Published in final edited form as:

Magn Reson Med. 2015 May ; 73(5): 1812–1819. doi:10.1002/mrm.25301.

SWIFT MRI Enhances Detection of Breast Cancer Metastasis to the Lung

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Abstract

Purpose—To evaluate the capability of longitudinal MR scans using sweep imaging with Fourier transformation (SWIFT) to detect breast cancer metastasis to the lung in mice.

Methods—Mice with breast cancer metastatic to the lung were generated by tail vein injection of MDA-MB-231-LM2 cells. Thereafter, MR imaging was performed every week using three different pulse sequences: SWIFT (TE~3 μ s), concurrent dephasing and excitation (CODE; TE~300 μ s), and 3D gradient echo (GRE; TE=2.2 ms). Motion during the long SWIFT MR scans was compensated for by rigid-body motion correction. Maximum intensity projection (MIP) images were generated to visualize changes in lung vascular structures during the development and growth of metastases.

Results—SWIFT MRI was more sensitive to signals from the lung parenchyma than CODE or 3D GRE MRI. Metastatic tumor growth in the lungs induced a progressive increase in intensity of parenchymal signals in SWIFT images. MIP images from SWIFT clearly visualized lung vascular structures and their disruption due to progression of breast cancer metastases in the lung.

Conclusion—SWIFT MRI's sensitivity to fast-decaying signals and tolerance of magnetic susceptibility enhances its effectiveness at detecting structural changes in lung parenchyma and vasculature due to breast cancer metastases in the lung.

Keywords

breast cancer metastasis; lung MRI; ultrashort TE; sweep imaging with Fourier transformation (SWIFT); concurrent dephasing and excitation (CODE); rigid body motion correction

INTRODUCTION

Metastatic breast cancer is the primary cause of the mortality in women diagnosed with breast cancer. Approximately 25–30 % of women treated for primary breast cancer are

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likely to have metastatic recurrence of the disease (1). The lung is one of the common sites of breast cancer metastasis, and prognosis is very poor once metastatic breast cancer develops (2,3). CT is the standard tool for assessing lung lesions in clinical and preclinical settings (4,5). Pulmonary imaging with CT provides a high contrast between air and soft tissues that enables the high resolution of anatomical detail and detection of lung lesions. However, the radiation exposure associated with CT is a risk factor for developing cancer (6). The use of CT is especially problematic for longitudinal investigations of cancer progression and monitoring response to therapies because repeated CT scans may lead to the accumulation of radiation-induced genetic damage, making it difficult to evaluate the effects of treatment. Therefore, non-invasive methods that do not use ionizing radiation are desirable.

MRI is an excellent tool for visualizing soft tissues. Until recently, however, MRI has played a limited role in pulmonary imaging because of inherent difficulties that include 1) low proton density in lung parenchyma, 2) susceptibility differences between air and pulmonary tissues, and 3) cardiac and respiratory motion. The first two properties make the signals from pulmonary tissues weak and fast-decaying (7,8). Indeed, the decay time constant (T_2^*) is often shorter than the minimum achievable echo time (TE) in conventional MRI sequences, including gradient echo (GRE) (~1 ms). Therefore, studies of lung using GRE have focused mainly on diseased tissues (e.g., tumor mass, inflammatory lesions, and emphysematous tissues) that appear as bright regions against a dark background of normal pulmonary tissues (9–11).

Non-standard MRI techniques known as ultra-short echo time (UTE) and zero echo time (ZTE) sequences have recently gained popularity in pulmonary imaging and achieved visualization of normal lung tissues with TEs far less than 1 ms (12–15). UTE and ZTE enable signal acquisition almost immediately after excitation with free induction decay (FID)-based acquisition. Sweep imaging with Fourier transformation (SWIFT) is another FID-based MRI technique (16). Based on its achievable TE, SWIFT is expected to provide comparable performance to UTE and ZTE in lung MRI; however, it uses a different excitation and acquisition strategy from UTE and ZTE. Excitation and acquisition are performed in separate time windows in UTE and ZTE, but they are quasi-simultaneous in SWIFT. A gapped frequency-modulated (FM) pulse enables time-shared excitation and acquisition by switching the transmit and receive mode alternately. Therefore, the length of the pulse is not a limiting factor for setting the TE in SWIFT (i.e., any length of the pulse (any flip angle) can be used while keeping the same TE).

In this study, we evaluated SWIFT MRI for detection of metastases and monitoring progression of metastases in the lung of mice. The breast cancer cell line MDA-MB-231-LM2 was selected for its propensity to establish lung metastases in vivo (17). The capability of SWIFT for visualizing pulmonary tissues with extremely short T_2^* was evaluated. Tumor growth and progression were evaluated by assessing the vascular structure.

METHODS

Materials

All reagents for tissue culture were purchased from Life Technologies (Grand Island, NY, USA), Corning Incorporated Life Sciences (Tewksbury, MA, USA), and Corning Life Sciences - Mediatech Inc. (Manassas, VA, USA).

Cell culture

MDA-MB-231-LM2 (17), a lung seeking metastatic variant of the basal-like triple negative breast cancer cell line MDA-MB-231, were cultured in Dulbecco's modified Eagles's medium supplemented with 10% fetal bovine serum. The MDA-MB-231 cell line was originally derived from the pleural effusion of a patient with metastatic breast cancer (18). MDA-MB-231-LM2 cells express firefly luciferase that allows noninvasive bioluminescence imaging (BLI) of metastases to lung and were obtained from Dr. Joan Massagué of the Howard Hughes Medical Institute, Memorial Sloan Kettering Cancer Center (New York, NY, USA).

Animal studies and model of breast cancer metastasis

All animal studies and care were approved by the University of Minnesota's Institutional Animal Care and Use Committee (IACUC) and were in accordance with the procedures detailed in the Guide for Care and Use of Laboratory Animals. MDA-MB-231-LM2 cells were injected in the tail vein of female athymic mice (Harlan Laboratories, Indianapolis, IN, USA) as a model of breast cancer metastasis to the lung (19). These cells form many metastatic colonies in the lung within 4–5 weeks after tail vein injection (17). 7.5×10^5 cells in 200 μ l of phosphate-buffered saline were injected into the tail vein of 5–6 week old female mouse ($n=4$).

MRI

All images were acquired with a 9.4T 31cm bore animal MR scanner equipped with 40 gauss/cm gradients (Agilent Technologies, Inc., CA, USA). A butterfly-shaped quadrature surface coil (diameter of each loop = 2 cm) tuned to ^1H resonance frequency (400.2 MHz) was used for transmit and receive. MR scans were performed on control mice without lung metastases ($n=3$) and on mice with lung metastases ($n=4$) with three different pulse sequences: SWIFT, CODE, and conventional 3D GRE. Mice injected with cells were imaged once before injection of cells and then once every week for 4 or 5 weeks thereafter. Of the four mice with lung metastases, two died from severe tumor burden after the MR scan at 4 weeks. Mice were laid prone on a custom-built Teflon holder and the coil was placed on the dorsal side over the thorax region. During the MR scan, mice were anesthetized with 1.5–2 % isoflurane mixed with oxygen and nitrous oxide ($\text{O}_2:\text{N}_2\text{O}=1:1$). Mice breathed freely during the MR scans using a nose cone. Core body temperature was maintained at 37 ± 1 °C with a home-built water circulation heating and warm air circulation system that was regulated by the temperature monitoring module of the small animal monitoring system (SA Instruments, Inc., Stony Brook, NY, USA). MRI acquisition was

synchronized with the small animal respiration monitor. Respiratory rates of mice were between 60–90 breaths/min.

Respiratory-gated acquisition was used for minimizing artifacts from respiratory motion during MR scans (Fig.1a). In SWIFT, spherical k-space is sampled in a radial manner by changing the gradient direction as is done in conventional radial MRI sequences (20). In the gated acquisition, 100 views were acquired in every respiratory cycle, in which an acquisition window was set in the plateau region in the expiratory phase. In each SWIFT acquisition, quasi-simultaneous (time-shared) excitation and acquisition were performed with a gapped FM pulse (Fig.1b) (16); a gapped hyperbolic secant (HS) pulse (21) with pulse width $T_p=1.024$ ms and frequency sweep $bw=125$ kHz was employed. Frequency of the pulse gapping was set to 125 kHz (duration of one pulse unit (dw), which was composed of one pulse segment and one gap, was 8 μ s). Signal acquisition was performed in gaps in the pulse by switching the transmit and receive mode in an order of 1 microsecond, which enabled a couple of microseconds of TE (Fig.1c). In each pulse unit, a short delay τ_d was inserted before the receiver gate opening to avoid signal contamination from coil ringing, which provided $\sim 3\mu$ s of TE herein. Signal acquisition continued after the pulse. Time fraction of the pulse over the acquisition time T_{acq} was $T_p/T_{acq}=0.5$. Repetition time (TR) was obtained by adding time for changing the gradient direction to acquisition time $T_{acq}+\tau$ ($=2.348$ ms). The other sequence parameters were as follows: nominal flip angle (FA) = 10° , field of view (FOV) = $40\times 40\times 40$ mm³ and number of views = 344,064 – 589,824 (336 – 576 spirals). The number of views was adjusted in each experiment (mouse) such that the total time under anesthesia did not exceed 3 hours, especially for mice in the later stages of tumor growth. The scan time was 60–80 min depending on the number of views and respiration rate.

For comparison with SWIFT, a short TE radial GRE known as CODE (22) and conventional Cartesian 3D GRE were performed. In CODE, asymmetric gradient echo acquisition can achieve down to approximately 200 μ s of TE (22). A sinc pulse with $T_p=200$ μ s and $bw\approx 60$ kHz was employed as an excitation pulse in CODE. Sequence parameters in CODE were as follows: TE=296 μ s, TR=3.32 ms, nominal FA= 18° , number of views=131,072 (128 spirals), FOV= $40\times 40\times 40$ mm³, $sw=62.5$ kHz, 82 views/respiration trigger and scan time = 20–30 min. In 3D GRE, a Gaussian pulse with $T_p=1$ ms was used for excitation. Sequence parameters for 3D GRE were as follows: TE=2.18 ms, TR=4.35 ms, nominal FA= 22° , FOV= $40\times 40\times 40$ mm³, $sw=156$ kHz, matrix= $256\times 256\times 256$ and 64 acquisitions/respiration trigger. The Cartesian k-space was segmented to 64 regions along one phase encoding dimension, and k-space lines were filled from the center out in each triggered acquisition (i.e., the k-space center region was filled with the first echoes). The scan time for 3D GRE was 15–20 min.

For all sequences, the FAs were determined experimentally and set to a relatively high angle so as to cover the entire lung using the surface coil; such high FAs were needed to get strong enough signals from lung regions far from the coil, whereas the regions close to the coil suffered SNR loss because the flip angle was larger than the optimal value (Ernst angle). Due to the relatively high FAs and short TRs, the images from the three sequences were consistently heavily T₁-weighted. To compare sensitivity of the three sequences to fast-

decaying signals from lung parenchyma, the SNR for lung parenchyma was determined for SWIFT, CODE and 3D GRE from MR images of normal lung by setting a common region of interest. Due to differences in sequence parameters, corrections to each measured SNR were made to account for differences in total scan time and receiver bandwidth. For SWIFT, compensation for the lower receiver duty cycle effect due to the time-shared excitation and acquisition was performed as described previously (23). Although corrected SNR values still contained contributions from unknown factors such as TR and FA differences, the SNR values should reasonably reflect the relative sensitivities to fast-decaying signals.

Image reconstruction

SWIFT image reconstruction was performed by 2 step processing (Fig. 2). Each signal acquired in SWIFT is a FID-projection convolved with the pulse (16). The FID-projection was retrieved by performing a deconvolution manipulation in the frequency domain (23). The obtained signals were sent to the following motion estimation process. All reconstruction and motion correction programs were written with C language. (Some of the SWIFT pulse sequence and image reconstruction codes used herein are available in the CMRRpack package: <http://www.cmrr.umn.edu/swift/>).

In radial acquisition, dense sampling around the k-space center inherently plays a role of self-navigation (24). The radial k-space data were divided into 14–24 time frame datasets. From each frame dataset, a 3D image was reconstructed. The images were transformed back to k-space domain, and then motion relative to one template frame chosen from the 14–24 datasets was estimated. In the motion estimation, the mouse body was assumed as a rigid body in 3D space (6 degrees of freedom). The estimation was performed for rotational and translational motion step by step separately (25). In motion estimation, the fully sampled k-space center region based on the Nyquist criterion was used. In the rotational motion estimation, the magnitude of each k-space subset was fitted to the template with 3 rotations around x, y, and z. After rotational motion estimation, translational motion in image domain was estimated by fitting a first order linear distribution to the phase difference between the rotated k-space and template in the weighted least squares method. On the basis of the estimated linear phase terms, the translational motion was corrected by removing the linear phase in k-space domain. Finally, root mean square error (RMSE) between the motion corrected and template k-space was calculated to evaluate goodness of k-space fitting.

After estimating rigid-body motion, the obtained motion parameters and RMSEs were sent to the image reconstruction pipeline (Fig. 2). First, the RMSEs (i.e., the rigid-body assumption) were evaluated by comparing a preassigned threshold; the time frame datasets that exceeded the threshold were eliminated from image reconstruction. The fraction of the data eliminated in this step was 4.4 % on average. For the data passing through the first step, motion correction was performed in radial k-space domain using the parameters in motion estimation. The motion corrected data were gridded onto a Cartesian k-space and reconstructed to a 3D image with Fourier transformation. The final image size was 288×288×288 (139 μ m isotropic resolution).

In CODE reconstruction, processing after the phase correction was the same as it was for SWIFT, but motion correction was not applied, since improvement of the image quality was

not seen. The matrix size of the CODE images was $256 \times 256 \times 256$ (156 μm isotropic resolution).

Image processing

The reconstructed images showed intensity variation along the dorsal-ventral axis of the mouse due to the B_1 field variation along the depth direction. To alleviate the intensity variation along the coil depth direction, the reconstructed image was divided by the image filtered with a Gaussian low-pass filter (before image division, a constant value was added to the filtered image to avoid enhancement of background noise).

Visualization of vasculature

In MR images of the lung, blood vessels appeared as bright signals relative to parenchymal tissues. Maximum intensity projection (MIP) was calculated to visualize vascular structure. The lung region in the images was extracted by manually masking out all non-lung tissues (e.g., muscle and fat under the skin). MIP images were calculated for the masked lung images.

Histology

After the final MRI, mice were euthanized and lungs were inflated with formalin and harvested. Lungs were fixed overnight in 10% buffered formalin, paraffin embedded, and cut into 5 μm sections. Sections were stained with hematoxylin and eosin (H&E). A Leica DM4000 B microscope and Leica Application Suite software (Leica Microsystems Inc., Wetzlar, Germany) was used to generate images of H&E stained sections of lungs from mice without injected breast cancer cells and mice with lung metastases.

RESULTS

MRI of normal mouse lung without breast cancer metastases

MRI of the lungs of control mice without breast cancer metastases were acquired with SWIFT, CODE, and 3D GRE (Fig. 3a–i). SWIFT was more sensitive to signals from pulmonary tissues (i.e., alveoli and interstitium) than CODE and 3D GRE because of its extremely short TE of $\sim 3 \mu\text{s}$ (Fig. 3a–f). The relatively short TE in CODE ($\sim 300 \mu\text{s}$) preserved pulmonary signals; however, visualization of pulmonary tissues with CODE was limited. 3D GRE failed to capture pulmonary signals (TE $\sim 2.2 \text{ ms}$). MIP images from SWIFT visualized fine vascular structures including small blood vessels due to its tolerance of susceptibility effects (Fig. 3g). Although MIP images for CODE and 3D GRE showed large vessels, strong susceptibility effects caused the small vessels to be nearly invisible (Fig. 3h,i). MIP images projected in different directions are shown in Supporting Information (Fig. S1). SWIFT was more sensitive to parenchymal signals than the other two sequences as determined by SNR values corrected for total scan time, receiver bandwidth and receiver duty cycle (Fig. 4). The SNR of CODE was remarkably higher than that obtained with 3D GRE.

BLI and MRI of mouse lung with breast cancer metastases

Growth of metastases in the lungs was monitored by BLI at 1 hour, day 2, and weeks 1–5 after injection of metastatic breast cancer cells and these data demonstrate that lung colonization of metastatic breast cancer cells and metastatic tumor burden increased over time (data not shown).

Growth of metastases in the lungs was monitored by MRI on weeks 1–5 after injection of metastatic breast cancer cells (Fig. 5). During the early stages of tumor growth (1–2 weeks), images of the lung acquired with SWIFT, CODE, and 3D GRE were similar to those from normal mice (Fig. 5a–f). As time elapsed, pulmonary tissues (i.e., interstitium and alveoli) showed progressive inhomogeneous intensity variation and some bright spots in SWIFT images by weeks 3 and 4 (Fig. 5g,j); almost the entire lung looked abnormal by 4 weeks post-injection of cells. In CODE images, signal intensities in pulmonary tissues gradually increased as the metastases grew from week 2–3 (Fig. 5h,k). Some regions were visible even with 3D GRE at week 4, but the majority of pulmonary tissues were still invisible (Fig. 5l). At week 5, the structure of lung pulmonary tissues was severely disrupted and almost all regions were visible with all sequences (Fig. 5m–o).

Visualization of vasculature

MIP images from SWIFT, CODE and 3D GRE clearly visualized the progress of metastatic tumor growth in the lung (Fig. 6). MIP images projected in different directions are shown in Supporting Information (Fig. S2). In SWIFT MIP images, contrast between blood vessels and pulmonary tissues gradually decreased as the tumor cells colonized and proliferated in the lung (i.e., signals from pulmonary tissues increased relative to signals from blood vessels). At 4 and 5 weeks, the lung vasculature was severely disrupted. CODE MIP images showed similar trend to SWIFT: pulmonary tissue signal intensities increased along with tumor growth, and some granular spots were seen at 3–4 weeks post-injection of cells, but these changes were more conspicuous in the SWIFT MIP images. MIP images from 3D GRE did not show signs of tumor growth and metastases at weeks 2–3 because of low pulmonary tissue signal intensities. Thus, MIP images from SWIFT visualized fine structural changes better than the other two techniques because of its extremely short TE and immunity to susceptibility effects. The lung volume of the mice with lung metastases monotonically increased as the lung metastases grew (see Supporting Information Fig. S3).

Histological evaluation of metastases of breast cancer

To verify that mice injected with cells developed lung metastases, H&E stained lung sections were examined by light microscopy. Normal control lungs showed no metastases (Fig. 7a), whereas lungs from mice 5 weeks after injection of cells showed increased cellularity due to many macroscopic and microscopic metastases (Fig. 7b).

DISCUSSION

One of the primary difficulties with lung MRI is visualizing short T_2^* signals from pulmonary tissues. In the current study, we tested the sensitivity to pulmonary signals at 9.4 T for three MRI sequences with different TEs: 3 μ s, 300 μ s, and 2.2 ms for SWIFT, CODE

and 3D GRE, respectively. SWIFT showed higher sensitivity to pulmonary signals than CODE or 3D GRE. CODE suffered from considerable signal loss despite its much shorter TE as compared to 3D GRE. According to previous reports, the T_2^* value for lung parenchyma in normal mice is $\sim 500 \mu\text{s}$ at 4.7 T (26,27); the T_2^* value for lung parenchyma is expected to be comparable to or even less than TE in CODE (300 μs) at 9.4 T. The extremely short TE in SWIFT can minimize effects from the fast signal decay due to the short T_2^* and push lung MRI toward higher field strengths, which makes it possible to take advantage of the higher SNR of high field MRI.

Along with the T_2^* shortening, the large susceptibility difference in pulmonary tissues is another difficulty with lung MRI. The large susceptibility difference disturbs B_0 field homogeneity (i.e., off-resonance). MIP images from SWIFT visualized the lung vasculature more precisely than CODE and 3D GRE. There is a large susceptibility difference expected between blood vessels and pulmonary tissues. The extremely short TE in SWIFT achieves almost perfect coherence at the center of k-space for all isochromats regardless of their off-resonance frequencies, and the off-resonance phase evolves from the center out; therefore, the k-space center region is nearly free from off-resonance (28). In contrast, the k-space center in CODE is impacted by off-resonance because the refocus time of the gradient echo for each local isochromat is dependent on its off-resonance frequency, which induces errors in k-space center registration. Moreover, the lower bandwidth in the current CODE setting is another source of the severe off-resonance blurring relative to the SWIFT images.

A gradual increase in pulmonary tissue signal intensities was found in SWIFT and CODE MIP images from 2–3 weeks as the tumors colonized the lungs. During the early stage of the metastatic cascade, breast cancer cells reach the lung following injection in the tail vein and extravasate out of blood vessels. As the surviving cancer cells grow and colonize the lung, the cancer cells attract their own blood supply (angiogenesis), which promotes hypervascularity (3,29). This tumor-induced vascular remodeling leads to a gradual increase in water content (blood fraction) and probably a reduction in susceptibility effects in lung, resulting in an increase in the pulmonary signals. At later metastatic stages, growing tumor cells disrupt the pulmonary structure and develop into high-cell density masses that induce an additional increase in the pulmonary tissue signal. Such tumor masses were visible even with conventional 3D GRE at 4–5 weeks after injection of cells.

The lung volume of mice harboring breast cancer metastases in the lung increased monotonically as time elapsed (Fig. S3). A previous longitudinal volumetric analysis of normal mouse lung with CT revealed that lung volume is stable after 6 weeks from birth in two mouse strains (30). Therefore, the volume increase seen in this study in the lungs of mice with breast cancer metastases (>5–6 weeks) reflects changes associated with metastatic disease (e.g., inflammatory response), although the mouse strain herein was different from the ones in the previous report. The increase in lung volume was accelerated around 3 weeks after injection of breast cancer cells, which corresponded with the time that metastases were visible by MRI (Fig. 5). Aggressive metastatic growth would be expected to accelerate the increase in lung volume. In this study, we included visual inspection and volumetric analyses of lung and avoided quantification of metastases based on signal intensity (image contrast) because of the severe B_1 field variation associated with the surface coil.

The high rate of respiratory and cardiac motion is another challenge to conducting MRI in a rodent model. Radial MRI sequences are inherently tolerant to motion artifacts (31). However, inconsistency among radial projections induces image blurring and reduction of signal-to-noise ratio. In this study, in addition to the respiratory-gated acquisition, rigid-body motion correction was retrospectively applied in SWIFT reconstruction using its inherent self-navigation property. In the current setting, motion correction was performed focusing on relatively slow motion (every 3–4 minutes). The time resolution is adjustable depending on time scale of the motion by changing the number of time frames in the motion estimation. High temporal resolution makes the fully sampled center region small which can result in degradation of the estimation accuracy. However, previous work for translational motion correction with self-navigation demonstrated that even if the k-space sampling pattern is not preserved, the rigid-body motion correction works well when the motion between successive acquisitions (time frames herein) is small and the sampled k-space subsets have an adequate amount of correlation (32). These assumptions are reasonable in the radially sampled k-space, provided the size of the k-space region used in motion estimation is selected appropriately.

CONCLUSIONS

Longitudinal MRI for mice with breast cancer metastasis to the lung was performed with three pulse sequences: SWIFT, CODE, and 3D GRE. In SWIFT image reconstruction, motion during the long MR scans was corrected by rigid-body motion correction using the inherent self-navigation property. For normal mouse lungs, SWIFT was better at preserving fast-decaying signals from lung pulmonary tissues than the other two techniques. Weekly MR scans of mice with breast cancer metastases in the lung visualized metastatic disease and disruption of lung structures within 3 weeks of initial tumor seeding in the lung. Furthermore, the MIP images from SWIFT clearly visualized the fine vascular structures and their disruption by metastatic breast cancer growth. Lung volume measurements based on MRI revealed a monotonic increase in lung volume as the lung structures were disrupted by tumor growth. In conclusion, SWIFT MRI visualizes lung structural change and metastases better than GRE and CODE and is an effective imaging technique for detecting metastatic breast cancer progression in the lung.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

ACKNOWLEDGEMENTS

Grant sponsor: Susan G. Komen for the Cure; Grant number: KG110716

Grant sponsor: NIH; Grant numbers: P41 RR008079 and P41 EB015894

Grant Sponsor: Prospect Creek Foundation

Grant sponsor: WM KECK Foundation

The authors thank Lynn Utecht (University of Minnesota) for assistance with mouse anesthesia monitoring and Michael Franklin (Department of Medicine, University of Minnesota) for editorial assistance.

REFERENCES

1. Clarke M, Collins R, Darby S, et al. Effects of radiotherapy and of differences in the extent of surgery for early breast cancer on local recurrence and 15-year survival: an overview of the randomised trials. *Lancet*. 2005; 366(9503):2087–2106. [PubMed: 16360786]
2. Kennecke H, Yerushalmi R, Woods R, Cheang MC, Voduc D, Speers CH, Nielsen TO, Gelmon K. Metastatic behavior of breast cancer subtypes. *J Clin Oncol*. 2010; 28(20):3271–3277. [PubMed: 20498394]
3. Nguyen DX, Massague J. Genetic determinants of cancer metastasis. *Nat Rev Genet*. 2007; 8(5): 341–352. [PubMed: 17440531]
4. Gotway MB, Reddy GP, Webb WR, Elicker BM, Leung JW. High-resolution CT of the lung: patterns of disease and differential diagnoses. *Radiol Clin North Am*. 2005; 43(3):513–542. viii. [PubMed: 15847814]
5. Kirsch DG, Grimm J, Guimaraes AR, et al. Imaging primary lung cancers in mice to study radiation biology. *Int J Radiat Oncol Biol Phys*. 2010; 76(4):973–977. [PubMed: 20206017]
6. Kubo T, Lin PJ, Stiller W, Takahashi M, Kauczor HU, Ohno Y, Hatabu H. Radiation dose reduction in chest CT: a review. *AJR Am J Roentgenol*. 2008; 190(2):335–343. [PubMed: 18212218]
7. Case TA, Durney GH, Ailion DC, Cuttillo AG, Morris AH. A Mathematical Model of Diamagnetic Line Broadening in Lung Tissue and Similar Heterogeneous Systems: Calculations and Measurements. *J Magn Reson*. 1987; 73:304–314.
8. Kveder M, Zupancic I, Lahajnar G, Blinc R, Suput D, Ailion DC, Ganesan K, Goodrich C. Water proton NMR relaxation mechanisms in lung tissue. *Magn Reson Med*. 1988; 7(4):432–441. [PubMed: 3173058]
9. Garbow JR, Wang M, Wang Y, Lubet RA, You M. Quantitative monitoring of adenocarcinoma development in rodents by magnetic resonance imaging. *Clin Cancer Res*. 2008; 14(5):1363–1367. [PubMed: 18316556]
10. Tigani B, Schaeublin E, Sugar R, Jackson AD, Fozard JR, Beckmann N. Pulmonary inflammation monitored noninvasively by MRI in freely breathing rats. *Biochem Biophys Res Commun*. 2002; 292(1):216–221. [PubMed: 11890695]
11. Quintana HK, Cannet C, Zurbrugg S, Ble FX, Fozard JR, Page CP, Beckmann N. Proton MRI as a noninvasive tool to assess elastase-induced lung damage in spontaneously breathing rats. *Magn Reson Med*. 2006; 56(6):1242–1250. [PubMed: 17029230]
12. Johnson GA, Cofer GP, Hedlund LW, Maronpot RR, Suddarth SA. Registered (1)H and (3)He magnetic resonance microscopy of the lung. *Magn Reson Med*. 2001; 45(3):365–370. [PubMed: 11241691]
13. Kuethe DO, Adolphi NL, Fukushima E. Short data-acquisition times improve projection images of lung tissue. *Magn Reson Med*. 2007; 57(6):1058–1064. [PubMed: 17534926]
14. Zurek M, Bessaad A, Cieslar K, Cremillieux Y. Validation of simple and robust protocols for high-resolution lung proton MRI in mice. *Magn Reson Med*. 2010; 64(2):401–407. [PubMed: 20665784]
15. Bianchi A, Lux F, Tillement O, Cremillieux Y. Contrast enhanced lung MRI in mice using ultra-short echo time radial imaging and intratracheally administrated Gd-DOTA-based nanoparticles. *Magn Reson Med*. 2013; 70(5):1419–1426. [PubMed: 23233439]
16. Idiyatullin D, Corum C, Park JY, Garwood M. Fast and quiet MRI using a swept radiofrequency. *J Magn Reson*. 2006; 181(2):342–349. [PubMed: 16782371]
17. Minn AJ, Gupta GP, Siegel PM, Bos PD, Shu W, Giri DD, Viale A, Olshen AB, Gerald WL, Massague J. Genes that mediate breast cancer metastasis to lung. *Nature*. 2005; 436(7050):518–524. [PubMed: 16049480]
18. Cailleau R, Olive M, Cruciger QV. Long-term human breast carcinoma cell lines of metastatic origin: preliminary characterization. *In Vitro*. 1978; 14(11):911–915. [PubMed: 730202]
19. Sachdev D, Zhang X, Matise I, Gaillard-Kelly M, Yee D. The type I insulin-like growth factor receptor regulates cancer metastasis independently of primary tumor growth by promoting invasion and survival. *Oncogene*. 2010; 29(2):251–262. [PubMed: 19838209]

20. Lauterbur PC. Image Formation by Induced Local Interactions: Examples Employing Nuclear Magnetic Resonance. *Nature*. 1973; 242:190–191.
21. Silver MS, Joseph RI, Chen CN, Sank VJ, Hoult DI. Selective population inversion in NMR. *Nature*. 1984; 310(5979):681–683. [PubMed: 6472448]
22. Park JY, Moeller S, Goerke U, Auerbach E, Chamberlain R, Ellermann J, Garwood M. Short echo-time 3D radial gradient-echo MRI using concurrent dephasing and excitation. *Magn Reson Med*. 2012; 67(2):428–436. [PubMed: 21702064]
23. Garwood, M.; Idiyatullin, D.; Corum, CA., et al. Capturing Signals from Fast-Relaxing Spins with Frequency-Swept MRI: SWIFT. In: Bydder, Graeme M.; Fullerton, Gary; Young, Ian R., editors. *MRI of Tissues with Short T2s or T2*s*. New York: John Wiley & Sons; 2013. p. 125–142.
24. Dadok J, Sprecher RF. Correlation NMR spectroscopy. *J. Magn. Reson*. 1974; 13:243–248.
25. Schaffter T, Rasche V, Carlsen IC. Motion compensated projection reconstruction. *Magn Reson Med*. 1999; 41(5):954–963. [PubMed: 10332879]
26. Welch EB, Manduca A, Grimm RC, Ward HA, Jack CR Jr. Spherical navigator echoes for full 3D rigid body motion measurement in MRI. *Magn Reson Med*. 2002; 47(1):32–41. [PubMed: 11754440]
27. Beckmann N, Tigani B, Mazzoni L, Fozard JR. MRI of lung parenchyma in rats and mice using a gradient-echo sequence. *NMR Biomed*. 2001; 14(5):297–306. [PubMed: 11477650]
28. Olsson LE, Lindahl M, Onnervik PO, Johansson LB, Palmer M, Reimer MK, Hultin L, Hockings PD. Measurement of MR signal and T2* in lung to characterize a tight skin mouse model of emphysema using single-point imaging. *J Magn Reson Imaging*. 2007; 25(3):488–494. [PubMed: 17279537]
29. Lehto LJ, Sierra A, Corum CA, Zhang J, Idiyatullin D, Pitkanen A, Garwood M, Grohn O. Detection of calcifications in vivo and ex vivo after brain injury in rat using SWIFT. *Neuroimage*. 2012; 61(4):761–772. [PubMed: 22425671]
30. Norton L, Massague J. Is cancer a disease of self-seeding? *Nat Med*. 2006; 12(8):875–878. [PubMed: 16892025]
31. Mitzner W, Brown R, Lee W. In vivo measurement of lung volumes in mice. *Physiol Genomics*. 2001; 4(3):215–221. [PubMed: 11161000]
32. Glover GH, Pauly JM. Projection reconstruction techniques for reduction of motion effects in MRI. *Magn Reson Med*. 1992; 28(2):275–289. [PubMed: 1461126]
33. Mendes J, Kholmovski E, Parker DL. Rigid-body motion correction with self-navigation MRI. *Magn Reson Med*. 2009; 61(3):739–747. [PubMed: 19097240]

Figure 1. Pulse sequence diagram of SWIFT with respiratory-gated acquisition (a) and magnification of one repetition time (TR) region (b) and pulse gapping (c).

Figure 2.

Flowchart of SWIFT image reconstruction processing. The SWIFT image reconstruction was performed by 2 step processing: 1) rigid-body motion estimation (shaded box) and 2) main image reconstruction pipeline. Based on the estimated motion, motion correction processing was done in the radial k-space domain (before gridding onto a Cartesian k-space).

Figure 3.

MR images of control lung of a mouse without lung metastases (**a–f**) acquired with SWIFT, CODE, and 3D GRE and their maximum intensity projection (MIP) images (**g–i**). Coronal slices of the lung are shown in **a–f**. Extremely short TE ($\sim 3 \mu\text{s}$) in SWIFT preserved signals from lung parenchyma (**a,d**) better than CODE (**b,e**) or GRE (**c,f**) ($\text{TE}=300 \mu\text{s}$ and 2.2 ms). In GRE, majority of lung parenchyma appeared dark. Lung vasculature was seen in MIP images (**g–i**). Fine vascular structures including small vessels were visualized better by

SWIFT (**g**) compared to CODE (**h**) or GRE (**i**). Images shown are from the same mouse and are representative of images acquired for 3 mice.

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Figure 4.

Comparison of SNR for SWIFT, CODE and 3D GRE for lung parenchyma in normal lung.

The graph shows both the SNR measured directly from images (raw SNR) and corrected for differences in total scan time, receiver bandwidth and receiver duty cycle (corrected SNR).

The values were normalized with the raw SNR for SWIFT.

Figure 5.

MR images of lung metastases from a single mouse monitored at the indicated times after injection of MDA-MB-231-LM2 cells. Coronal slices of images of the lung acquired with SWIFT (**a,d,g,j,m**), CODE (**b,e,h,k,n**), and 3D GRE (**c,f,i,l,o**) are shown. At week 1, images from all sequences were similar to normal mice (**a–c**). As time elapsed, lung tissues progressively showed inhomogeneous intensity variations in images from SWIFT (**d,g,j**). A gradual increase in parenchymal signal intensities was seen in CODE images (**e,h,k**). At week 4, some bright spots representing metastatic cell masses were seen in images from

SWIFT (**j**) and CODE (**k**). In contrast, lung tissues were invisible with GRE at week 2 and 3 (**f,i**). Some parts of lung tissues were visualized with GRE at week 4, but the majority of the lung tissues appeared dark (**l**). At week 5, lung tissues were mostly visible with all sequences (**m–o**). All images shown here are from the same mouse as in Figure 4 and are representative of data acquired for 4 mice.

Figure 6.

Maximum intensity projection (MIP) images of lung metastases from a single mouse at the indicated times after injection of breast cancer cells. MIP images from SWIFT (**a,d,g,j,m**), CODE (**b,e,h,k,n**), and 3D GRE (**c,f,i,l,o**) were generated from the lung MR images in Figure 5. As time elapsed, signals from lung tissues gradually increased in MIP images from SWIFT (left) and CODE (center). At week 3 and 4, spots representing lung metastases were visible in images from SWIFT (**g,j**) and CODE (**h,k**), but changes of fine structures were more clearly visualized in SWIFT MIP images especially around small vessels. In contrast,

structural changes due to metastases could not be detected with GRE until week 3 (**c,f,i**). At week 4, some bright spots appeared in GRE MIP images (**l**). At week 5, disruption of entire lung structures was clearly seen in MIP images from all three sequences (**m,n,o**). MIP images shown here are from the same mouse as in Figure 5 and are representative of data acquired for 4 mice.

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Figure 7.

H&E stained sections of control lung from a mouse that was not injected with cells **(a)** and lung from a mouse with breast cancer metastases in the lung 5 weeks after injection of cells **(b)**. The H&E-stained section of lung in (b) is from the mouse whose MRI data are shown in Figs. 5 and 6.