood velocity was 106.3 cm/s (range, 74.8–185.0 cm/s), and vessel-antenna spacing was 21.1 mm (range, 10.0–29.4 mm). As expected, the peak velocity of the hepatic artery was significantly greater than the peak velocity of the hepatic vein or portal vein (106.3 cm/s ± 27.2 [mean ± 1 SD] for hepatic artery, 24.0 cm/s ± 8.9 for hepatic vein, and 15.6 cm/s ± 9.0 for portal vein; P < .0001) (Fig 3a). The diameter of the hepatic artery was significantly smaller than the diameter of the hepatic vein or portal vein (2.2 mm ± 0.2, 7.3 mm ± 1.9 and 7.3 mm ± 3.4; P < .0001) (Fig 3b). No differences were noted in vessel-antenna spacing between the microwave antenna and hepatic arteries, hepatic veins, or portal veins (21.1 mm ± 5.5, 17.7 mm ± 10.5, and 20.7 mm ± 5.2; P > .05) (Fig 3c).

**Thrombosis Analysis**

**Hepatic Artery.** No thrombus formations were identified in any hepatic arteries. Owing to the absence of thrombus formation, a logistic function and its corresponding ROC curve could not be created.

**Hepatic Vein.** Thrombus formation was demonstrated in 13% of the hepatic veins (2 of 15; 1 partial

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**Figure 2.** Vessel imaging after ablation. (a) Thrombus formation seen in a nearby portal vein on US immediately after the ablation procedure at 100 W for 5 minutes. (b) Patent hepatic vein immediately after creation of microwave ablation zone. Note the absence of thrombus in the hepatic vein, even though the ablation zone abutted the vessel wall, as denoted by the hyperechoic gas bubbles made from the ablation zone (arrowhead). (c) Axial slice of ablation zone with a thrombosed portal vein (arrowhead) inside an ablation zone. (Available in color online at www.jvir.org.)
Logistic regression showed a significant negative correlation of thrombus formation with blood flow velocity ($P = .032$) and with vessel-antenna spacing ($P = .014$) and a strong negative correlation with vessel diameter ($P = .083$). Thrombus formation was more likely with slower flow, smaller vessels, and shorter vessel-antenna spacing in hepatic veins (Fig E1 [available online at www.jvir.org]). Univariate modeling predicted a 50% chance of thrombus formation for peak blood flow velocity $< 12.45$ cm/s, vessel diameter $< 5.10$ mm, or vessel-antenna spacing $< 3.75$ mm. The areas under the ROC curve generated from each univariate logistic function were high (0.885, 0.904, and 0.923 for the velocity, vessel diameter, and vessel-antenna spacing predictors) indicating that the selected factors of vessel diameter, blood flow velocity, and vessel-antenna spacing were able to separate thrombosis and nonthrombosis events in hepatic veins (Fig E2 [available online at www.jvir.org]). Details of the quantitative statistical metrics are outlined in Table 1.

Multivariate logistic regression, which adjusted an individual variable to account for the confounding effect of the other two variables, showed an inflated area under the ROC curve secondary to small sample size. Using vessel diameter, blood flow velocity, and vessel-antenna spacing simultaneously did not show improvements in predictive power over an individual variable alone. Perfect separability between events and nonevents also caused unreliable estimates of standard errors, preventing further significance testing.

**Portal Vein.** Thrombus formation was demonstrated in 53% of the portal veins (8 of 15; 4 partial and 4 full occlusions). In contrast to the results found for hepatic veins, peak flow velocity ($P = .264$), vessel diameter ($P = .563$), and vessel-antenna spacing ($P = .256$) were found not to be significant predictors of thrombosis in portal veins. Portal vein thrombosis occurred across a range of peak flow velocity, vessel diameter, and vessel-antenna spacing (Fig E3 [available online at www.jvir.org]). Correspondingly, the area under the ROC curve of the portal vein logistic functions was significantly less than that of the hepatic veins (0.607, 0.536, and 0.607 for velocity, vessel diameter, and vessel-antenna spacing predictor) (Fig E4 [available online at www.jvir.org]). Details of the quantitative statistical metrics are outlined in Table 2.

Multivariate logistic regression using blood flow velocity, vessel diameter, and vessel-antenna spacing simultaneously was marginally more predictive, with only
Table 1. Individual Contributions of Peak Velocity, Vessel Size, and Vessel-Antenna Spacings Toward to Thrombus Formation in Hepatic Veins

<table>
<thead>
<tr>
<th>Variables</th>
<th>OR</th>
<th>95% CI</th>
<th>P Value</th>
<th>Area</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peak velocity (cm/sec)</td>
<td>0.773</td>
<td>0.5292–1.280</td>
<td>0.0832</td>
<td>0.885</td>
<td>0.617–0.989</td>
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<tr>
<td>Vessel size (mm)</td>
<td>0.096</td>
<td>0.0018–5.2305</td>
<td>0.0321</td>
<td>0.904</td>
<td>0.641–0.994</td>
</tr>
<tr>
<td>Vessel-antenna spacing (mm)</td>
<td>0.725</td>
<td>0.4718–1.1143</td>
<td>0.0136</td>
<td>0.923</td>
<td>0.667–0.997</td>
</tr>
</tbody>
</table>

Cl = confidence interval; OR = odds ratio.
*Statistically significant.

Table 2. Individual Contributions of Peak Velocity, Vessel Size, and Vessel-Antenna Spacings Toward to Thrombus Formation in Portal Veins

<table>
<thead>
<tr>
<th>Variables</th>
<th>OR</th>
<th>95% CI</th>
<th>P Value</th>
<th>Area</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peak velocity (cm/sec)</td>
<td>0.942</td>
<td>0.7875–1.0847</td>
<td>0.2642</td>
<td>0.607</td>
<td>0.329–0.842</td>
</tr>
<tr>
<td>Vessel size (mm)</td>
<td>0.917</td>
<td>0.6829–1.2321</td>
<td>0.5633</td>
<td>0.536</td>
<td>0.268–0.789</td>
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<tr>
<td>Vessel-antenna spacing (mm)</td>
<td>0.881</td>
<td>0.6980–1.1114</td>
<td>0.2557</td>
<td>0.607</td>
<td>0.329–0.842</td>
</tr>
</tbody>
</table>

Cl = confidence interval; OR = odds ratio.

a slight increase in the area under the ROC curve, compared with any one univariate predictor alone (0.75 ± 0.13 vs 0.61 ± 0.16, 0.75 ± 0.13 vs 0.54 ± 0.08, and 0.75 ± 0.13 vs 0.607 ± 0.08; P < .05 for all comparisons).

DISCUSSION

This in vivo pilot study demonstrated that vascular thrombosis occurs four times more commonly in portal veins than hepatic veins during microwave ablation. Although vessel diameter, peak blood flow velocity, and vessel-antenna spacing were predictive of hepatic vein thrombosis, those factors were unable to predict portal vein thrombosis with sufficient sensitivity and specificity. Combining factors improved predictions compared with single factors alone, but the fit of the portal vein model was still less conclusive than the hepatic vein model. No incidents of hepatic arterial thrombus were identified in this study despite the close proximity of the microwave antenna to hepatic arteries of various sizes. This finding raises the possibility of a protective effect of rapid or pulsatile blood flow.

The primary goal of thermal ablation is to heat the target tumor and a margin of normal tissue to cytotoxic temperatures. Achieving an appropriate margin on average tumors of 2–3 cm has been challenging with existing radiofrequency ablation systems and early microwave ablation devices. In particular, the slower heating produced by those systems was not always sufficient for ablation near vascular structures > 3–5 mm in diameter (11). More recent high-power systems have demonstrated an improved ability to overcome perfusion-mediated cooling and create larger ablation zones at a faster rate, potentially decreasing the risk for tumor recurrence, especially in perivascular tissue (12–15). However, unintended collateral damage to hepatic vascular structures is now a greater clinical concern, especially in patients with cirrhosis (16,17). The results of this study may help guide clinical treatment planning to avoid damage to critical vessels, particularly in cases of slow blood flow and when tumors are located in close proximity to portal vein branches. For example, analysis of portal veins showed potential utility in using multiple predictors to anticipate when a thrombus may form. One sample combination showed that vessels > 7 mm, coupled with vessel-antenna spacing > 27 mm, predicted a < 30% chance of thrombosis formation. Likewise, in the vicinity of a 15-mm vessel, the vessel-antenna spacing can be decreased to 19 mm while maintaining a 30% chance of thrombosis formation. Conversely, in hepatic veins, where thrombus formation is rare, a physician may want to use a more aggressive approach to treat the tumor. Multiple antennas or larger power-time combinations that deliver greater energy deposition can be used to ensure complete thermal necrosis in the absence of vessel occlusion.

The results reported in this study confirm the previously established relationship between vessel diameter and acute thrombosis. An early in vivo porcine study on the effect of radiofrequency ablation on vessels found that vessels < 3 mm in diameter had an increased risk for occlusion (1). The same group later found similar results with microwave ablation, showing that vessels < 6 mm in diameter had an increased risk for thrombosis, whereas larger vessels had less of a risk for thrombosis (18). The aforementioned studies did not distinguish between vessel types. Clinical studies of hepatic radiofrequency ablation have also demonstrated that the incidence of inadvertent portal vein throm-
bosis (0.87%; 12 of 1,379) is greater than hepatic vein thrombosis (0.29%; 4 of 1,379) (19). These findings are concordant with our study, albeit at lower overall rates of thrombosis. Our higher rates can be attributable to the fact that we specifically targeted vessels in normal liver parenchyma compared with placing antennas in the center of a tumor.

The underlying reasons for the observed differences in thrombosis rates or the substantially lower ability for basic geometry parameters to predict portal vein thromboses are unclear. A plausible explanation is the difference in blood flow dynamics. Specifically, portal vein flow is relatively steady and into a capillary bed (hepatic sinusoids). Thrombus or increased portal pressure caused by elimination of the capillary bed is likely to increase resistance to forward flow. The resulting stagnation would subsequently increase the chances of activating the coagulation cascade; this is especially true in patients with portal hypertension, in whom portal venous resistance is already elevated. In contrast, hepatic vein flow is pulsatile, which decreases the likelihood of thrombus formation owing to turbulence, and hepatic veins empty into progressively larger vessels leading into the heart (20–22). Any hepatic venous thrombus that does develop is more likely to be dislodged from the vessel.

Limitations to this study were due to the simultaneous interplay of heat transfer, blood flow, and clotting in an in vivo environment. Evaluating a broad range of vessel-antenna spacing and velocity combinations (needed to assess their relationship to thrombus formation) was challenging. In particular, we had limited control over blood flow velocity. Such control may be more possible in an ex vivo system, but the use of anticoagulated blood or fluids in such systems would not allow us to evaluate clotting. In an effort to reduce variability among ablations, we used a single ablation setting of 100 W for 5 minutes. Our results can potentially be generalized to other power-time settings by integrating a scaling factor, but such broad analysis was beyond the scope of this investigation. Another limitation of this study was the lack of a tumor model. The vasculature near or within a tumor differs from the vasculature in normal tissue and can affect how microwave energy and heat propagate. These limitations need to be taken into account when extrapolating the results of this study to a clinical environment where liver cirrhosis and tumor are present. With regard to sample size, the small number of thrombus formations in hepatic veins led to difficulties in performing error analysis. Although there are drawbacks to forming conclusions based on such a small number of events, the results still illustrate the resistance of hepatic veins to thrombus formation. Finally, the objective of our study was to focus on acute thrombus formation. Rates of delayed thrombus formation, which would result not only from the decreased blood flow from upstream acute thrombus but also from the thermally damaged endothelial layers, were not studied (8). In human patients with cirrhosis, arteriovenous shunts near tumors, and portal hypertension, portal vein velocities would be expected to be lower than in healthy pig livers, and a higher rate of thrombosis would be expected compared with what was seen in this study.

In conclusion, our in vivo study demonstrated that portal veins exhibit more frequent, but less predictable, rates of thrombus formation than hepatic veins during microwave ablation. Hepatic veins showed a clear correlation between thrombus risk and peak flow velocity, vessel diameter, and vessel-antenna spacing. We did not identify thrombus formation in any hepatic arteries. Follow-up studies to refine further the risk of vascular thrombosis in the clinical setting would be helpful for procedural planning and establishing risk profiles.

ACKNOWLEDGMENT

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REFERENCES


Figure E1. Logistic regression model of hepatic veins. Vessel velocity and spacing were significantly correlated with thrombosis, whereas vessel size was correlated but not significant. CI = confidence interval; OR = odds ratio.

Figure E2. Receiver operator characteristic curve for hepatic vein logistic regression model. Because of the small number of events, the regression model could classify thrombus formation with high sensitivity and specificity.
Figure E3. Logistic regression model of portal veins. Size, velocity, and vessel-antenna spacing were not considered significant contributors to portal vein thrombosis. CI = confidence interval; OR = odds ratio.

Figure E4. Receiver operator characteristic curve for portal vein logistic regression model. The model was poorly discriminating, with limited performance in separating the likelihood of a thrombotic event during an ablation procedure. The areas under the curve of each predictor were not significantly different from each other.