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SNR Relaxivity and Stability of Agarose-based MRI Phantoms with Iron (III) Chloride and Gadolinium (III) Chloride as Relaxation Modifier: A Longitudinal Study

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ABSTRACT

Magnetic resonance imaging (MRI) quality control (QC) relies heavily on phantom relaxation to maintain image consistency. Current research indicates that many laboratory-fabricated phantoms often fall short of image uniformity and stability over time. This study aimed to assess the signal-to-noise ratio (SNR) relaxivity and stability of agarose gel phantoms added with relaxation modifiers against time. A total of twenty-two agarose gel phantoms added with different volumes of gadolinium (III) chloride ($GdCl_3$) and iron (III) chloride ($FeCl_3$) were prepared. The phantoms were scanned using a 3-T Siemens Magnetom Skyra MRI system at an interval of 30 days apart. A Turbo Spin Echo (TSE) pulse sequence was used in acquiring T1 and T2 measurement images. The SNR of all the T1 and T2 images acquired were calculated, analyzed, and compared. The $GdCl_3$ and $FeCl_3$ added agarose gel phantoms showed a decreasing T1 SNR value when the relaxation modifier content increased and a gradually decreasing T1 SNR value in the range of TR used. The prepared phantoms displayed an unstable T1 SNR over an interval of time after preparation. The T2 curves for all phantoms appear the same at all time points (TP) indicating the very small influence of TP on T2 relaxation. While the T1

values for the phantoms were undetermined in this study, their T2 values show an inconsistent trend with increasing relaxation modifier content and at different TP. The use of $FeCl_3$ and $GdCl_3$ as relaxation modifiers seemed promising especially for the quality control of a fast imaging sequence due to their ability to reduce T1 relaxation time markedly. In addition, the consistency of T2 relaxation over time indicates the T2 SNR stability of the phantoms.

Keywords: Relaxivity; SNR; longitudinal; agarose; gel; modifier

INTRODUCTION

MRI phantom is a scientific device that is specially designed to mimic the photon properties of human tissue. It is used to evaluate, analyze, and fine-tune the performance of an MRI scanner during the quality control (QC) procedure of an MRI system. A standard water phantom, as provided by the American College of Radiology (ACR) accredited phantom, has all the characteristics that an MRI phantom has to offer. However, this phantom is not available in all MRI facilities due to its prohibitive cost [1].

As an alternative to the common water phantom, various phantoms that mimicked human tissue have been designed and assessed in recent decades [2]. Gel phantoms are more practical and cost-effective for medicine and biomedical research for quality assessment purposes and their lifespan can be easily extended by adding preservatives [3]. The behaviors of phantoms made of agarose gel when compared to the characteristics of soft tissues were found to be similar in many ways compared to other phantoms [4]. The significance of this product is due to its low viscosity, low melting point, and low gelling agent [1].

There are several essential criteria of an MRI phantom which are relaxation times in the range of soft tissues, constant T1 and T2 relaxation times all over the phantom itself, ability to be formed into similar shape and size of human organs, ease to handle and chemically and physically stable over a prolonged period [1]. To fulfill such requirements, a longitudinal study should be done to assess the stability of the phantom. In our previous study, iron (III) oxide (Fe_2O_3) nanoparticles as relaxation modifiers were diluted into a hydrochloric (HCl) solution and mixed with an agarose gel phantom that resulted in a larger surface area [5]. It will increase the chemical reactivity that will enable efficient magnetic relaxivity and allow higher sensitivity of detecting any pathology in MRI [6]. The Fe_2O_3 nanoparticles are specified as inorganic nanoparticle-based contrast agents and have received significant recognition for their unique magnetic properties at the nanoscale [6]. The relaxation modifiers are expected to reduce both T1 and T2 relaxation times by dipole-dipole interaction between the magnetic moments of hydrogen protons and the magnetic moment of particles [5]. Longitudinal T1 relaxation times in human soft tissue range from about 380 to 1700 ms and transversal T2 relaxation on the other hand is measured to range from 30 to 140 ms at 3 T. The agarose gel doped with relaxation modifier would be able to mimic the human's T1/T2 such as grey matter (1607ms/63 ms), white matter (838 ms/77 ms), and renal cortex (1261 ms/121 ms) [7].

The cylindrical ACR magnetic resonance accreditation phantom is filled with water and has relaxation durations and conductivities that are relevant to biological tissues. However, water phantoms can also be affected by vibration and cause air bubbles that may lead to non-uniformity. The relaxation time of water, which is significantly longer than that of human tissue, is another challenge when utilizing a water phantom. Consequently, research into alternative materials with relaxation times that are more

similar to those of human tissue is growing [5]. Agarose gel is the most used material to fabricate the MRI phantom besides polyvinyl alcohol (PVA), carrageenan, and distilled water [8]. In a previous study, the behaviors of phantoms made of agarose gel are similar in many ways compared to other phantoms when compared to the characteristics of soft tissues [4]. The addition of ionic solutions as the relaxation modifiers to alter the T1 and T2 relaxation times may added to the choice of agarose gel as an MRI phantom in a clinical setting.

Our previous study on gadolinium (III) chloride (Gd_2Cl_3) and iron (III) chloride (Fe_2Cl_3) solutions as a relaxation modifier for agarose gel phantom exhibited excellent T1 and T2 SNR uniformity [9]. In another study, agar gel phantoms exhibited SNR uniformity but showed instability of the SNR after four weeks of preparation [5]. This occurs due to the evaporation process inside the phantom resulting in the loss of water molecules, which significantly impacts the phantom's T1 and T2 relaxation times and SNR [5]. Thus, the prepared phantoms need to be scanned at different time points (TP) to evaluate the condition of the phantom over time. In this study, 22 agarose gel phantoms added with Gd_2Cl_3 and Fe_2Cl_3 as relaxation modifiers were scanned at different TP (TP1, TP2, TP3, TP4, TP5). The T1 and T2 weighted images were analyzed to evaluate the SNR relaxivity and uniformity of the agarose gel phantom.

EXPERIMENTAL METHODS

Materials and Apparatus

This project was approved by the Internal Review Board (UKM JEP-2021-616). The materials and apparatus used in the preparation of agar gel phantom were as follows: 1) 2.55 g of Agarose Powder (Sigma Aldrich, Malaysia); 2) 0.1 g of each gadolinium (III) oxide (Gd_2O_3) and Fe_2O_3 (Sigma Aldrich, Malaysia) nanoparticles (100 nm) as the relaxation modifier; 3) 0.01 g of copper oxide (CuO) (Sigma Aldrich, Malaysia) nanoparticle (100 nm) as the antifungal; 4) 10 ml of hydrochloric acid (HCl) 37% (Fisher Scientific (M) Sdn. Bhd.); 5) 80 ml of distilled water; 6) digital weighing scale (Ohaus); 7) pipet; 8) thermometer; 9) Glass rod; 10) sterile plastic container; 11) magnetic stirrer with hotplate; 12) beaker; 13) cylindrical measuring tube; 14) grease and 15) seal tape for sealing the container.

Location of Research

The agarose gel phantoms were prepared in the Biomedical Laboratory, Center for Diagnostic, Therapeutic and Investigative Studies (CODTIS) Faculty of Health Science (FHS), Universiti Kebangsaan Malaysia (UKM) Kuala Lumpur. The prepared phantoms were scanned using a 3T Siemens Magnetom Skyra MRI system in Hospital Pakar Kanak-Kanak (HPKK), Universiti Kebangsaan Malaysia (UKM) at 5 TP 30 days apart. This interval is the chosen period to assume the characteristics of the phantoms were reliable to evaluate the SNR whereas in the previous study, the water content in the agar gel phantoms was evaporated after 2 weeks of preparation [5]. Thus, it was thought that by having 5 TP with 30 days interval, the SNR can still be measured and analyzed despite the loss of water vapor from the phantoms.

Preparation of the Agarose Gel Phantom

Twenty-two agarose gel phantoms were prepared using conventional mixing methods. The agarose powder (2.55 g) was mixed with distilled water (80 ml) at room temperature by using a magnetic stirring technique while heated at 80°C on a hot plate for 8 minutes

to homogenize the solution. The color changes of the solution from cloudy to crystal clear indicate that the solution is ready to be used. This solution was labeled as Solution A.

An amount of 0.1 g Gd_2O_3 and Fe_2O_3 nano-powders were mixed with 10 ml HCL in two different beakers separately. The magnetic stirring technique was applied again to ensure the uniformity of the relaxation modifier solution for 2 minutes on a hot plate at 80°C [10]. A reaction between an inorganic compound such as Gd_2O_3 or Fe_2O_3 with an inorganic acid such as HCL has produced a colorless aqueous solution of $GdCl_3$ and water (H_2O) as stated in Eqn. (1) or a pale yellow-green aqueous solution of $FeCl_3$ and H_2O as stated in Eqn. (2) through double displacement reaction.

$$Gd_2O_3 + 6HCl = 2GdCl_3 + 3H_2O \quad (1)$$
$$Fe_2O_3 + 6HCl = 2FeCl_3 + 3H_2O \quad (2)$$

These two solutions were labeled as Solution B. Solution A and Solution B were mixed in sterilized plastic containers following a systematic volume fraction as shown in Table 1. The total volume of the mixture was 80 ml. The plastic containers were sealed with grease and PTFE seal tape to prevent leakage and let to cool down to room temperature. The chemical reaction may be visualized as the presence of bubbles or a change in the color of the solution which is not shown in this study. Leakage prevention is very crucial in this study because water leakage will affect the SNR value in such a way that a higher SNR value indicates higher water content [5]. All the phantoms were kept in a desiccator before the MRI scan and inspected visually for any leakage, presence of air bubbles, and fungal growths.

Table 1. The systematic volume fraction of the agarose gel phantoms

Phantom	Agarose Gel Phantom with Gd_2O_3			Agarose Gel Phantom with Fe_2O_3		
	Volume of Solution A (Distilled water + Agarose Gel Powder)/ml	Volume of Solution B (HCl + Gd_2O_3)/ml	Total Volume /ml	Volume of Solution A (Distilled water + Agarose Gel Powder)/ml	Volume of Solution B (HCl + Fe_2O_3)/ml	Total Volume /ml
1	80	0	80	80	0	80
2	79	1	80	79	1	80
3	78	2	80	78	2	80
4	77	3	80	77	3	80
5	76	4	80	76	4	80
6	75	5	80	75	5	80
7	74	6	80	74	6	80
8	73	7	80	73	7	80
9	72	8	80	72	8	80
10	71	9	80	71	9	80
11	70	10	80	70	10	80

Data Acquisition

The dates chosen for phantom scanning were: 1) 12th November 2021 (TP1); 2) 9th December 2021 (TP2); 3) 6th January 2022 (TP3); 4) 3rd February 2022; 5) 3rd March 2022 (TP5). The agarose gel phantoms were positioned horizontally inside a head coil at

the iso-centre of the magnet bore. This was to ensure that all the phantoms were subjected to the same magnitude of the magnetic field during the MRI scan. The turbo spin echo (TSE) sequence was applied to obtain T1 and T2 measurement images in axial projection for all phantoms. To produce T1 measurement images, a short echo time (TE) = 18 ms was used with the repetition time (TR) changed in a range of 400 ms to 6000 ms. For T2 measurement images, a long TR = 2000 ms was used with TE changed in a range of 26 ms to 235 ms [11]. All other imaging parameters were kept constant such as the field of view (FOV) of 200 mm 200 mm, slice thickness of 5.0 mm, and matrix size of 256. All images acquired were stored in a CD-ROM in DICOM format.

Data Analysis

This analysis is conducted based on the data scanned on five different TP. The SNR of the images is measured using The Image J software (The National Institute of Health, NIH) similar to previous studies [12]. The SNR is determined by placing the single large region of interest (ROI) that fits the size of the phantom image on all phantom images to obtain the signal intensity (I_p). In the previous study, the SNR of the phantom on different ROIs shows a similar trend of T1 and T2 relaxation in 1, 3, and 25 ROIs [5]. The size of the ROI is drawn as large as possible until it covers the whole phantom image. Background intensity (I_b) is measured by placing a large ROI on the background of the image. The I_b obtained from each phantom image was averaged to acquire the mean I_b and its standard deviation (σ_b). The SNR of the T1 and T2 measurement images is determined using Eqn. (3).

$$SNR = (I_p - I_b)/\sigma_b \tag{3}$$

The SNR obtained from the ROIs drawn on the T1 and T2 measurement images will be compared using the analysis of variance (ANOVA) test in The Jamovi Project (Jamovi). This test will be conducted to determine the stability of the T1, and T2 SNR obtained from the phantoms at 5 different TP.

To determine the T1 and T2 values of all the phantoms under study, the experimental SNR data will be fitted to the exponential equations shown below for T1 (Eqn. 4) and T2 (Eqn. 5) curves, respectively. Matlab Curve Fitting Toolbox will be used for this purpose.

$$SNR(TR) = SNR_o\,(1 - e^{-TR/T1}) \tag{4}$$
$$SNR(TE) = SNR_i e^{-TE/T2} \tag{5}$$

For T1 curves that do not show a relaxation process, T1 determination will not be conducted. For T2 curves that do not fit Eqn. (5), a transformation of Eqn. (5) into a natural logarithm will be done to derive an equation shown by Eqn. (6) below:

$$\ln SNR(TE) = -(TE/T2) + \ln SNR_i \tag{6}$$

and line fitting will be performed using EXCEL programming software.

RESULTS

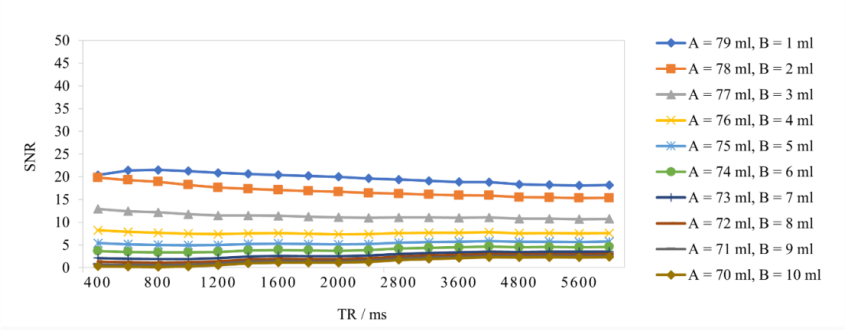
T1 and T2 curves for agarose gel added with $FeCl_3$ in different proportions for TP1 to TP5

Figures 1 and 2 show the SNR as a function TR (T1 curve) and TE (T2 curve) for agarose gel phantoms added with $FeCl_3$ obtained from an ROI drawn on each phantom, respectively. The T1 curves (Figure 1) show a nearly constant value of SNR for all TR values resembling the saturation part of a T1 curve. The relaxation part of the T1 curves is absent in the present range of TR used in this study. For each TP, the T1 SNR decreases markedly with increasing $FeCl_3$ content but interestingly, the SNR increases with time after preparation for all phantoms. This large influence of TP on T1 SNR means that the T1 SNR is quite unstable over a time interval after preparation. The T2 curves for all agarose gel phantoms demonstrate an exponentially decreasing trend as the TE value increases (Figure 2). However, the curves start to show a constant trend starting from TE = 91 ms. The T2 SNR curves for all phantoms look the same at all TP indicating a very little influence of TP on T2 relaxation.

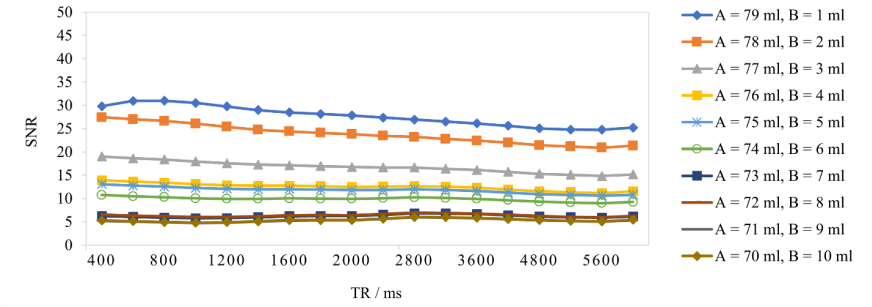
T1 and T2 curves for agarose gel added with $GdCl_3$ in different proportions for TP1 to TP5

Figs. 3 and 4 show the SNR as a function of TR (T1 curve) and TE (T2 curve) for agarose gel phantoms added with $GdCl_3$ obtained from an ROI drawn on each phantom, respectively. The T1 curves (Figure 3) show a nearly constant value of SNR for all TR values resembling the saturation part of a T1 curve. The relaxation part of the T1 curve is also absent in the present range of TR used for this study. For each TP, the T1 SNR decreases markedly with increasing $GdCl_3$ content. Unlike phantoms added with $FeCl_3$, the phantoms added with $GdCl_3$ show a noticeable influence of TP on T1 SNR values which indicates a fluctuation in SNR values for all phantoms over time. This influence of TP on T1 SNR means that the T1 SNR is quite unstable over a time interval after preparation. Similar to phantoms added with $FeCl_3$, the T2 curve for all agar gel phantoms demonstrates an exponentially decreasing trend as the TE value increases. However, it starts to show a constant trend starting from TE = 91 ms (Figure 4). Again, the T2 curves for all phantoms look the same at all TP indicating very little influence of TP on T2 relaxation.

(a)



(b)



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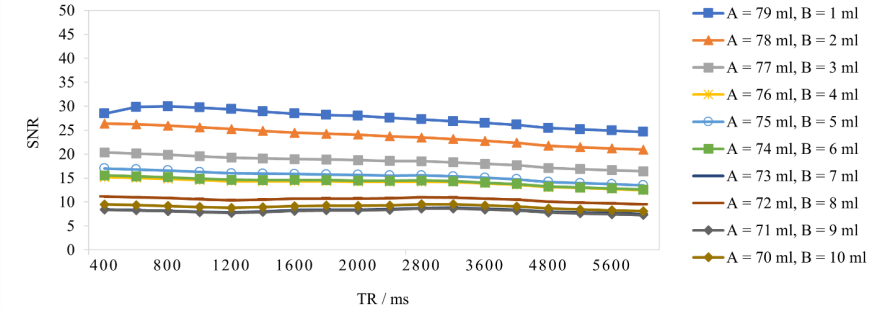

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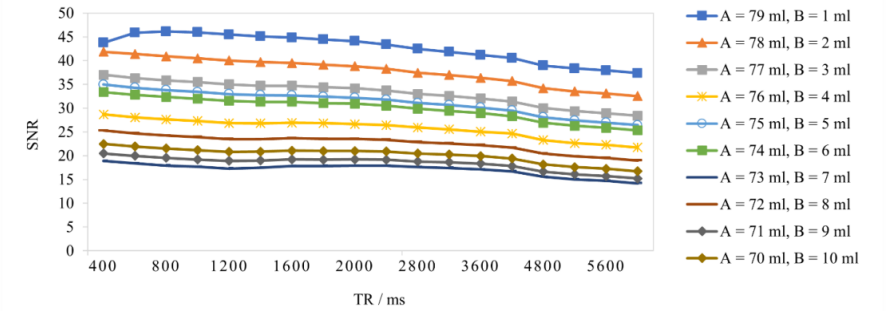

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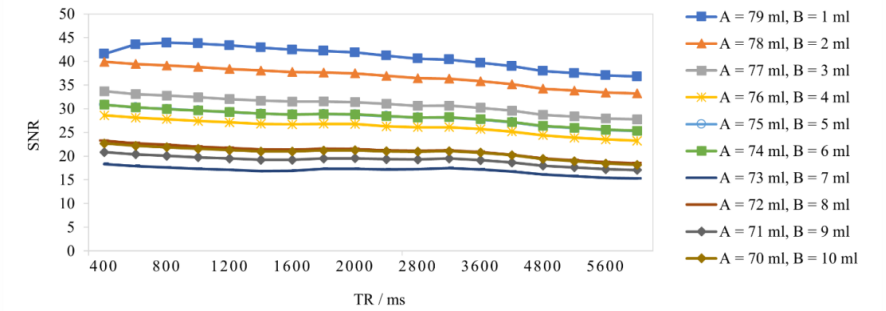
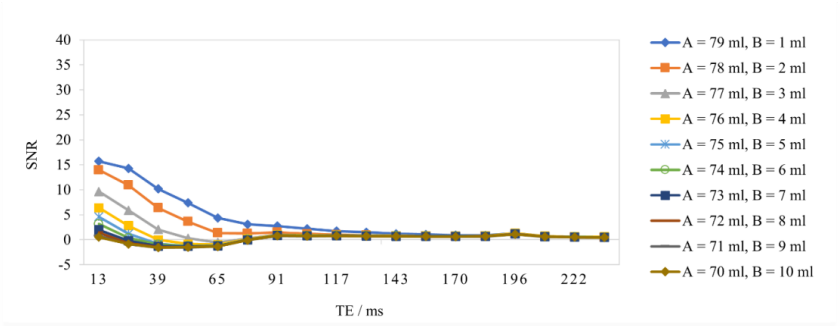


Figure 1. T1 plots obtained from agarose gel added with $FeCl_3$ solution at different TP (a) TP 1 (b) TP 2 (c) TP 3 (d) TP 4 and (e) TP 5

(a)



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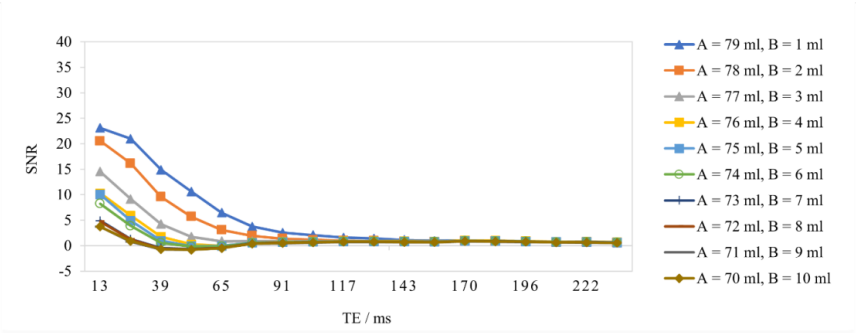

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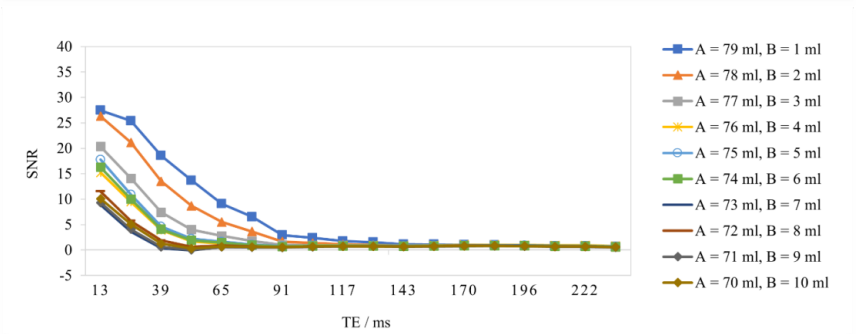

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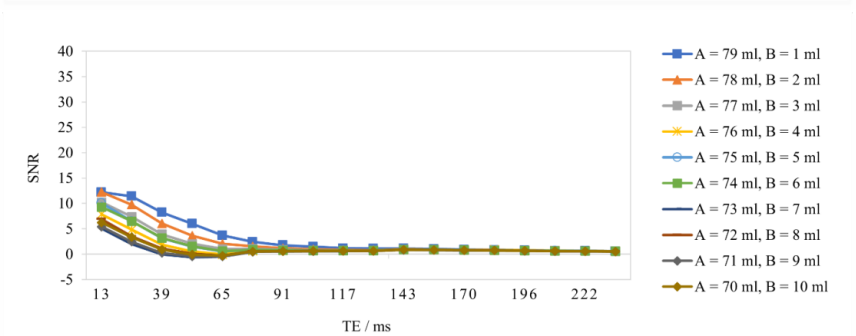

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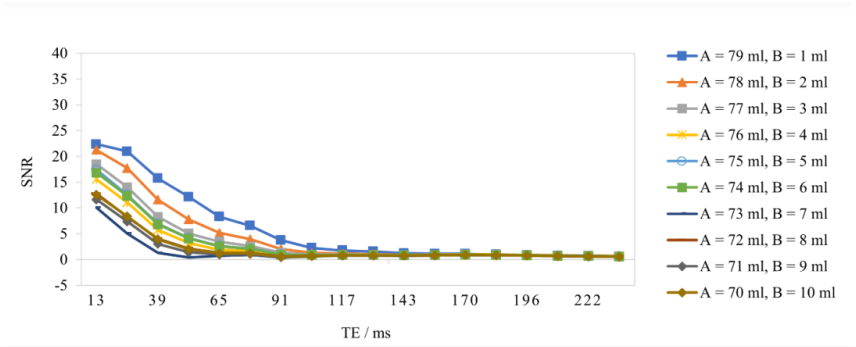


Figure 2. T2 plots obtained from agarose gel added with $FeCl_3$ solution at different TP (a) TP 1 (b) TP 2 (c) TP 3 (d) TP 4 and (e) TP 5

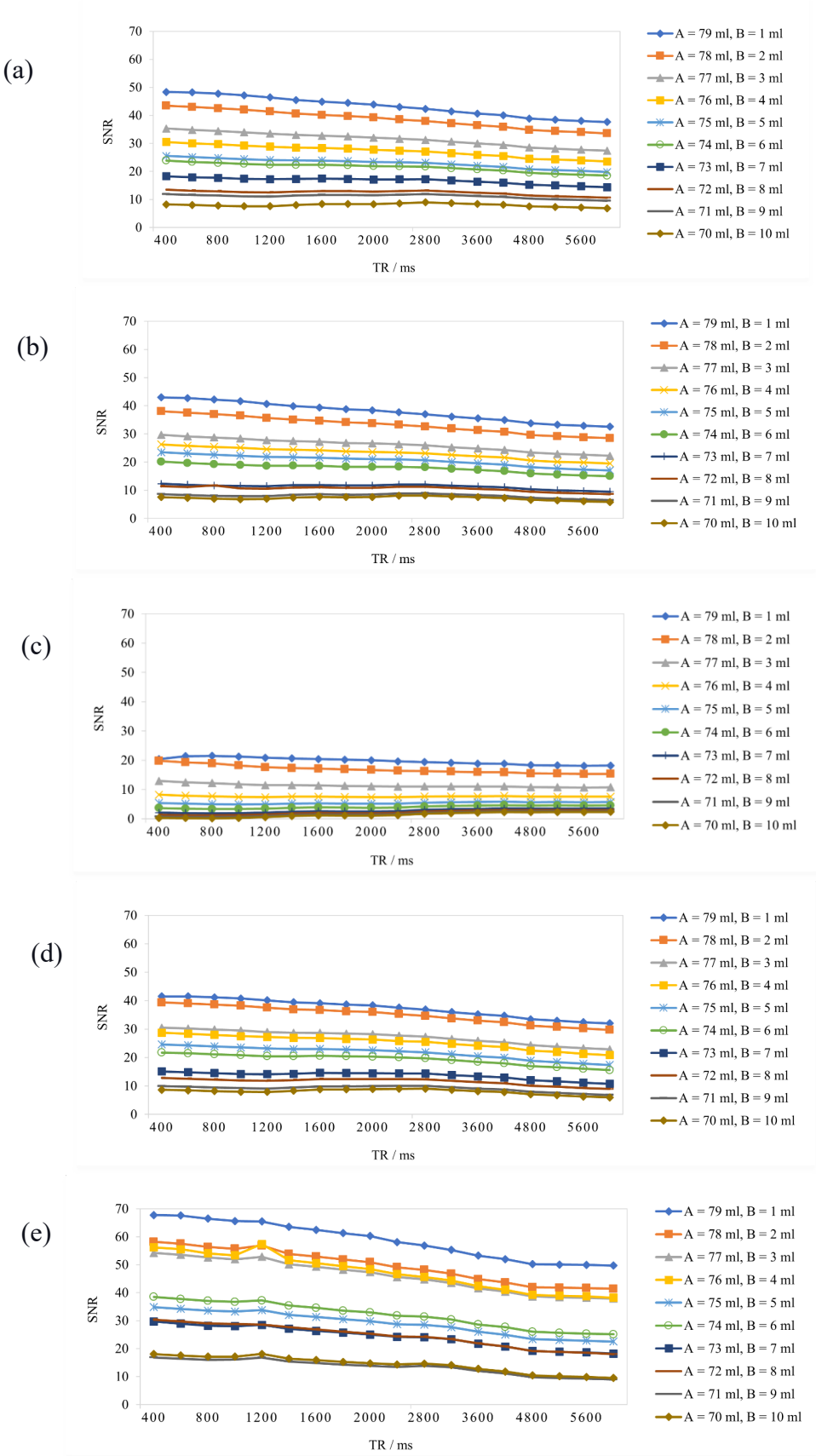
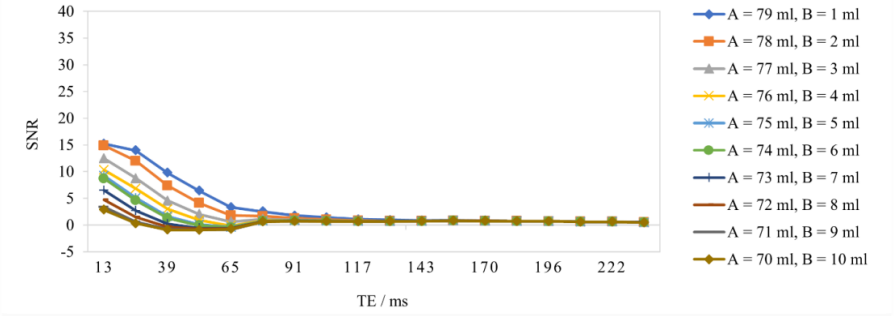


Figure 3. T1 plots obtained from agarose gel added with Gd_2O_3 solution at 5 different TP (a) TP 1 (b) TP 2 (c) TP 3 (d) TP 4 and (e) TP 5

(a)



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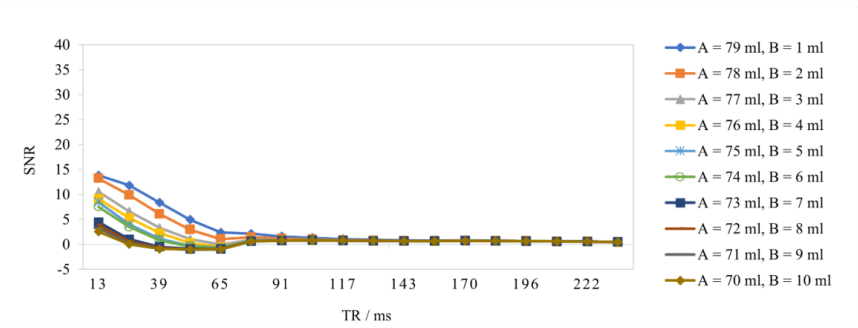

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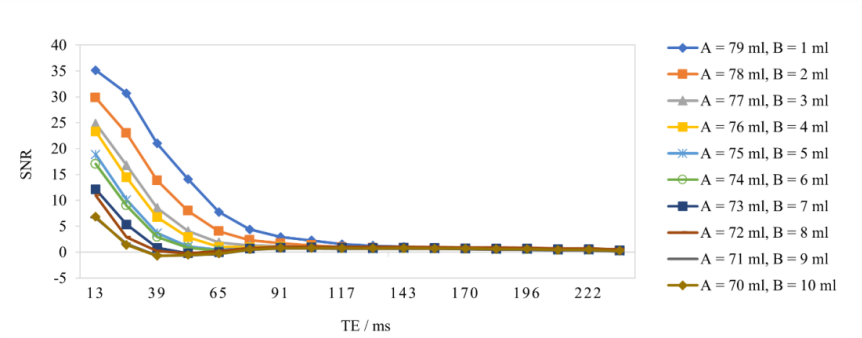

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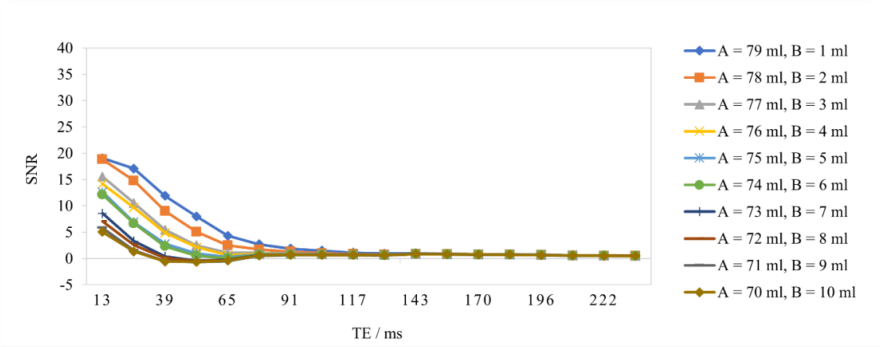

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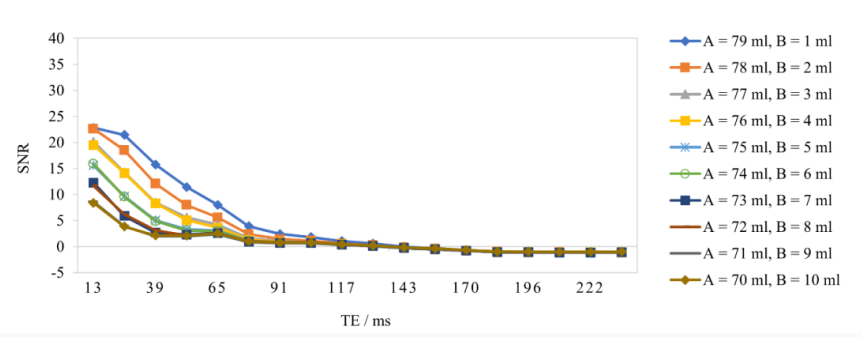


Figure 4. T2 plots obtained from agarose gel added with Gd_2O_3 solution at 5 different TP (a) TP 1 (b) TP 2 (c) TP 3 (d) TP 4 and (e) TP 5

Stability of SNR across 5 TP

SNR stability over time can be confirmed by comparing the SNR values between TP for each of the phantoms. The statistical analysis was conducted to evaluate the hypothesis of no difference in SNR with time after preparation for all phantoms. The results of ANOVA on T1 curves for all phantoms with $FeCl_3$ addition show a significant difference ($p < 0.05$) for all comparisons except several comparisons involving pure agarose and comparisons involving TP4 vs. TP5 (Table 2). For phantoms with $GdCl_3$ addition, the results also show a significant difference ($p < 0.05$) of SNR for all comparisons except several comparisons involving pure agarose and comparisons involving TP1 vs. TP2 and TP1 vs. TP4 (Table 3). Insignificant differences are given as bold-faced figures in both tables. However, the results of ANOVA for T2 curves for all phantoms with $FeCl_3$, and $GdCl_3$ additions show insignificant differences for all comparisons (Tables 4 and 5). It can be summarized that the majority of the phantoms added with relaxation modifiers show significant differences in T1 SNR over time while the difference in T2 SNR is not comparable. The significant difference in SNR between TP indicates an instability (or time-dependent) of the SNR of the agar phantoms produced in this study.

Table 2. One-Way ANOVA (Fisher's) for T1 curve of agar gel phantoms added with $FeCl_3$

$FeCl_3$ volume	TP1 vs TP2	TP1 vs TP3	TP1 vs TP4	TP1 vs TP5	TP2 vs TP3	TP2 vs TP4	TP2 vs TP5	TP3 vs TP4	TP3 vs TP5	TP4 vs TP5
0	**0.163**	**0.215**	<0.001	<0.001	**1.000**	0.003	**0.057**	0.002	0.039	**0.847**
1	<0.001	<0.001	<0.001	<0.001	1.000	<0.001	<0.001	<0.001	<0.001	**0.112**
2	<0.001	<0.001	<0.001	<0.001	1.000	<0.001	<0.001	<0.001	<0.001	**0.624**
3	<0.001	<0.001	<0.001	<0.001	0.017	<0.001	<0.001	<0.001	<0.001	<0.001
4	<0.001	<0.001	<0.001	<0.001	0.004	<0.001	<0.001	<0.001	<0.001	**0.868**
5	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
6	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.006
7	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	**0.987**
8	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
9	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	**0.144**
10	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	**0.441**

Bold-face figures = not significant ($p>0.05$)

Table 3. One-Way ANOVA (Fisher's) for T1 curve of agar gel phantoms added with $GdCl_3$

$GdCl_3$ volume	TP1 vs TP2	TP1 vs TP3	TP1 vs TP4	TP1 vs TP5	TP2 vs TP3	TP2 vs TP4	TP2 vs TP5	TP3 vs TP4	TP3 vs TP5	TP4 vs TP5
0	**0.901**	0.007	**0.869**	**0.089**	**0.080**	**1.000**	0.008	**0.097**	<0.001	0.006
1	0.002	<0.001	0.001	0.001	0.001	0.996	0.001	0.001	0.001	0.001
2	0.001	0.001	0.037	0.001	0.001	0.634	0.001	0.001	0.001	0.001
3	0.001	0.001	0.002	0.001	0.001	0.838	0.001	0.001	0.001	0.001
4	0.006	0.001	**0.522**	0.001	0.001	0.293	0.001	0.001	0.001	0.001
5	0.044	0.001	**0.532**	0.001	0.001	0.699	0.001	0.001	0.001	0.001
6	0.001	0.001	**0.092**	0.001	0.001	0.369	0.001	0.001	0.001	0.001
7	0.001	0.001	0.001	0.001	0.001	0.006	0.001	0.001	0.001	0.001
8	**0.054**	0.001	**0.697**	0.001	0.001	0.588	0.001	0.001	0.001	0.001
9	0.001	0.001	0.001	0.001	0.001	0.201	0.001	0.001	0.001	0.001
10	**0.492**	0.001	**1.000**	0.001	0.001	0.430	0.001	0.001	0.001	0.001

Bold-face figures = not significant (p>0.05)

Table 4. One-Way ANOVA (Fisher's) for T2 curve for agar phantoms added with $FeCl_3$

$FeCl_3$ volume	TP1 vs TP2	TP1 vs TP3	TP1 vs TP4	TP1 vs TP5	TP2 vs TP3	TP2 vs TP4	TP2 vs TP5	TP3 vs TP4	TP3 vs TP5	TP4 vs TP5
0	**0.801**	**0.436**	**0.98**	**0.79**	**0.975**	**0.454**	**1.000**	**0.164**	**0.978**	**0.442**
1	**0.975**	**0.771**	**0.996**	**0.925**	**0.980**	**0.871**	**1.000**	**0.549**	**0.997**	**0.765**
2	**0.976**	**0.713**	**1.000**	**0.88**	**0.962**	**0.962**	**0.997**	**0.664**	**0.998**	**0.845**
3	**0.976**	**0.632**	**0.998**	**0.583**	**0.928**	**0.999**	**0.903**	**0.818**	**1.000**	**0.779**
4	**0.964**	**0.59**	**0.993**	**0.367**	**0.929**	**0.999**	**0.772**	**0.835**	**0.996**	**0.627**
5	**0.953**	**0.361**	**0.854**	**0.183**	**0.795**	**0.998**	**0.561**	**0.92**	**0.995**	**0.742**
6	**0.956**	**0.331**	**0.770**	**0.112**	**0.758**	**0.99**	**0.409**	**0.951**	**0.98**	**0.701**
7	**0.946**	**0.465**	**0.926**	**0.237**	**0.89**	**1.000**	**0.667**	**0.915**	**0.993**	**0.71**
8	**0.968**	**0.289**	**0.748**	**0.071**	**0.674**	**0.98**	**0.273**	**0.94**	**0.961**	**0.604**
9	**0.959**	**0.367**	**0.792**	**0.05**	**0.785**	**0.992**	**0.229**	**0.956**	**0.871**	**0.464**
10	**0.965**	**0.318**	**0.708**	**0.039**	**0.717**	**0.972**	**0.181**	**0.968**	**0.869**	**0.497**

Bold-face figures = not significant (p>0.05)

Table 5. One-Way ANOVA (Fisher's) for T2 curve for agar phantoms added with $GdCl_3$

$GdCl_3$ volume	TP1 vs TP2	TP1 vs TP3	TP1 vs TP4	TP1 vs TP5	TP2 vs TP3	TP2 vs TP4	TP2 vs TP5	TP3 vs TP4	TP3 vs TP5	TP4 vs TP5
0	**1.000**	**0.214**	**0.999**	**0.958**	**0.155**	**0.994**	**0.912**	**0.333**	**0.601**	**0.992**
1	**1.000**	**0.568**	**0.999**	**0.989**	**0.447**	**0.989**	**0.961**	**0.743**	**0.849**	**1.000**
2	**1.000**	**0.821**	**0.999**	**0.995**	**0.707**	**0.989**	**0.974**	**0.931**	**0.962**	**1.000**
3	**0.999**	**0.871**	**0.999**	**0.995**	**0.741**	**0.989**	**0.966**	**0.947**	**0.98**	**1.000**
4	**1.000**	**0.816**	**0.994**	**0.966**	**0.72**	**0.978**	**0.922**	**0.961**	**0.993**	**0.999**
5	**1.000**	**0.922**	**0.995**	**0.995**	**0.869**	**0.983**	**0.984**	**0.993**	**0.992**	**1.000**
6	**1.000**	**0.939**	**0.996**	**0.99**	**0.879**	**0.982**	**0.968**	**0.995**	**0.998**	**1.000**
7	**0.996**	**0.942**	**0.999**	**0.998**	**0.798**	**0.973**	**0.964**	**0.986**	**0.991**	**1.000**
8	**1.000**	**0.853**	**0.996**	**0.949**	**0.742**	**0.978**	**0.88**	**0.968**	**0.999**	**0.996**
9	**1.000**	**0.995**	**0.992**	**0.988**	**0.988**	**0.984**	**0.977**	**1.000**	**1.000**	**1.000**
10	**1.000**	**0.964**	**0.99**	**0.934**	**0.941**	**0.98**	**0.902**	**1.000**	**1.000**	**0.998**

Bold-face figures = not significant (p>0.05)

T1 and T2 values for agarose gel added with $GdCl_3$ in different proportions for TP1 to TP5

The experimental SNR data are supposed to be fitted to the exponential equations for T1 (Eqn. 4) and T2 (Eqn. 5) curves respectively to determine the values of T1 and T2 of all phantoms in this study. However, the T1 determination could not be accomplished due to the absence of the relaxation part of T1 curves for agarose gels added with relaxation modifiers as shown in Figs. 1 and 3. For T2 determination, a line fitting technique was performed using EXCEL programming software by using the derived equation as shown by Eqn. (6). The T2 values determined from line fitting are shown in Tables 6 and 7 for agarose gel phantom added with $FeCl_3$ and $GdCl_3$ respectively.

Table 6. T2 values for agarose gel phantom added with $FeCl_3$ relaxation modifier

$FeCl_3$ volume	T2/ms				
	TP1	TP2	TP3	TP4	TP5
0	79.37	65.79	75.76	75.19	112.36
1	39.84	33.44	35.84	37.59	43.67
2	28.74	27.03	28.65	30.67	33.56
3	26.46	24.94	25.64	28.99	30.03
4	62.50	36.50	32.57	30.12	26.60
5	103.09	29.33	24.21	26.88	27.55
6	250.00	51.55	23.92	27.47	27.62
7	322.58	78.13	40.32	59.52	26.95
8	312.50	78.13	25.91	44.25	25.64
9	312.50	126.58	52.08	98.04	24.45
10	333.33	120.48	52.08	71.43	25.32

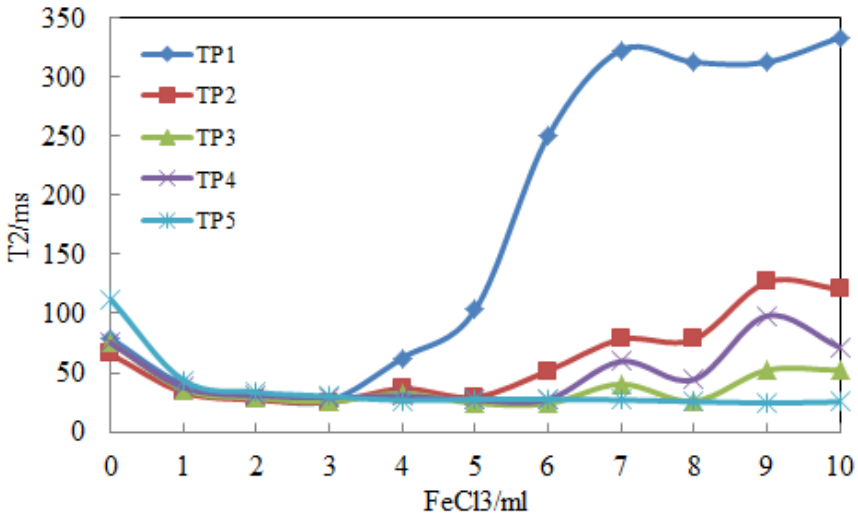
Table 7. T2 values for agarose gel phantom added with (b) $GdCl_3$ relaxation modifier

$GdCl_3$ volume	T2/ms				
	TP1	TP2	TP3	TP4	TP5
0	78.13	76.34	64.52	52.08	53.19
1	33.33	32.57	32.57	31.25	33.67
2	28.57	28.25	25.97	26.67	27.86
3	26.46	36.1	23.04	25.06	26.32
4	34.25	30.49	33.56	23.81	24.75
5	41.84	48.54	33.56	24.63	26.53
6	44.64	38.17	22.17	21.55	26.11
7	70.92	88.5	47.85	46.51	29.85
8	70.42	88.5	81.97	77.52	41.15
9	116.28	243.9	68.97	60.61	34.72
10	217.39	105.26	78.74	68.49	37.31

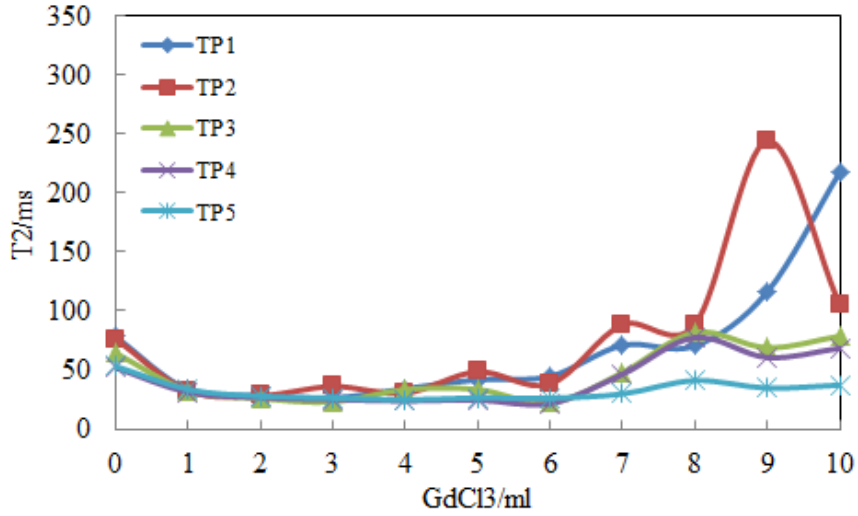
For agarose gel phantoms added with $FeCl_3$, it can be said that for TP1 to TP4, the value of T2 initially decreases (0 ml to 1 ml), remains constant (1 ml to 3 ml), and increases as the volume of $FeCl_3$ added to the agarose phantom increases. However, for TP5, the T2 value shows an initial decrease for $FeCl_3$ volume below 1 ml but remains constant for $FeCl_3$ volume of 1 ml onwards. The comparisons of the T2 values among the 5 TP demonstrate a constant T2 value for the modifier volume of 1 ml to 3 ml. However, for a lower modifier volume (below 1 ml), T2 values are inconsistent but for a higher modifier volume i.e., 4 ml to 10 ml, T2 value decreases with TP.

For agarose gel phantoms added with $GdCl_3$, it can be said that for TP1 to TP4, the value of T2 initially decreases (0 ml to 1 ml), remains nearly constant (1 ml to 6 ml) and increases as the volume of $GdCl_3$ added to the agarose phantom is further increased. For TP5, the T2 value shows an initial decrease for $GdCl_3$ volume below 1 ml but remains constant for $FeCl_3$ volume of 1 ml onwards. The behavior is similar to $FeCl_3$-added agarose gel. The comparisons of the T2 values among the 5 TP demonstrate a constant T2 value for the modifier volume of 1 ml to 6 ml. However, for a lower modifier volume (below 1 ml), T2 values are inconsistent but for a higher modifier volume i.e., 6 ml to 10 ml, T2 value decreases with TP.

The trend of change of T2 values as a function of $FeCl_3$ and $GdCl_3$ content can be seen in Figure 5. It can be said that the stability of the T2 values can only be achieved at lower relaxation modifier concentration and for a longer interval after preparation for $FeCl_3$-added agarose gel phantoms.



(a)



(b)

Figure 5. Plots of T2 values against the modifier volume for the five TP used in this stud: (a) $FeCl_3$ and (b) $GdCl_3$

DISCUSSION

A typical T1 phenomenon should exhibit a time-dependent exponentially increased curve characterized by relaxation, at shorter TR values and saturation, at longer TR values [5]. However, in this study, the agarose gel phantoms added with various volumes of $FeCl_3$ or $GdCl_3$ solutions exhibit only the saturation part. The relaxation part of the T1 curves was absent for all agarose gel phantoms within the range of TR applied during the experiment. The graphs in Figs. 1 and 3 demonstrate that all the phantoms with $FeCl_3$ or $GdCl_3$ solution had reached saturation. These results were mainly caused by the selection of a higher TR range far beyond the range within which the relaxation should be displayed by all phantoms as proven by our study conducted at a lower TR range [9].

The T1 relaxation process corresponds to the transfer of energy from the spins to the lattices/surroundings. When the RF pulse is turned off, this energy transfer takes place and can be explained as the behavior of proton spins seeking equilibrium. As a result, the T1 curve which represents the change in magnetization in the *z* direction, begins to show

relaxation at shorter TR values [1, 9]. If TR is sufficiently long, magnetization achieves saturation, which is represented by a constant T1 SNR. However, the T1 SNR of the phantoms containing $FeCl_3$ or $GdCl_3$ solutions shows a slight decrease with TR which is attributed to the net effect of spin-lattice interaction. As more and more spins reached equilibrium at a longer TR, the T1 SNR gradually decreased. This behavior is less prominent for phantoms containing higher $FeCl_3$ or $GdCl_3$ solutions probably due to the net effects of the presence of higher water molecules and magnetic ions in the agarose gel phantoms.

T1 SNR can be affected by the water content of the phantoms. The addition of $FeCl_3$ or $GdCl_3$ as a relaxation modifier altered the water content of the agarose gel phantoms which in turn affected the T1 SNR in such a way that it decreased as $FeCl_3$ or $GdCl_3$ solutions were added to the phantom. By adding $FeCl_3$ or $GdCl_3$ into the agarose gel, an equal amount of agarose gel is displaced so that the total volume is constant at 80 ml. Pure agarose gel contains fewer water molecules than the ones added with $FeCl_3$ or $GdCl_3$ according to Eqn. (1) and (2). Water molecules are the primary source of T1 SNR and supposedly the T1 SNR increases with the addition of both relaxation modifiers. However, due to the presence of magnetic ions (Fe^{2+} and Gd^{2+}) in the relaxation modifier solutions, the increase in T1 SNR with water content is overcompensated with the increase in signal loss from magnetic field inhomogeneity that was caused by the presence of magnetic ions. Thus, the net effect is the reduction of T1 SNR with the increase in relaxation modifier content.

The T1 SNR for most of the prepared phantoms appears to fluctuate with time after preparation. The 30-day separation between TPs may have caused loss of water content resulting in the inconsistency of the T1 SNR values. Water vapor may have found its way out of the plastic container despite the tight sealing of the container cap. Furthermore, changes in the matrix of the agarose gel phantom due to chemical reactions may also contribute to the inconsistencies. Further investigations need to be conducted to eliminate these causes and to explain why T1 SNR could have been greatly affected by these changes.

A typical T2 relaxation, on the other hand, should exhibit an exponential decrease in T2 SNR as TE increases. The spin-spin interactions that accelerate the decay of the magnetization in the *x-y* plane are the basis of this T2 relaxation phenomenon [1]. The agarose gel phantoms added with various volumes of $FeCl_3$ or $GdCl_3$ solutions used in this study exhibit an exponentially decreasing trend with increasing TE. The curves also show negative T2 SNR for several TE values. According to Eqn. (3), the negative SNR displayed on the T2 curves for all phantoms at several TE values demonstrated that the intensity of the background was higher than the intensity of the T2-weighted images of the phantom. The intensity of the background is quite constant for all images, thus it can be said that the T2 measurement images have lost signal intensity resulting in a drop in SNR possibly due to the spin-spin interaction which causes more signals lost as TE increases [10]. The spin-spin interaction is supposed to last longer for agarose gel phantoms added with $FeCl_3$ or $GdCl_3$, as the surrounding has a higher number of water molecules compared to the pure agarose gel phantom [13, 14] as mentioned earlier. However, the presence of magnetic ions increased the spin-spin interaction and shortened T2 relaxation. For several $FeCl_3$ or $GdCl_3$-added agarose gel phantoms, this mechanism caused the signal intensity to decrease markedly below the intensity of the background at certain TE values.

The difference in T2 SNR curves among all phantoms for every TP can only be observed at lower TE values below 91 ms. For higher TE values, the curves overlapped.

At shorter TE, the spins within the phantoms have yet to reach the state where their precessions are in sync. This is due to the precession frequency of a particular spin that is very much dependent on the surrounding spins, hence spin-spin interaction, resulting in the T2 SNR of the phantoms being comparable. In addition, the T2 SNR at lower TE is still relatively and acceptably high. At higher TE values, the precession of the spins of all phantoms is almost in sync resulting in the overlapping of the T2 curves. Not only has the T2 SNR between phantoms become incomparable, but they are also of low SNR.

As opposed to T1 relaxation, T2 relaxation is not influenced by the time interval after preparation. T2 curves for all phantoms are incomparable among all TPs as confirmed by ANOVA. It can be concluded that spin-spin interaction does not depend on water content or microstructure of the phantoms but rather relies on the interaction between spins which does not change much with time after preparation. T2 relaxation can be said to be sufficiently stable with time after preparation.

The $FeCl_3$ and $GdCl_3$ added agarose gel phantoms under study possess a very short T1 value and can only be observed if TR is lowered below 400 ms [9]. Previous studies found that the presence of relaxation modifiers in the form of magnetic ions was able to shorten T1 relaxation time [15, 16] by 10 to 80 times [9] depending on the modifier content. The T2 relaxation time however was able to be determined from the T2 relaxation curves and shows a large fluctuation at higher relaxation modifier content which could be due to the spins within the phantoms that have yet to reach the state where their precessions are in sync as explained earlier. This is true because T2 determination in this study utilizes T2 SNR at shorter TE values lower than 91 ms. It is worth noting that the T2 values calculated for both $FeCl_3$ and $GdCl_3$ added agarose gel phantoms at TP5 exhibit stability regardless of the relaxation modifier content.

Prototyping the $FeCl_3$ and $GdCl_3$ added agarose gel phantoms as a systematic MRI phantom [17] should be the focus of future studies to investigate their suitability to be used as test phantoms for different MRI sequences and QC parameters. In addition, studies on alternative materials other than agar and agarose gel [16] should continue so that future works will have a wider choice of materials that can be prepared at a lower cost [18]. The use of $FeCl_3$ and $GdCl_3$ solutions seems promising due to their ability to reduce T1 relaxation time markedly.

This study is limited by the choice of TR and TE values during the T1 and T2 measurements. The lower bound for TR should be at least 50 ms for the relaxation part of the T1 curve to become observable, allowing T1 relaxation time to be estimated. The upper bound is not necessarily longer than 1000 ms because the curves have already reached saturation even for TR = 800 ms. For TE, the range of 10 ms – 100 ms should be sufficient for the relaxation modifiers used in this study. The results for $FeCl_3$ and $GdCl_3$ added agarose gel phantoms obtained from this study should also be compared with the results at different TP for distilled water, pure agarose, pure $FeCl_3$, and pure $GdCl_3$ at least qualitatively to avoid biases.

CONCLUSIONS

In conclusion, agarose gel phantoms added with $GdCl_3$ and $FeCl_3$ displayed unstable T1 SNR after the time of preparation. In addition, T1 SNR showed a slight decrease with TR but a marked decrease with $GdCl_3$ and $FeCl_3$ addition. The T2 curves for all phantoms decrease exponentially with TE but appear the same at all TP indicating that TP is noninfluential on T2 relaxation. Changes in T2 SNR with $GdCl_3$ and $FeCl_3$ addition can only be observed below TE = 91 ms. While the T1 values for the phantoms were not able

to be determined in this study, their T2 values show an inconsistent trend with increasing relaxation modifier content at any different TPs. The T1 SNR of the laboratory-prepared $GdCl_3$ and $FeCl_3$ agarose gel phantoms were not stable over the time range used in this study. Even though T2 SNR showed an acceptable consistency, the signal is relatively small and the phantoms' T2 relaxation can only be differentiated at TE values lower than 91 ms. The T1 relaxation of the agarose gel phantoms added with $GdCl_3$ and $FeCl_3$ cannot be compared with that of the water phantom due to the absence of the relaxation part of the T1 curves. For T2, the values obtained from the phantoms are much shorter than the T2 of the water phantom due to the difference in the content of both phantoms.

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