**Research Strategy (6 pages)**

**A. Significance**

**A.1 Soft tissue tumors**

Soft tissue tumors may arise from connective tissues including muscle, tendons, fat, lymph and blood vessels, and nerves. These tumors may be benign, locally aggressive, or malignant, and while rare in older populations, are relatively common in younger people (<30 years old) (REF ARNDT,HaDuong). The treatment of soft tissue sarcomas is multimodal, including surgery and/or radiation for primary tumors, and chemotherapy for potential metastatic disease. Advances in treatment have led to greatly improved survival rates over the last half century. However, the young patient population has increased risk of recurrence and metastases that result in poor prognoses (give numbers). Younger patients are also at risk of long-term treatment effects that severely and negatively impact quality of life and are at times life-threatening (numbers?, references).

Better treatments are still needed to improve the outcome of patients with soft tissue tumors, especially for those with aggressive, recurrent or metastatic disease, to reduce long-term complications from surgery and radiation, and to improve long-term quality of life. Magnetic resonance-guided focused ultrasound (MRgFUS) is an emerging therapy that meets this need with paradigm-changing potential in the treatment and management of soft tissue tumors of all varieties.

**A.2 MRgFUS**

MRgFUS is a completely non-invasive therapy that generates localized necrosis in diseased tissues. Ultrasound waves enter the body from an external transducer, propagate through healthy tissues without causing damage, and focus inside the target tissue, quickly increasing the local tissue temperature and inducing coagulative necrosis. Benefits of MRgFUS include limited risk of infection, fewer side effects than radiation or chemotherapy, decreased morbidity, and significantly reduced recovery times with excellent long-term outcomes [8–10]. In the last two decades, the FDA has approved the use of MRgFUS for the treatment of uterine fibroids, bone metastases, prostate disease, and essential tremor, with many other indications, including soft tissue tumors, under preclinical and clinical investigation [11–14].

Several groups have investigated the use of MRgFUS (and ultrasound-guided FUS) to treat desmoids, a non-malignant, yet aggressive soft tissue tumor [15–18]. A recent retrospective multi-center study in 15 desmoid patients found that after the initial MRgFUS treatment, the median targeted and total tumor volumes decreased by 63% and 58%, respectively, with average pain scores improving from 6  2.3 to 1.3  2 [17]. 7 of 9 patients that did not receive other therapies after MRgFUS with follow up greater than 6 months achieved durable clinical benefit (>50% reduction in viable tumor volume). These encouraging results highlight MRgFUS as a promising therapeutic technology to displace traditional therapies for the control and elimination of desmoid and other soft tissue tumors.

**A.3 Fat and focused ultrasound**

Several challenges associated with fatty tissues exist in MRgFUS therapies. The clinically utilized proton resonance frequency (PRF) technique for monitoring tissue temperatures during MRgFUS treatments does not work in fat-based tissues. While methods for monitoring fat temperatures using MR T1 or T2 properties are under development, these are not yet implemented clinically leaving fat at risk for unchecked heating and damage. Also, subcutaneous fat, fat near or surrounding the target tissue, and even fat within a tumor invariably play a role in the ultrasound pathway. Several studies have demonstrated that fat in the ultrasound beam path negatively impacts efficiency and accuracy of ultrasonic focusing and heating. While phase aberration correction (PAC) techniques can recover some of the losses associated with inhomogeneous tissues, they are not currently implemented in clinical treatments.

Another fat-specific MRgFUS challenge has been observed by interventional radiologists performing MRgFUS therapies of desmoid tumors. They have found that the T2-weighted MR signal of subcutaneous fat in the ultrasound near field increases in intensity during the treatment (see figure 1). After this change occurs, focal temperatures within the tumor decrease and getting sufficient energy to the target tissue for ablation becomes difficult. Treatment times are extended, power requirements for effective ablation rise, and treatment uncertainty increases along with the likelihood of normal tissue damage in the near field. Since MR temperature measurement within fat is difficult and not utilized clinically, it is unclear what drives this phenomenon.

A thorough characterization of fat tissue properties will provide data to improve T1- or T2-based MR thermometry in fatty tissues, inform PAC techniques for increasing MRgFUS treatment efficiency and accuracy, and offer clinicial insights regarding observed difficulties with subcutaneous fat in desmoid treatments. Altogether, these outcomes will lead to safer, faster, and better MRgFUS treatments of soft tissue tumors.

**A.4 MRgFUS treatment planning**

In practice, when a patient is preparing for MRgFUS, they attend the clinic for a MR imaging session 24-48 hours prior to treatment. In the interim between the imaging and treatment sessions, the treating clinician will prepare a plan on how the MRgFUS will proceed. Decisions during treatment planning include where to begin the treatment and the path to follow as the treatment progresses, what is the best acoustic window to minimize near- or far-field damage to healthy tissues, what power and duration should be used for each sonication. The development of the treatment plan relies heavily on the knowledge and experience of the clinician performing the MRgFUS treatment.

An underutilized opportunity exists for predictive computational models to assist the clinician in preparing the treatment plan. The models could inform the clinician on the optimal treatment path, acoustic window, powers and sonication durations. They could identify potential risks to healthy or sensitive tissues for a prescribed treatment plan. They could use *a priori* knowledge of the desired beam path to implement PAC for improved focal accuracy and power delivery. However, before such opportunities become a reality, the acoustic and thermal models used for computational models require improvement and extensive validation. Current models often overestimate the local temperature at the target tissue and underestimate the focus size. A growing body of literature is showing the value and need for temperature-dependent properties in treatment planning. However, actual values for those properties are scarce. Finally, accurate models are computationally expensive, making optimization of the treatment parameters extremely difficult given the limited time window available for treatment planning. The temperature-dependent properties and computational studies projected in this proposal will set up a framework for future efforts to make computational modeling a meaningful contributor in MRgFUS treatment planning.

**A.5 Summary and Need**

MRgFUS shows great promise for treating soft tissue tumors and reducing morbidity and long-term side effects associated with current therapies. However, fat can impact focused ultrasound treatments in several ways, reducing certainty due to difficulties in adipose MR temperature imaging, shifting focal locations or lowering intensities through phase aberration, and driving unexplained clinical phenomena in the treatment of tumors with subcutaneous fat in the ultrasound near field. This proposal aims to overcome these challenges and accelerate clinical acceptance of MRgFUS for soft tissue tumor treatments through the systematic characterization of temperature-dependent fat properties and development of tools for improved MRgFUS treatment planning.

**B. Innovation**

**B.1 Technical innovation**

**B.1.A Temperature-dependent fat properties**

* The approach described in C.1 is unique for its combination of temperature control and extensive characterization of fat properties. The experimental setups developed for this work will be useful in the temperature-dependent characterization of other tissue types.
* Identifying correlations between acoustic, thermal, MR, and mechanical properties measured in this study may provide useful surrogate information regarding ongoing efforts to develop fatty tissue temperature measurements or to non-invasively monitor for tissue damage in MRgFUS treatments.
* While the literature has room-temperature or body-temperaturevalues (IT IS foundation ref) for fat properties determined in this study, there is currently no information available regarding temperature-dependent fat properties.

**B.1.B Computational modeling for treatment planning**

* While applying temperature-dependent thermal properties for MRgFUS computational modeling is gaining some traction (Guntur, Prakash references), the lack of temperature-dependent acoustic properties in modeling is a common shortcoming of current efforts (REFS that show tighter focus).
* The computational tools proposed in C.2.J to set up a treatment planning pipeline are being developed with computational efficiency in mind, in addition to accuracy. This will bolster clinical relevance and applicability since the treatment planning timeline is often no more than a day or two.

**B.2 Impact and premise**

The tissue characterization and computational modeling proposed herein will identify why MRgFUS desmoid tumor treatments become more difficult as the treatment progresses. Opportunities to avoid or mitigate those difficulties will likely be identified with further application to other soft tumor MRgFUS therapies. New computational tools will be extensively validated through direct comparison to clinical data, increasing confidence in their applicability and utility in MRgFUS treatment planning. With further development, those tools will provide meaningful information to the treatment planning physician that will increase treatment safety, reduce treatment times, and improve treatment outcomes for the MRgFUS treatment of soft tissue tumors. Together, such improvements will accelerate the adoption of MRgFUS as an alternative therapy that reduces morbidity and increases quality of life compared to the current standard of care.

**C. Approach**

This work will characterize the temperature dependence of fat properties (Aim 1), retrospectively evaluate the impact of those properties on MRgFUS treatments (Aim 2), and establish a path toward clinical utilization of temperature-dependent fat properties in MRgFUS treatment planning (Aim 2). The completion of the two outlined specific aims will provide quantifiable results to better understand why subcutaneous fat modifies MRgFUS treatments and include the initial development of novel computational approaches that leverage this new understanding to improve treatment outcomes.

**C.1 Specific Aim 1**

As described in A.3, clinicians have observed that the T2-weighted signal intensity of subcutaneous fat increases during MRgFUS treatments while the ability to effectively deliver ultrasonic energy to deeper target tissues is reduced. Because the subcutaneous fat in the ultrasound near field is consistently exposed to low intensity ultrasound, its temperature may be gradually increasing throughout the treatment. This temperature change is difficult to observe because traditional PRF MR thermometry does not work in adipose tissues. This aim hypothesizes that increased temperatures in subcutaneous fat alter local tissue properties (for example, increasing acoustic absorption) in a manner that reduce the temperatures and sonication efficiency at the deeper target tissue.

***C.1.a. Tissue acquisition and preparation***. Freshly excised bovine fat tissue will be acquired from a local slaughterhouse. Human subcutaneous fat tissue will be acquired from local plastic surgeons. The fat will be packed into cylindrically shaped tissue holders with mylar end faces that ensure near acoustic transparency. Excess space will be filled with water to prevent air bubbles (degas before acoustic measurements?). The tissue holders will be sealed and cooled to room temperature. All property characterization will be performed within 24 hours of excision to minimize tissue degradation.

***C.1.b. Temperature control***. Water circulators (PolyScience VWR 1157, Niles, IL) and sous vides (1000-W SOUSVIDE ART, Perch, Wilmington, DE) will be used to heat and control the tissue temperature in the various property measurement setups. Thermocouples (RISEPRO 4-channel K-type digital thermocouples, Hong Kong) will be placed in the water outside the tissue holder and inside the fat to ensure temperature uniformity during property measurements. Property measurements will be made in 5 C increments from 20 C up to 70 C. Additional measurements will then be made in decreasing increments of 10 C back to room temperature to investigate potential hysteresis effects.

***C.1.c. MR properties***. Tissue samples will be placed inside the XX-channel head coil of a Siemens Tim 3T scanner (MAGNETOM Prismafit, Erlangen, Germany). For MR experiments, thermocouples will be replaced with fiberoptic temperature probes (Neoptix ReFlex 4-channel box with Neoptix T1 probes, Quebec, Canada) and temperature control will require hardware external to the MR scanner room (see Dillon 2019). The MR imaging protocol used to characterize the T1 and T2 relaxation times is shown in Table 1 (Include MR sequence, TR, TE, Matrix size, Resolution, Slices, Bandwidth, …). The anticipated accuracy of MR property measurements is 10% for both T1 and T2 relaxation times (Does Steven have a REF for this?).

***C.1.d. Acoustic properties***. Through transmission measurements will be utilized to characterize the fat’s temperature-dependent speed of sound at frequencies of 600 kHz, 1 MHz, 1.8 MHz, and 3 MHz. Radiation force balance-generated insertion-loss measurements of acoustic attenuation will be performed at frequencies of 500 KHz, 800 kHz, 1.5 MHz, and 2.8 MHz. Based on previous experimental studies, expected accuracy of the speed of sound and acoustic attenuation measurements is 0.1 and 16% respectively (Sara Johnson 2018).

***C.1.e. Thermal properties***. A commercially available thermal property analyzer (TEMPOS, METER Group, Pullman, WA) will be used to evaluate temperature-dependent thermal conductivity and thermal diffusivity. Specific heat capacity will be measured with a differential scanning calorimeter (MCDSC, TA Instruments, New Castle, DE). Anticipated accuracy for thermal conductivity and thermal diffusivity are 10% and 12%, respectively (Use Dillon 2014 as REF?), and the reported accuracy of the MCDSC is 10% (REF).

***C.1.f. Mechanical properties***. While not contributing directly to the challenge of delivering ultrasonic energy through fat, the mechanical property of shear modulus has been proposed as an acute indicator of tissue viability in MRgFUS treatments (REFS) and is included for completeness. The shear modulus will be characterized with a commercially available ultrasound scanner ().

***C.1.g. Statistical analysis***. Temperature (C.1.b) represents the explanatory variable that will be used to statistically model the properties (C.1.c-f). The analysis will account for potential nonlinearities in the relationships among the variables, as well as the dependence induced by using the same tissue samples across temperatures. Further, the model will test for and accommodate hysteresis effects. The correlations among the property variables will be investigated using Pearson and Spearman correlations.

***C.1.h. Ensuring rigor and reproducibility***. Sample size required?

***C.1.i. Measures of success***. A Bayesian statistical modeling that allows for the computation of probabilities associated with hypotheses will be used. Success in characterizing the relationships among property measurements and the temperature we be declared if the probability of no relationship is less than 0.05.

***C.1.j. Potential problems and alternatives***. Acquiring human tissues. Ex vivo vs in vivo: could use in vivo treatment data to compute diffusivity, absorptivity, and pennes perfusion (Dillon 2018, clinical property estimation). Hardware temperature limits?

**C.2 Specific Aim 2**

As described in A.4, acoustic and thermal computational models of MRgFUS are not used extensively in current treatment planning protocols. Treatment planning software only crudely accounts for tumor tissue heterogeneity caused by variation in fat, cellularity, fibrous components, and vascularity and no temperature-dependence is considered at all. Computational models are underutilized in the clinic due in part to a lack of model validation. This aim hypothesizes that implementing temperature-dependent properties in treatment modeling will provide more accurate predictions of the temperature rise location, magnitude, and distribution in MRgFUS treatments than using literature-derived constant properties. These efforts at model improvement and validation will increase confidence in predictive computational models proposed for MRgFUS treatment planning.

***C.2.a. Clinical data for analysis***. Desmoid tumor MRgFUS treatments performed at Stanford University from 2016 to 2022 (N=?) will be retrospectively evaluated in this aim. Any exclusion criteria? MR data included for the analysis will include anatomic imaging, fat/water separation images, MR temperatures acquired during focused ultrasound sonications, and post-treatment contrast enhanced images. Additional information including the time stamp of sonications, sonication durations, transducer positioning (including element positioning), and phases of all elements applied for electronic steering will also be required. All data will be deidentified to protect private health information.

***C.2.b. Tissue segmentation***. Volumetric MRI scans and fat/water separation images will be interpolated to 0.5-isotropic spacing (ZFI ref). Initial segmentation will be performed on the fat- and water-separated images using Seg3D software (REF). Manual cleanup of the segmented images will likely be necessary due to signal inhomogeneity from nonuniform distribution of radiofrequency coils. Further manual and semi-automatic segmentation will result in a model including skin, fat, muscle, tumor, and bone.

***C.2.c. Acoustic modeling***. Acoustic simulations will utilize the hybrid angular spectrum (HAS) algorithm (REF). The transducer position with respect to the patient will be determined from anatomic images. Element positions, power, and phasing will be taken from the treatment log files. Constant property simulations will utilize acoustic properties of the various tissue types taken from the literature (REF). Temperature-dependent property simulations will implement the fat speed of sound and acoustic attenuation values described in C.1.d. Absorption will be assumed equivalent to acoustic attenuation, neglecting scattering in the tissue.

***C.2.d. Thermal modeling***. A finite-difference time-domain thermal solver (0.1-s temporal resolution) of the Pennes bioheat equation (REF) will compute temperature distributions from the MRgFUS sonications. The initial conditions for each sonication will be drawn from MR baseline temperature reconstructions of the treatment data as described in C.2.e. Simulations will be performed using (a) constant thermal properties from the literature with constant acoustic property power deposition from C.2.d, and (b) temperature-dependent thermal properties from C.1.e with temperature-dependent acoustic property power deposition from C.2.d. Pennes perfusion parameter will be taken from the literature and assumed constant for each tissue type for all thermal simulations. Both heating and cooling data will be simulated to correspond with MR temperature data acquired during the treatment.

***C.2.e. MR temperature reconstruction***. The background tissue temperature will be computed using a prior baseline method from non-heating images acquired incrementally throughout the treatment (Bitton REF). Temperature changes from ultrasound sonication will be reconstructed using referenceless (?) PRF thermometry methods (REF). All temperature data will be zero-filled interpolated to 0.5-mm in-plane resolution (REF). Final temperatures will be computed by summing the background baseline temperatures with local temperature changes during sonication.

***C.2.f. Computational-clinical comparison***. For each sonication of the MRgFUS clinical data and computational data (both constant and temperature-dependent properties), three metrics of the temperature data will be assessed: focal location, focal distribution, and focal intensity. The focal location will be determined by computing the center of thermal mass for all voxels with temperature changes exceeding 4 C (Dillon 2018, Clinical property estimation). The focal distribution is characterized by the total contiguous volume in which temperature changes exceed 50% of the maximum temperature rise. Finally, focal intensity is defined as the peak temperature change at the end of the heating period.

***C.2.g. Statistics***. *Essentially, we have two models (constant property and temperature-dependent properties) and would like to statistically evaluate if one is better at predicting the three metrics computed from the clinical data. What kind of test should we use? How many datasets will be required? We’ll likely have hundreds of sonications.*

There are two competing physical models to predict the three metrics: a constant property model and a temperature-dependent property model. Each of these physical models will be characterized as a statistical model. It is hypothesized that the statistical model for the temperature-dependent property model will lead to better predictions.

***C.2.h. Ensuring rigor and reproducibility***. Hundreds of sonications will likely be available, thereby permitting ample opportunity to test for an improvement in predictive ability of the temperature-dependent property model and to assess the magnitude of the improvement.

***C.2.i. Measures of success***. For each model, the deviance information criterion (DIC) and root mean squared prediction error (from *k*-fold cross validation) will be computed. These calculations will allow for the assessment of statistical significance and for the estimation of the magnitude of the improvement.

***C.2.j. Treatment planning pipeline***. A forward-looking portion of this Aim is to begin the framework for treatment planning computational tools that could be implemented in the clinic to assist in (a) treatment optimization and (b) reducing phase aberration during treatments. A small number of clinical MRgFUS sonications (N=?) where significant reduction in sonication efficiency was observed will be retrospectively investigated to see if a change in the acoustic path or utilization of phase aberration correction tools (Dillon 2018, Experimental assessment…, other REFs?) might have resulted in more efficient delivery of the ultrasound energy. This retrospective analysis will make use of the temperature-dependent property changes characterized in Aim 1. Effort will also be put into improving the computational efficiency of such tools through parallelization, high performance computing resources, code optimization, reduced order methods, and/or machine learning, so that these analyses can be performed in clinically relevant time frames.

***C.2.k. Potential problems and alternatives***. Segmentation challenges? Neglecting scattering? Challenge of updating acoustic field for changing temperatures at the focus (Guntur 2015 ref?). Prior baseline method should give reasonably accurate values for near field heating, except in fat which is what we care about. Extrapolate into the fat? Pennes perfusion being held constant throughout the treatment.

**D. Summary**

The expected outcome of this high risk-high impact Trailblazer proposal…This is a first step toward our long-term goal of…. Doing this in the way we’ve proposed is awesome because…