Blood Oxygen Level-Dependent Liver MRI: Can It Predict Microvascular Invasion in HCC?

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Purpose: To assess Blood Oxygen Level-Dependent (BOLD) Magnetic Resonance Imaging (MRI) for noninvasive preoperative prediction of Microvascular Invasion (MVI) in Hepatocellular Carcinoma (HCC).

Materials and Methods: In this prospective, institutional review board approved study, 26 patients (21 men and 5 women age range, 34–77 years with mean age of 61 years) with HCC were evaluated preoperatively with liver MRI including baseline and post oxygen (O2) breathing BOLD MRI. Post processing of MRI data was performed to obtain R2* values (1/s) and correlated with histopathological assessment of MVI. Statistical analysis was performed to assess correlation of baseline R2*, post O2 R2* and R2* ratios to presence of MVI in HCC by binary logistic regression analysis.

Results: MVI was present in 15/26 (58%) of HCC on histopathology. The mean R2* values ± SD at baseline and post O2 with and without MVI were 35 ± 12, 36 ± 12, 38 ± 10, 42 ± 17. The R2* values between the groups with and without MVI were not significantly different statistically.

Conclusion: BOLD MRI is unable to accurately predict MVI in HCC. The noninvasive preoperative MRI detection of MVI in HCC remains elusive.

Key Words: hepatocellular carcinoma (HCC); magnetic resonance imaging (MRI); blood oxygen level dependent MRI (BOLD MRI); microvascular invasion (MVI); cirrhosis; liver transplantation

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LIVER RESECTION AND transplantation are the only curative treatment options currently available for hepatocellular carcinoma (HCC) (1–3). However, long-term survival is still poor as a result of a high rate of recurrence among other drawbacks, such as lack of reliable prognostic factors, phenotypic diversity of the disease, and the lack of effective systemic treatment. Given the limited organ availability as well as morbidity and or mortality risks associated with surgical options, it is critical to select patients who are likely to have long-term curative outcome without recurrence. Among many outcome prediction parameters such as tumor grade, differentiation, size, multiplicity, and vascular invasion (macro and microscopic), microvascular invasion (MVI) has been correlated to be one of the most significant independent risk factors affecting recurrence-free survival following curative resection and or liver transplantation (4). Preoperative prediction of microvascular invasion could allow appropriate patient selection for liver transplantation and predicting prognosis.

Although the combined use of imaging modalities, including MRI, computed tomography (CT), and ultrasonography, can detect tumor invasion of the major branches of the portal and hepatic veins in 81–95% of cases at the time of diagnosis, imaging studies currently do not have the ability to detect microvascular invasion (5–14). The diagnosis of MVI cannot be reliably achieved by a biopsy due to sampling errors (15) as only a small region of the tumor is being evaluated in a biopsy as compared to surgical pathology. Intratumoral hypoxia has been shown to enhance proliferation, angiogenesis, metastasis, chemoresistance, and radioresistance of HCC resulting in overall increased tumor invasiveness (16–18). Blood oxygen level dependent (BOLD) MRI is a noninvasive diagnostic method capable of assessing tumor oxygenation and indirectly hypoxia, by detecting signal changes secondary to changes in blood flow and oxygenation. The purpose of this study was to correlate prospectively the ability of BOLD MRI to predict MVI preoperatively compared with histopathology in patients with HCC who were undergoing liver resection.

MATERIAL AND METHODS

Patients

This was a prospective single center HIPAA compliant, institutional review board approved study with enrolled patients giving approval to participate by means
of written informed consent. Over a 2-year period from September 2009 to 2011, 28 patients with suspected HCC who were to undergo liver resection (partial hepatectomy) where accrued to this study and underwent routine staging liver MRI along with BOLD MR before surgery with an average time interval between MRI and surgery of 25 days (range, 1 to 138 days). Exclusion criteria included specifically lack of histopathology; prior tumor treatment such as transcatheter chemoembolization, radiofrequency ablation, or chemoradiation; and general contraindications to MRI and or gadolinium-based contrast agents. Two patients were excluded as their surgery was canceled. Thus, our final study cohort included 26 patients (21 men and 5 women age range, 34–77 years with mean age of 61 years) who underwent liver MRI and subsequent surgical resection and histopathology. Serum alpha fetoprotein (AFP) levels were in the range of 2–15408 μg/L with mean of 1101 μg/L. Chronic viral hepatitis B and or C was present in 65% of patients.

**MR Imaging**

MR imaging was performed on all subjects on a 1.5 Tesla (T) MR system (Siemens Avanto, Siemens Healthcare, Erlangen, Germany). Torso phased-array coils were used. All subjects underwent a routine Liver MR protocol (Table 1) that included coronal single-shot T2-weighted HASTE, fat suppressed motion corrected axial T2 fast spin echo (BLADE), fat suppressed axial single-shot T2 HASTE (long TE), axial gradient-refocused echo T1-weighted in-phase and out-of-phase, axial diffusion-weighted imaging (DWI), and three-dimensional (3D) T1-weighted imaging (T1WI) before and after IV injection of gadolinium contrast material. Bold MR imaging was an addition to this routine liver protocol before gadolinium contrast injection. The BOLD MR imaging comprised of a 12 multiecho gradient refocused echo (GRE) T2* imaging pulse sequence with TE ranging from 1 to 41 ms. BOLD MR Imaging was performed before and after inhalation of oxygen. Oxygen (100%, 5L/min) was administered through a nasal mask for 5 min before a repeat BOLD acquisition was obtained following oxygenation. The post oxygenation BOLD imaging was followed by multiphasic gadolinium enhanced T1WI.

**Imaging Analysis**

**Primary Analysis**

The primary analysis comprised of quantitative estimations of R2*. All the MRI scans including the BOLD data were anonymized and saved to a password protected encrypted hard drive for analysis purposes. R2* Analysis was obtained using commercial software (Image J, NIH) (Fig. 1). Multiple ROIs were placed on HCC, liver, and muscle on multiple slices to obtain T2* and thereafter R2* (1/T2* in 1/s) for HCC, liver, and muscle. Areas of obvious susceptibility artifacts were excluded from ROI measurement. The ROI tracing was performed manually taking care to be within confines of tumor at all times. Multiple R2* indices such as ratios and or differences in R2* between tumor and liver before and after oxygenation were

**Table 1**

Technical Parameters of the Liver MRI Protocol Utilized

| Pulse sequence | TR (ms) | TE (ms) | Slice thickness (mm) | No. of slices | Matrix | Averages | Parallel imaging factor | Time (min:s) |
|----------------|---------|---------|----------------------|--------------|--------|----------|------------------------|--------------|
| T2 HASTE BREATHING LOC | 1000 | 84 | 8 | 10 | 256x192 | 1 | 2 | 0:20 |
| T2 HASTE BH LOC | 1000 | 84 | 8 | 10 | 256x192 | 1 | 2 | 0:20 |
| T2 COR HASTE BH | 1000 | 181 | 8 | 40 | 320X256 | 1 | 2 | 0:40 |
| T2 AX HASTE FS | 1000 | 181 | 8 | 40 | 320X256 | 1 | 2 | 0:42 |
| T1 AX IN/OUT PHASE | 174 | 2.2, 4.5 | 5 | 40 | 256X216 | 1 | 2 | 0:43 |
| T2 AX BLADE FS TE 90 | 7910 | 92 | 8 | 30 | 320X320 | 1 | 2 | 2:22 |
| DWI (b100,600) | 5300 | 68 | 8 | 30 | 192X144 | 6 | 2 | 3:27 |
| GRE BOLD (12 echo) pre O2 | 80 | 1,35,7,...,41 | 8 | 4 | 128X128 | 2 | 2 | 0:49 |
| GRE BOLD (12 echo) post O2 | 80 | 1,35,7,...,41 | 8 | 4 | 128X128 | 2 | 2 | 0:49 |
| AX T1 VIBE Pre Contrast | 3.5 | 1.31ms | 5 | 48 | 320X192 | 1 | 2 | 0:21 |
| Care_Bolus | 35.08 | 1.43ms | 8 | 1 | 256 | 1 | Off | 1:42 |
| AX T1 VIBE Spair Dynamic | 3.5 | 1.31ms | 5 | 48 | 320X192 | 1 | 2 | 1:04 |
| AX T1 VIBE Spair 5 min. delay | 3.5 | 1.31ms | 5 | 48 | 320X192 | 1 | 2 | 0:21 |

**Figure 1.** R2* map of the liver generated using ImageJ software. The trace demarcates the HCC. Note the visual difference in gray scale shade of the tumor from the background liver.
calculated to seek discriminative thresholds for prediction of MVI.

Secondary Analysis

The secondary imaging analysis included multiparametric clinical, morphological, and DWI assessment. Detailed morphological observations were recorded performed by two readers in consensus using the departmental PACS viewer for review regarding the tumor size, shape location, signal intensity on T1- and T2-weighted images, multiphasic enhancement patterns and contrast washout, presence or absence of tumoral capsule, satellite nodules of tumor, and intra and extrahepatic metastatic disease. DWI analysis was done by a single observer calculating the ADC values from the ADC maps generated during the DWI imaging acquisition. Regions of interest (ROIs) were drawn on the ADC maps in areas of interest encompassing the tumor as well as background liver on a MRI postprocessing workstation (Siemens Healthcare, Erlangen, Germany).

Pathological Assessment

Tumor characteristics were evaluated by review of the pathological specimens. Tumor size was measured as the largest diameter of the major tumor in centimeters. Macrovascular invasion is defined as gross vascular invasion into major portal vessels or hepatic veins. Microvascular invasion was determined on pathologic analysis as microscopic vascular invasion of small vessels within the peritumoral parenchyma of the liver (Figs. 2, 3). The predominant histopathologic grade of differentiation of the tumors was assessed according to Edmondson-Steiner criteria (G1, well differentiated; G2, moderately differentiated; G3, poorly differentiated; G4, undifferentiated). Immunohistochemistry using the monoclonal antibody QBEnd10 (anti-CD34) was performed in cases microvascular invasion is suspected but not unequivocal to help to confirm or exclude microvascular invasion. For the purpose of this study, sections from the blocks that had already been used for routine histological examination were stained by the streptavidin-biotin complex immunohistochemical technique. The slides were counterstained with hematoxylin, and blindly and randomly examined by two pathologists who had already evaluated hematoxylin and eosin (HE) sections. The presence of neoplastic cells inside the lumina of the vessels whose endothelium had been immunohistochemically stained at the periphery of the HCC nodules was carefully searched for the presence of attached tumor embolus.

Data and Statistical Analysis

Binary logistic regression was used to assess the ability of R2 parameters and quantitative morphological parameters to predict MVI. Stepwise variable selection in the context of binary logistic regression was also performed to evaluate the combination of MRI and morphological parameters in predicting MVI. Furthermore, BOLD MRI parameters (R2* values) and quantitative morphological outcomes (AFP value, length dimension, volume, and ADC values) were compared by means of the Wilcoxon rank-sum test (also known as the Mann-Whitney U test). Fisher’s exact test was used to compare between patients with and without MVI in regards to the qualitative morphological parameters. The analysis was done using the statistical software IBM SPSS Version 20.

RESULTS

Histopathologic Findings

All 26 patients who had surgical resection for presumed HCC were confirmed at histology. The tumor size ranged from 1.5 cm to 14 cm with mean size of
5.84 cm. The degree of differentiation in HCC was as follows: 01 (3.8%) well differentiated, 22 (84.6%) moderately differentiated, and 3 (11.6%) poorly differentiated. The grade of liver fibrosis was as follows: Grade 1 = 1, (3.8%), Grade 2 = 3 (11.5%), Grade 3 = 3 (11.5%), Grade 4 = 13 (50%), No fibrosis = 3 (11.5%), Regressed cirrhosis = 3 (11.5%). The degree of fatty infiltration exhibited by the background liver parenchyma was as follows: Fatty liver was present in 19 (73.1%) cases and background liver was non fatty in 7 (26.9%) cases. Iron deposition was noted in 11 (42.3%) cases. MVI was present in 15 tumors (57.7 %) and absent in 11 (42.3 %). No macroscopic vascular invasion was seen.

### MRI Findings and Analysis

All 26 HCC were identified and imaging analysis was performed both quantitatively as well as qualitatively for primary and secondary analysis as described in the methods section. In the primary analysis, the mean baseline (preoxygenation) R2* values (1/s) in tumors with and without MVI were 35 ± 12 (24–74) and 38 ± 12 (25–58), respectively. The mean post oxygenation R2* values (1/s) in tumors with and without MVI were 36 ± 10 (24–72) and 42 ± 17 (24–83), respectively. The mean difference (delta) in R2* values (1/s) before and after oxygenation with and without MVI were −1 (−15 to +7) and −4 (−50 to +26). Various other indices to reflect R2* ratios between the HCC and liver before and after oxygenation as well ratios of difference in R2* values between HCC and liver were calculated to seek any discriminative threshold of correlation with presence or absence of MVI on histopathology. Table 2 displays descriptive information about the MRI parameters of interest in each group and the P value from the Wilcoxon rank-

| Variable                           | MVI = YES (n = 15) | MVI = NO (n = 11) | P value |
|-----------------------------------|--------------------|-------------------|---------|
|                                  | Mean  | SD    | Min | Max | Mean  | SD    | Min | Max |         |
| HCC PRE                           | 35    | 12    | 24  | 74  | 38    | 10    | 25  | 58  | 0.30    |
| HCC POST                          | 36    | 12    | 24  | 72  | 42    | 17    | 24  | 83  | 0.47    |
| HCC PRE – HCC POST                | -1    | 0.5   | -15 | 7   | -4    | 1.9   | -50 | 26  | 0.54    |
| HCC PRE/LIVER PRE                 | 0.705 | 0.170 | 0.496 | 1.131 | 0.696 | 0.192 | 0.402 | 1.055 | 0.96 |
| HCC POST/LIVER POST               | 0.732 | 0.148 | 0.543 | 1.121 | 0.821 | 0.505 | 0.410 | 2.113 | 0.64 |
| HCC POST/HCC PRE                  | 1.039 | 0.168 | 0.821 | 1.414 | 1.151 | 0.526 | 0.562 | 2.535 | 0.57 |
| LIVER PRE – HCC PRE               | 14    | 9     | -9  | 27  | 19    | 17    | -3  | 58  | 0.88    |
| LIVER POST – HCC POST             | 12    | 7     | -8  | 20  | 14    | 27    | -44 | 56  | 0.47    |
| LIVER POST – HCC POST/LIVER PRE   | 0.949 | 0.295 | 0.546 | 1.668 | -0.567 | 2.857 | -7.496 | 1.363 | 0.47 |
| PERCENT CHANGE                    | 3.91% | 16.79% | -17.9% | 41.38% | 15.1% | 52.6% | -43.8% | 153.5% | 0.57 |
sum test comparing the two groups. A series of logistic regression models were examined to identify which of the baseline R2 and post oxygenation R2 variables can significantly predict MVI status. However, none of the R2 variables (HCC or liver) were statistically significant. The insignificant results may be due to the relative small sample size in each group. Nevertheless, bootstrap methods for logistic regression that are appropriate for small data sets were applied as well yielding insignificant results.

Thus, overall, no significant differences were found between those who had MVI and those who did not with regard to the BOLD MRI parameters (R2*).

Secondary analysis included multiparametric clinical and morphological qualitative and quantitative analysis including ADC values from DWI acquisition. Morphological included assessment of tumor size, volume, T1 and T2 signal intensities, enhancement patterns, tumor margins, capsule characteristics if present, satellite lesions, and ADC values. The mean tumor size was 5.8 cm (1.5–14 cm) with 15 tumors located in the right lobe (57.7%), 9 in the left lobe (34.6%), and 2 (7.7%) straddling both lobes. Six of 26 (23%) tumors were at least partially in an exophytic location in relation to the liver surface. Thirteen of 26 (50%) had smooth margins, while 5 (19%) had irregular or 8 (31%) had lobulated margins. Sixteen of 26 (61.5%) were hypointense, 5 (19%) hyperintense, 1 (3.8%) isointense, and 4 (15.7%) had mixed signal intensity on T1. Twenty-one of 26 (88.5%) had hyperintense signal, 2 (7.7%) had hypointense signal, while 1 (3.8%) and 2 (7.7%) had mixed signal intensity on T2. Twenty-three of 26 (88.5%) tumors were hyper enhancing while 3 (11.5%) were hypoenhancing on arterial phase. Portal venous phase hypoenhancement (washout) was noted in 25 (96%) and only 1 (4%) tumor did not display this characteristic. All (100%) tumors were hypointense on the 5-min delayed post-contrast phases. Tumoral capsule (Fig. 4) was identified in 22 (85%) and absent in 4 (15%) of cases. When present, the capsule was incomplete in 8 (36%) and complete in 14 (64%). Satellite lesions were observed in only 2 (8%) HCC. The mean ADC value of HCC was $1.040 \times 10^{-3}$ mm$^2$ s$^{-1}$ (range, $0.417 \times 10^{-3}$ mm$^2$ s$^{-1}$ to $1.354 \times 10^{-3}$ mm$^2$ s$^{-1}$) while that of background liver parenchyma was $0.981 \times 10^{-3}$ mm$^2$ s$^{-1}$ (range, $0.728 \times 10^{-3}$ mm$^2$ s$^{-1}$ to $1.293 \times 10^{-3}$ mm$^2$ s$^{-1}$). The clinical and morphological variables are displayed in Table 3. There was no statistically significant difference in morphological variables between those who had MVI and those who did not. Clinical variables assessed for prediction of MVI such as AFP level and tumor differentiation/grade were also not statistically significant between the two groups of tumors with and without MVI.

**DISCUSSION**

Treatment decisions for HCC currently based on tumor number, size, and liver function continue to be hampered by poor long-term survival and high recurrence rates (19–21). The combination of tumor recurrence with vascular invasion limits additional attempts at various therapies, such as repeat hepatic resections, percutaneous ethanol injection, microwave coagulation therapy, and radiofrequency ablation, thereby contributing to poor survival (22–24). Tumor invasion into microscopic branches of the portal and/or hepatic veins (MVI) is associated with increased risk of early tumor recurrences following surgical

**Table 3**

Predicted Microvascular Invasion in Patients With HCC Using Morphological, Clinical, and Histological Parameters

| Parameter                                      | MVI Present (n = 15) | MVI absent (n = 11) | P value  |
|------------------------------------------------|----------------------|---------------------|----------|
| Tumor size (cm) Mean +/- SD                    | 5 +/- 2.4            | 6.2 +/- 4.1         | 0.73     |
| Capsule present (n)                            | 14 (93.3%)           | 8 (72.7%)           | 0.28     |
| Incomplete capsule (n)                         | 7 (50%)              | 1 (12.5%)           | 0.17     |
| Irregular, Lobulated margins (n)               | 9 (60%)              | 4 (36.4%)           | 0.120.43 |
| Satellite nodule present (n)                   | 2 (13.3%)            | 0                   | 0.5      |
| Exophytic tumor location (n)                   | 4 (26.7%)            | 2 (18.2%)           | 1        |
| T1 signal -hyper or hypo (n)                   | 13 (86.7%)           | 8 (72.7%)           | 0.850.62 |
| T2 signal -hyper or hypo (n)                   | 13 (86.7%)           | 10 (90.9%)          | 0.561    |
| Arterial hyperenhancement (n)                  | 13 (86.7%)           | 10 (90.9%)          | 1        |
| Venous hypoenhancement (n)                     | 15 (100%)            | 10 (90.9%)          | 0.42     |
| ADC (mm2/s) Mean                               | 1.32 x 10-3          | 1.03 x 10-3         | 0.35     |
| Serum AFP level (ug/L) mean                    | 1325                 | 787                 | 0.15     |
| Moderately or poorly differentiated tumor (n)  | 15 (100%)            | 10 (90%)            | 0.750.42 |
treatment of HCC and shorter survival (25–32). Accurate prediction of MVI would impact treatment decisions based on anticipated tumor biology, therapeutic response and clinical outcomes, but because current imaging techniques as well as preoperative biopsy are unable to accurately predict MVI in HCC, this important prognostic marker remains unconsidered while making treatment decisions for HCC.

Lack of sufficient oxygenation, hypoxia, is a common tumor microenvironmental characteristic caused by the imbalance between oxygen supply by abnormal tumor vasculature and demand by rapidly proliferating tumor cells. Evidence suggests that the heterodimeric transcription factor hypoxia inducible factor 1a (HIF-1a) controls the expression of a variety of genes, which play crucial roles in the acute and chronic adaptation of tumor cells to oxygen deficiency, including enhanced erythropoiesis and upregulated glycolysis, promotion of cell survival, inhibition of apoptosis, inhibition of cell differentiation, and increased angiogenesis. Hypoxic stress accelerates the invasion of hepatoma by up-regulating ETS-1 and the matrix metalloproteinases family by the HIF-1a-independent pathway. These adaptive changes of gene expression in neoplastic cells result in tumor invasion, metastases, and chemoradiation resistance (33–37). Specifically related to the liver, fibrogenesis associated with cirrhosis reduces hepatic blood flow leading to hypoxia. High proliferation of tumor cells also induces local hypoxia within HCC also stimulating angiogenesis to support the tumor growth by inducing the expression of angiogenic factors (16,17). To summarize, hypoxia enhances proliferation, angiogenesis, metastasis, and chemo- and radioresistance of HCC resulting in increased tumor invasiveness (MVI), which is essentially the primary step for future development of macrovascular invasion and or metastases in HCC. Thus, the level of intratumoral oxygenation, precisely hypoxia, could be a contributing factor to development of MVI.

BOLD MRI is a noninvasive technique that could estimate tumor oxygenation would find broad applications in prediction of MVI in HCC. The blood oxygen level-dependent (BOLD) MRI imaging technique has the unique capability to study tumor pathophysiology noninvasively. By accentuating the susceptibility effect of deoxyhemoglobin (dHb) in the blood with gradient-echo techniques, image contrast reflects the blood oxygen level. Changes in R2 (1/T2) reflect the capillary microvasculature whereas R2* (1/T2*) is sensitive to both microand macrovasculature (38). Some studies have shown a linear relationship between R2* and content of deoxyhemoglobin while others have reported a quadratic function between blood R2* and oxygen saturation, suggesting that the technique should be more sensitive in regions with low oxygen saturation, e.g., in tumors (39,40). Tumor basal R2* may potentially be considered as an intrinsic marker of pO2, because it is related to the oxygenation state of hemoglobin and to the arterial blood pO2, which is in equilibrium with tissue pO2. MRI using R2* quantification (BOLD MRI) has been reported to be a promising tool for noninvasive imaging of prostate cancer hypoxia (41,42).

In this study, we correlated the presence of MVI on histology by MRI estimation of BOLD effect (R2*) in HCC. We performed tumoral and background liver R2* estimations at baseline as well as after oxygen inhalation. Oxygen inhalation was performed to extract physiological changes induced in tumoral oxygen content which might allude to better determination of intratumoral hypoxia and thereby MVI. Our data analysis of absolute estimations of baseline R2* and postoxygenation R2* did not reveal a statistically significant threshold for prediction of MVI. We also assessed the difference in R2* before and after oxygenation, which also failed to provide a positive result statistically. Calculations of the ratios of R2* values between HCC and liver parenchyma both pre and post oxygenation were also unable to deliver a quantitative threshold to predict MVI. Finally, ratio of the delta R2* values between HCC and liver before and after oxygenation as well as percentage change in R2* values in HCC before and after oxygenation also did not result in a significant result. Our negative results, although disappointing, may be relevant to other investigators in this area. The use of BOLD contrast in tumors is a relatively new area of research and brings with its challenges of understanding and interpretation. As with any technique, BOLD MRI has both advantages and disadvantages. One advantage of BOLD MRI is that it is noninvasive BOLD MRI also has high spatial resolution, allowing it to address the issue of the spatial heterogeneity of the tumor response. To distinguish the contribution from the inflow and blood oxygenation to the BOLD signals, multiple gradient-echo imaging sequences is used instead of using conventional gradient-echo techniques. Carbogen induced changes in R2* or basal R2*, which reflect vascular development, may also be monitored with BOLD MRI to predict radiotherapy sensitivity. As for disadvantages, BOLD MRI is unfortunately an indirect method for monitoring tumor pO2. This is the result of the extreme sensitivity of changes in R2* to the basal state of tumor oxygenation and blood volume fraction. The intra- and intertumoral distribution of these parameters may be greatly heterogeneous, making it very difficult to compare estimated pO2 changes between two regions or individuals. Even more problematic is the fact that the change in R2* is not always indicative of the change in pO2 only. Concomitant changes in blood volume, blood pH, and metabolic status can lead to smaller-than-expected or even negative changes in R2*. In our secondary analysis, we also performed multiparametric morphological and DWI analysis to assess for any significant factors that could correlate with presence of MVI. However, neither of these parameters yield a statistically encouraging result. Although we did observe an association of MVI with the presence of an incomplete tumoral capsule (Figs. 4, 5) the correlation was not statistically relevant. This secondary result is, however, in keeping with prior and more recent studies (14) that have attempted to correlate tumoral morphologic characteristics with prediction of MVI and returned negative results.
is also not a static entity, but can be rather than
may interfere with the depiction of hypoxia. Hypoxia
accounted for by the complexity of hypoxia. Factors
relation between BOLD MR and MVI might be
that necessarily correlates with MVI. The lack of cor-
mitation could revolve around the simplistic assump-
could only worsen the results. Finally, a significant li-
result in temporal and spatial discrepancies and
acquisitions instead of breathheld acquisitions would
hold given the nature of multi-echo gradient echo ac-
ber of slices that can be obtained in a single breath-
estimation. There is a technical limitation to the num-
may be responsible for inaccurate oxygenation status
were larger than 5 cm, we may not be sampling the
limitations for BOLD evaluation. Because many tumors
and or morphological imaging methods remains
as well. The prediction of MVI by existing functional
ing is already known to be unable to determine MVI
as well. The prediction of MVI by existing functional
and morphological imaging methods remains elusive.

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Figure 5. Contrast enhanced T1 weighted MR image show-
ing focal capsule break (black arrow) and adjacent satellite
nodule (white arrow) in HCC which demonstrated MVI on
histology.

There are several limitations to our study that we
would like to acknowledge. First, is the small sample
size of only 26 subjects. However, this was an explora-
tory pilot study and herein we report our preliminary
findings. Second, a technical limitation may be due to
employment of a 1.5T MR system. Possibly higher
field strengths of 3T may widen the quantitative R2*
spectrum and perhaps have better statistical signifi-
cance. The choice of a 1.5T system was based on well
established MR image quality results for body imaging
and well established values of T2 and T2* for normal
liver tissue. A further limitation may be usage of nasal
oxygen as methods to induce changes in tumoral oxy-
genation. Perhaps this does not have the desired
impact, and there may be a need to explore stronger
stimuli such as carbogen or CO2 breathing. Further-
more, they may be limitations to detecting or demon-
strating hypoxia in HCC on the basis of fast blood
flow in these mostly hypervascular tumors that may
not allow enough time for hypoxia to be reflected in
the BOLD signal. Limitations relating to data analysis
include inclusion of only up to a maximum of five sec-
tions for BOLD evaluation. Because many tumors
were larger than 5 cm, we may not be sampling the
entire tumor for analysis and tumoral heterogeneity
may be responsible for inaccurate oxygenation status
estimation. There is a technical limitation to the num-
ber of slices that can be obtained in a single breath-
hold given the nature of multi-echo gradient echo ac-
quision. The move to free breathing or triggered
acquisitions instead of breathheld acquisitions would
result in temporal and spatial discrepancies and
could only worsen the results. Finally, a significant li-
mitation could revolve around the simplistic assump-
tion that BOLD contrast necessarily reflects hypoxia
that necessarily correlates with MVI. The lack of cor-
relation between BOLD MR and MVI might be
accounted for by the complexity of hypoxia. Factors
such as blood volume will affect BOLD contrast and
may interfere with the depiction of hypoxia. Hypoxia
is also not a static entity, but can be rather than
dynamic with perhaps acute and chronic hypoxic
states affecting BOLD contrast differently impacting
final results.

In conclusion, BOLD MR imaging does not appear to
be promising as an accurate method for preopera-
tive prediction of MVI in HCC under the circumstan-
ces wherein it was tested by us. Morphological imag-
ing is already known to be unable to determine MVI
as well. The prediction of MVI by existing functional
and or morphological imaging methods remains
elusive.
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