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Quantitative ferumoxytol-enhanced MRI in pregnancy: A feasibility study in the nonhuman primate

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Abstract

Objectives: To assess the feasibility of ferumoxytol-enhanced MRI in pregnancy with a nonhuman primate model.

Materials and methods: In this prospective study, eleven pregnant rhesus macaques at day 98 ± 5 of gestation were divided into three groups, untreated control (UC) ($n = 3$), saline control (SC) ($n = 4$) and interleukin 1 beta (IL-1 β) treated (IT) ($n = 4$), which were administered with either saline or IL-1 β into the amniotic fluid. All animals were imaged at multiple time points before and after ferumoxytol administration (4 mg/kg). Longitudinal $R2^*$ and susceptibility of tissues were obtained using region-of-interest analysis and the longitudinal changes were assessed using linear mixed models and Student's t -test.

Results: In fetuses, a slope of $0.3s^{-1}/\text{day}$ ($P = 0.008$), $0.00\text{ppm}/\text{day}$ ($P = 0.699$) and $-0.2s^{-1}/\text{day}$ ($P = 0.023$) was observed in liver $R2^*$, liver susceptibility, and lung $R2^*$, respectively. In

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Declaration of competing interest

Dr. Diego Hernando has no relevant conflicts. Dr. Diego Hernando has ownership interest in Calimetrix. Dr. Scott B. Reeder has no relevant conflicts. Dr. Scott B. Reeder consults for ArTara Therapeutics, and has ownership interests in Calimetrix, Reveal Pharmaceuticals, Collectar Biosciences, and Elucent Medical. The other authors have no conflicts of interest. The University of Wisconsin receives research support from GE Healthcare and Bracco Diagnostics.

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.mri.2019.10.006>.

placentas, $R2^*$ and susceptibility increased immediately after ferumoxytol administration ($P < 0.001$) and decreased to baseline within two days. The mean change from baseline showed no significant difference between the SC group and the IT group at all scan time points. In maternal livers, $R2^*$ increased immediately after ferumoxytol administration, further increased at one-day, and then decreased but remained elevated ($P < 0.001$). The mean change from baseline showed no significant difference between the SC group and the IT group at all scan time points.

Conclusions: This work demonstrates the feasibility of quantitative ferumoxytol-enhanced MRI to measure dynamics of ferumoxytol delivery and washout in the placenta. Stable MRI measurements indicated no evidence of iron deposition in fetal tissues of nonhuman primates after maternal ferumoxytol exposure.

Keywords

Placenta; Ferumoxytol; Quantitative; $R2^*$; Magnetic susceptibility

1. Introduction

Preeclampsia is an inflammatory condition of human pregnancy that can lead to peripheral edema, hypertension, seizures and maternal death [1,2]. Maternal vascular dysfunction at the maternal-fetal interface in preeclampsia leads to insufficient placental perfusion and activation of the uteroplacental inflammatory response [3,4]. Altered immune cell (e.g. macrophage) activation and distribution within the maternal-fetal interface may also play an important role in the development of adverse pregnancy outcomes [5]. Evaluation of macrophage distribution may enable early assessment of the developing pathophysiologic conditions in preeclampsia and/or inflammation-induced fetal growth restriction before their clinical manifestation. Thus, evaluation of macrophage distribution and activity is highly desirable for identifying immune cell homing in pregnancy.

Ferumoxytol is an FDA-approved iron compound for treatment of anemia [6] which recently has received substantial interest as an off-label contrast agent in MRI [7,8]. Ferumoxytol particles are phagocytosed by macrophage in the reticuloendothelial system or sites with activated inflammation responses [9]. Importantly, the presence of ferumoxytol affects MR contrast parameters, including $R2^*$ ($= 1 / T2^*$) and magnetic susceptibility. Quantitative $R2^*$ mapping and quantitative susceptibility mapping (QSM) have been used for detection of ferumoxytol [10–12] and characterization of microscopic iron distribution [13,14]. Ferumoxytol-enhanced MRI has been shown to enable non-invasive assessment of macrophages in applications including pancreatic inflammation in type I diabetes, high-grade gliomas, and inflammation after myocardial infarction [15–17]. Therefore, ferumoxytol-enhanced MRI may enable sensitive detection of macrophage homing in inflammation at the maternal-fetal interface.

However, the use of ferumoxytol for the assessment of macrophage distribution in pregnancy, and the safety of the ferumoxytol use, including the potential uptake in the fetus after maternal administration, have not been investigated. Unlike gadolinium-based contrast agents with an approximate diameter of 0.4 nm [9], which cross the placenta into the fetal circulation [18,19], ferumoxytol with an approximate diameter of 30 nm [9] may not

transport directly into fetal circulation [20]. The nonhuman primate has the most similar placentation, physiology and immunology with the human reproductive system [21–23]. Thus, quantification of iron accumulation at the maternal-fetal interface and in the fetus after ferumoxytol exposure in nonhuman primates would constitute an important step towards evaluating the feasibility and safety of ferumoxytol-enhanced MRI in human pregnancy.

The hypothesis for this work was that maternal ferumoxytol exposure will not lead to increased iron deposition in fetal tissues. Further, we hypothesize that ferumoxytol will be taken up by macrophages in the placenta or the decidua, the endometrium of pregnancy, in inflammation with macrophage homing and will be detectable by MRI. Therefore, the purpose of this study was to assess the feasibility of ferumoxytol-enhanced R2* mapping and QSM in a pregnant nonhuman primate model.

2. Materials and methods

2.1. Animals and experimental procedure

This prospective animal study was approved by our institution's animal care and use committee. Interleukin 1 beta (IL-1 β), a pro-inflammatory cytokine, was used to generate inflammation at the maternal-fetal interface. Eleven pregnant rhesus macaques at day 98 ± 5 of gestation (average term pregnancy = 165 days), equivalent to the late second trimester of human pregnancy, were obtained. Animals were divided into three groups, *i.e.*, untreated control (UC) ($n = 3$), saline control (SC) ($n = 4$), and IL-1 β treated (IT) ($n = 4$). For all procedures and MRI scans, the animal was anesthetized by administration of up to 10 mg/kg ketamine and the sedation was prolonged by delivering oxygen with 1.5% isoflurane through inhalation. The animal was monitored during all procedures, and subsequently until fully recovery from the anesthesia. Veterinarian staff were alerted for treatment if the recovery took > 90 mins, or if there were symptoms of compromised health.

Each animal in the UC group was imaged first as baseline (Day 0-pre). Subsequently, ferumoxytol (AMAG Pharmaceuticals, Waltham, MA, USA) was administered intravenously at a dose of 4 mg/kg, diluted 5:1 with sterile saline. Immediately after ferumoxytol administration, the animal was imaged again (Day 0-post) and follow-up MRI scans were performed afterwards at four time points within 25 days (Day 1–25), as shown in the flow chart of Fig. 1. Animal information, drug dose, ferumoxytol administration, and scan time points are detailed in Supporting information Table S1.

Each animal in either the SC or IT group was imaged first as baseline (Day $-2/-1$). On the same day after the baseline imaging, animals were administered either saline (0.5 mL) or IL-1P (10 μ g IL-1 β in 0.5 mL saline) into the amniotic using a needle inserted through an aseptically prepared site of the abdominal wall and introduced until the tip touched the uterus wall. Subsequent scans were started one or two days later, in order to allow for inflammation to develop. The animals were then imaged before and immediately after ferumoxytol administration (Day 0 pre/post) and follow-up scans were performed afterwards within three days (Day 1–3), as shown in the flow chart of Fig. 1. Animal information, drug dose, ferumoxytol administration and scan time points are detailed in Supporting

information Table S1. Histology and iron quantification analysis on placental and fetal tissues were performed after the last MRI scan.

2.2. MRI acquisition

MRI scans were performed on a 3.0 T MRI system (Discovery 750, GE Healthcare, Waukesha, WI, USA) using a 32-channel torso coil (Neocoil, Pewaukee, WI, USA) with the animal in the left lateral decubitus position. An axial 3D multi-echo spoiled gradient-echo (SGRE) acquisition of the maternal pelvis covering the uterus cavity was obtained to evaluate the ferumoxytol distribution at the maternal-fetal interface. Additionally, the ferumoxytol distribution in the maternal reticuloendothelial system including the maternal liver was also evaluated in this study. For this purpose, an axial 3D multi-echo SGRE radial ultra-short TE (UTE-SGRE) acquisition of the maternal abdomen covering the maternal liver was performed. Respiratory-gating [24] was used in both acquisitions. Scan parameters are detailed in Table 1.

2.3. Imaging reconstruction and data analysis

R2* maps of the maternal pelvis were generated from the SGRE images using a chemical shift-encoded reconstruction [25]. R2* maps of the maternal abdomen were obtained by using monoexponential fit of echoes in the UTE-SGRE images. In each animal at each scan time point, R2* of the amniotic fluid, the fetal liver, the fetal lung, the two separate placental discs, and the maternal liver were measured in oval regions of interest (ROIs). Information on ROIs is detailed in Supporting information Table S2.

Susceptibility maps of the maternal pelvis were generated from the SGRE images using a QSM reconstruction [26]. All susceptibility measurements were calculated relative to the adjacent amniotic fluid, which was used as a susceptibility reference as no ferumoxytol uptake was observed as shown below. The fetal liver susceptibility was measured by calculating the difference between the susceptibility values in the fetal liver and the amniotic fluid. The placental susceptibility was measured by calculating the difference between the susceptibility values in one of the placental discs and the amniotic fluid. Information on ROIs is detailed in Supporting information Table S2. Susceptibilities of the fetal lung and the maternal liver were not obtained due to the lack of adjacent susceptibility reference and the limited range of echo times in UTE-SGRE, respectively.

2.4. Statistical analysis

To investigate the longitudinal variation of R2* and susceptibility measurements of the fetal liver and R2* measurements of the fetal lung, a linear mixed-effects model was used. The model included the time interval of each scan to the baseline scan as a fixed continuous factor, the animal groups as a fixed classification factor, the interaction of the animal group and the time interval as a fixed factor, and animal as a random classification factor.

To investigate the longitudinal variation of placental R2* and susceptibility measurements, and maternal liver R2*, respectively, a linear-mixed effects model was used in each animal group, including the scan time point as a fixed classification factor and animal as a random classification factor. The changes at each scan time point from baseline were calculated for

placental $R2^*$ as $R2^*_p = R2^*_{p, \text{TimePoint}} - R2^*_{p, \text{Baseline}}$, for placental susceptibility as $\chi_p = \chi_{p, \text{TimePoint}} - \chi_{p, \text{Baseline}}$, and for maternal liver $R2^*$ as $R2^*_l = R2^*_{l, \text{TimePoint}} - R2^*_{l, \text{Baseline}}$. To investigate the inter-group difference on the longitudinal variation of MRI measurements, Student's t-test with conservative Bonferroni correction for multiple comparison was used to compare $R2^*_p$ at each time point between the SC and IT groups, as well as χ_p and $R2^*_l$. Statistical analyses were performed in Matlab (MathWorks, Natick, MA, USA). Statistical significance was selected as a P value < 0.05 .

3. Results

One animal in the UC group (Rhesus #9) and one in the IT group (Rhesus #4) which had a history of ocular swelling not associated with ferumoxytol were noted to have moderate periocular swelling/edema following the ferumoxytol administration. They were treated with 10 mg diphenhydramine hydrochloride and recovered without further medical intervention. No complications were observed in all the other animals during or after ferumoxytol administration. 3D SGRE acquisitions of the maternal pelvis were successfully obtained without apparent motion artifacts in the reconstructed $R2^*$ and susceptibility maps, except for the Day 0-pre acquisition in Rhesus #2 when the respiratory gating failed. 3D UTE-SGRE MRI of maternal abdomen was successfully obtained at most time points as listed in Supporting information Table S3.

The MRI scan time points for Rhesus #3 in the UC group, Rhesus #4 in the IT group and Rhesus #5 in the SC group are shown at the top of Figs. 2 and 3. Anatomic images, $R2^*$ maps, and susceptibility maps are shown at each scan time point for each animal. The fetal liver (red lines), the fetal lung (pink lines), the maternal liver (white lines), the uterine cavity (yellow lines), the two separate placental discs (orange lines), and the amniotic fluid (green arrows) are delineated and indicated in Figs. 2 and 3. Placental $R2^*$ and susceptibility measurements of animals in all groups increased substantially after ferumoxytol administration (Day 0-post) and decreased towards baseline at Day 1 and afterwards. Maternal liver $R2^*$ in Rhesus #3 increased immediately after ferumoxytol administration (Day 0-post), further increased at one-day follow-up (Day 1), and subsequently decreased but remained elevated three weeks afterwards (Days 7–21).

Longitudinal measurements and changes from baseline in $R2^*$ of the amniotic fluid, $R2^*$ and susceptibility of fetal liver and $R2^*$ of fetal lung in all animals are summarized in Fig. 4 and Table 2. Stable $R2^*$ measurements of the amniotic fluid before and after ferumoxytol administration indicated no ferumoxytol deposition there. It further indicated the amniotic fluid to be a reliable reference tissue for the susceptibility measurement. In the UC group, a slope of $0.3 \text{ s}^{-1}/\text{day}$ ($P = 0.008$) and a slope of $0.00 \text{ ppm}/\text{day}$ ($P = 0.699$) were observed in fetal liver $R2^*$ and susceptibility, respectively. A slope of $-0.2 \text{ s}^{-1}/\text{day}$ ($P = 0.023$) was observed in fetal lung $R2^*$. An intercept of -0.09 ppm ($P = 0.003$) was observed in susceptibility of fetal liver in the IT group compared to the measurements in the UC group. The remaining measurements of $R2^*$ and susceptibility in the fetal liver and the fetal lung in the SC and IT groups were not significantly different from the reference, *i.e.*, the UC group. Based on previous published calibrations in phantoms [27–30], the additional iron deposition after maternal ferumoxytol exposure, indicated by the MRI measurements, is $<$

9.0, 7.0 and 4.3 micrograms iron per gram of tissue in the amniotic fluid, the fetal liver and the fetal lung, respectively (see Supporting information S1).

Longitudinal measurements and changes from baseline in placental R2* and susceptibility and R2* of maternal liver in the UC group are summarized in Fig. 5 and Table 3. Example ROIs are shown in a previous study [31]. Placental R2* and susceptibility increased significantly after ferumoxytol administration at Day 0-post ($P < 0.001$). Placental R2* and susceptibility at Day 1 decreased back to the baseline and showed no significant differences at all four follow-up scans ($P = 0.395$, $P = 0.627$, $P = 0.905$ and $P = 0.559$ in R2*, respectively, and $P = 0.172$, $P = 0.859$, $P = 0.974$ and $P = 0.948$ in susceptibility, respectively). Maternal liver R2* increased after ferumoxytol administration at Day 0-post ($P < 0.001$), further increased at one-day follow-up ($P < 0.001$) and decreased but remained elevated at all three follow-up scans ($P < 0.001$).

Longitudinal measurements and changes from baseline in placental R2* and susceptibility and maternal R2* in the SC and IT groups are summarized in Fig. 6 and Table 4. After administration of either saline or IL-1 β measured at Day 0-pre, placental R2* and susceptibility did not significantly change. Immediately after ferumoxytol administration, placental R2* and susceptibility in both groups increased significantly ($P < 0.001$). At one-day follow-up, placental R2* and susceptibility decreased towards baseline but remained elevated in both groups. At two- or three-day follow-up, placental R2* and susceptibility further decreased towards baseline in both groups. At all time points, the mean change of either placental R2* or susceptibility from baseline showed no significant difference between the SC group and the IT group. Based on previous published calibrations in phantoms [27–29,32], the maximum increase of R2* at Day 2/3 relatively to the baseline in all animals indicated a maximum iron concentration of 16.3 micrograms iron per gram of placental tissue (see Supporting information S2).

Maternal liver R2* was not significantly altered after administration of either saline or IL-1 β measured at Day 0-pre. Immediately after ferumoxytol administration, maternal liver R2* increased significantly in both groups ($P < 0.009$). At one-day follow-up, maternal liver R2* further increased in both groups. At two- or three-day follow-up, maternal liver R2* decreased but remained elevated in both groups. At all time points, the mean change of maternal liver R2* from baseline showed no significant difference between the SC group and the IT group.

4. Discussion

This work demonstrated the feasibility of R2* mapping and QSM to measure the ferumoxytol delivery and washout from the placental tissues. Additionally, stable R2* and tissue susceptibility were observed in fetal tissues before and after ferumoxytol administration in nonhuman primates, indicating no increased iron deposition in the fetus after maternal exposure.

The elevated maternal liver R2* immediately after ferumoxytol administration and the further increase of R2* at one-day follow-up were likely due to the presence of ferumoxytol

in the blood pool [9,33]. Maternal liver R2* decreased subsequently over the following three weeks, but did not return to the baseline over this time span. This suggests iron uptake in the maternal liver as expected to occur following ferumoxytol administration in treatment of anemia [33].

The elevated placental R2* and susceptibility immediately after ferumoxytol administration were also likely due to the presence of ferumoxytol in the blood pool [9,34]. At one-day follow-up, placental R2* and susceptibility remained elevated, most likely due to the remaining ferumoxytol in the blood pool [9,34]. Further, the lack of significantly elevated placental R2* at one-, two- or three-day follow-up, compared to the elevated maternal liver R2*, indicated a lack of ferumoxytol uptake in the placenta.

Importantly, the lack of significant amount of iron deposit in fetal tissues, compared to the iron uptake in the maternal liver, suggests that ferumoxytol does not cross into the amniotic cavity and may not enter the fetal circulation. This is an important observation towards evaluating the safety implications for the use of ferumoxytol in pregnancy. However, delayed effects of elevated iron content in the maternal body (as shown in maternal liver R2*) on the fetus are unknown. To further investigate the safety of the ferumoxytol use in pregnancy, future studies are needed to assess iron contents in offspring with *in utero* exposure to ferumoxytol. In addition, ferumoxytol administration in animals at different gestation stages also need to be studied.

In the UC group, slightly elevated R2* measurements were observed in the fetal liver over time after maternal ferumoxytol exposure. However, a previous study has shown an increase of human fetal liver R2* over gestation stage in normal fetuses without maternal ferumoxytol exposure [35]. It cannot be determined from the current study whether the elevated fetal liver R2* is due to the normal development of the reticuloendothelial system over gestation stage [36,37], or due to the maternal exposure of ferumoxytol. Quantification of iron content in fetuses without maternal ferumoxytol exposure is under separate study [38].

Tissue R2* and susceptibility measurements were both obtained as potential imaging biomarkers of iron content in this study. In the placenta, both R2* and susceptibility measurements showed consistent behavior after ferumoxytol administration, even though R2* and susceptibility may have different dependence on the presence of iron [39]. The fact that R2* and susceptibility can be obtained as separate measurements from the same 3D SGRE data may enable a comprehensive evaluation of iron content and microscopic iron distributions [14] in a single acquisition.

This study has several limitations. The IL-1 β treatment was ineffective at inducing the pathology reported in a previous study [40] where neutrophil and macrophage infiltration of the decidua parietalis was demonstrated. However, in this previous study the decidua basalis or the placenta itself was not evaluated; it is possible that the inflammatory response on the maternal side is limited to fetal membranes. With the MRI techniques in this study, it was not possible to resolve the thin fetal membranes and decidua parietalis. The potential use of ferumoxytol-enhanced MRI for the detection of macrophage homing to the decidua basalis

and placenta needs further validation in an effective macrophage homing inflammation model. Additionally, a larger population of animals is needed to capture the biological variation on the iron biodistribution after maternal ferumoxytol exposure.

In conclusion, this study demonstrated the feasibility of longitudinal tracking of placental iron content in ferumoxytol-enhanced MRI. Additionally, this study suggested ferumoxytol may not cross the placenta and hence enter into the amniotic cavity or into the fetal circulation after maternal ferumoxytol exposure in nonhuman primates at late second trimester. Upon successful detection of macrophage homing in an effective inflammation model and being proven safe, ferumoxytol-enhanced MRI may enable assessment of the inflammation at the maternal-fetal interface in human pregnancy.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Fig. 1. Flow chart of the MRI scan time points relative to the administration of ferumoxytol, saline and IL-1 β for all eleven animals.

Fig. 2. Longitudinal axial anatomic image (1st and 4th row), R2* (2nd and 5th row) and susceptibility (3rd row) maps of fetal liver (red lines), fetal lung (pink lines) and maternal liver (white lines) of Rhesus #3 (day 91 of gestation) in the untreated control group. Stable R2* and susceptibility of fetal tissues after maternal ferumoxytol administration were observed. Maternal liver R2* increased after ferumoxytol administration, further increased at one-day follow-up (Day 1), and remained elevated at seven-, fourteen-, and twenty-one day follow-up.

Fig. 3. Placental R_2^* and susceptibility of Rhesus #5 (day 103 of gestation, bottom three rows) in the saline control group and Rhesus #4 (day 92 of gestation, top three rows) in the interleukin 1 beta (IL-1 β) treated group increased after ferumoxytol administration (at Day 0-post) and decreased towards baseline at Day 1 and afterwards. The amniotic fluid is indicated by green arrow. The uterus cavity and the two separate placental discs are delineated by yellow lines and orange lines, respectively.

Fig. 4.

Stable R_2^* measurements of the amniotic fluid indicate no ferumoxytol uptake, suggesting the amniotic fluid as a reliable reference tissue for the susceptibility measurement. Fetal liver R_2^* and susceptibility measurements and fetal lung R_2^* measurements in all animals are not significantly increased over time after maternal ferumoxytol administration at Day 0-post.

Fig. 5.

Longitudinal measurements of placental R_2^* and susceptibility and maternal liver R_2^* in the untreated control group. Both the placental R_2^* and susceptibility increased significantly after ferumoxytol administration at Day 0-post and decreased back to the baseline at follow-up scans. Maternal liver R_2^* increased after ferumoxytol administration and remained elevated at follow-up scans.

Fig. 6.

Placental R_2^* and susceptibility (χ) measurements of all animals in the saline control group and the interleukin 1 beta (IL-1 β) treated group increased after ferumoxytol administration at Day 0-post and decreased back to the baseline at follow-up scans. The difference of measurements at each scan time point compared to that at baseline, *i.e.*, R_2^* and χ , showed no difference between the saline control group and the IL-1 β treated group. Maternal liver R_2^* in both groups immediately increased after ferumoxytol administration, further increased at Day 1, and remained elevated at Day 2 or Day 3.

Table 1
Scan parameters of maternal pelvic MRI (SGRE) and maternal abdominal MRI (UTE-SGRE) of the animal.

	# TE	TE _{int} / TE (ms)	TR (ms)	Flip angle (°)	Field of view (mm ³)	Acquired resolution (mm ⁵)	Reconstructed resolution (mm ⁵)	Parallel imaging acceleration phase/slice	Scan time (min)
Maternal pelvic MRI: SGRE	8	1.6/1.3	12.9	9	160 × 160 × 140	1.1 × 1.1 × 1.9	0.6 × 0.6 × 1.9	1.5/1.0	4:51
Maternal abdominal MRI: UTE-SGRE	3	0.1/0.2	2.8	3	180 × 180 × 180	1.4 × 1.4 × 1.4	1.4 × 1.4 × 1.4	1.0/1.0	5:30

SGRE=spoiled gradient-echo, UTE=ultra-short echo time.

Table 2

Longitudinal variation of R2* and susceptibility measurements of the fetal liver and R2* measurements of the fetal lung over time in all animals.

	Estimated change in fetal liver R2* (s ⁻¹)	P value	Estimated change in fetal liver susceptibility (ppm)	P value	Estimated change in fetal lung R2* (s ⁻¹)	P value
UC group (reference)	Intercept: 30.0 (25.1, 35.0) Slope: 0.3 (0.1, 0.6)	< 0.001	Intercept: 0.09 (0.05, 0.14) Slope: 0.00 (-0.00, 0.00)	< 0.001	Intercept: 16.3 (14.0, 18.5) Slope: -0.2 (-0.4, 0.0)	< 0.001
SC group	Intercept: -2.3 (-8.7, 4.2) Slope: 0.9 (-0.8, 2.2)	0.008 0.489	Intercept: -0.06 (-0.11, 0.00) Slope: 0.00 (-0.03, 0.02)	0.699 0.046	Intercept: -2.0 (-4.8, 0.8) Slope: 0.4 (-0.8, 1.6)	0.023 0.081
IT group	Intercept: -2.7 (-9.2, 3.8) Slope: 0.3 (-1.5, 2.1)	0.365 0.411	Intercept: -0.09 (-0.15, -0.03) Slope: 0.03 (0.00, 0.06)	0.886 0.003	Intercept: 4.5 (-0.6, 9.5) Slope: 1.0 (-0.5, 2.4)	0.544 0.081
		0.765		0.057		0.186

Data in parentheses are 95% confidence intervals, IL-1β = interleukin 1beta, UC = untreated control, IT = interleukin 1beta treated, SC = saline control.

Table 3

Estimated change of placental R2* and susceptibility measurements at scan time points after ferumoxytol administration from the baseline in the untreated control group.

	Estimated change in placental R2* (s ⁻¹)			Estimated change in placental susceptibility (ppm)			Estimated change in maternal liver ^a R2* (s ⁻¹)		
	Intercept	P value	Intercept	Intercept	P value	Intercept	Intercept	P value	
Day 0-pre vs. Day 0-post	312.8 (289.6, 335.9)	<0.001	1.1 (0.9, 1.3)		<0.001	276.8 (217.1, 301.0)		<0.001	
Day 0-pre vs. Day 1	9.4 (-13.8, 32.6)	0.395	0.2 (-0.1, 0.4)		0.172	1049.4 (1013.8, 1085.0)		<0.001	
Day 0-pre vs. Day 4/7	5.3 (-17.9, 28.5)	0.627	0.0 (-0.2, 0.2)		0.859	405.2 (369.6, 440.8)		<0.001	
Day 0-pre vs. Day 12/14	1.3 (-21.9, 24.5)	0.905	0.0 (-0.2, 0.2)		0.974	364.7 (329.0, 400.3)		<0.001	
Day 0-pre vs. Day 21/25	6.4 (-16.8, 29.6)	0.559	0.0 (-0.2, 0.2)		0.948	326.2 (290.6, 361.8)		<0.001	

Data in parentheses are 95% confidence intervals, vs. = versus.

^aData of Rhesus #1 in the untreated control group were not available at all scan time points and thus not included.

Estimated change of placental R2* and susceptibility (χ) measurements at scan time points before and after ferumoxytol administration from the baseline in the saline control group and the untreated control group.

Table 4

	Saline control (<i>n</i> = 4)			IL-1 β treated (<i>n</i> = 4)			SC vs. IT	
	Estimated change placental	R2* (s ⁻¹)	<i>P</i> value	Estimated change placental	R2* (s ⁻¹)	<i>P</i> value	<i>P</i> value ^a	<i>P</i> value ^a
Day 0-pre vs. Day -1/-2	-0.5 (-47.3, 46.4)		0.984	0.2 (-82.1, 82.5)		0.996	0.867	
Day 0-post vs. Day -1/-2	444.0 (397.1, 490.8)		<0.001	426.3 (343.9, 508.6)		<0.001	0.824	
Day 1 vs. Day -1/-2	21.9 (-24.9, 68.8)		0.338	33.6 (-48.7, 115.9)		0.398	0.598	
Day 2/3 vs. Day -1/-2	9.4 (-37.4, 56.3)		0.677	7.7 (-74.7, 90.0)		0.845	0.733	
	Estimated change placental	χ (ppm)	<i>P</i> value	Estimated change placental	χ (ppm)	<i>P</i> value	<i>P</i> value ^a	<i>P</i> value ^a
	Estimated change maternal liver	R2* (s ⁻¹)	<i>P</i> value	Estimated change maternal liver	R2* (s ⁻¹)	<i>P</i> value	<i>P</i> value ^a	<i>P</i> value ^a
Day 0-pre vs. Day -1/-2	0.06 (-0.15, 0.28)		0.541	0.02 (-0.31, 0.34)		0.917	0.339	
Day 0-post vs. Day -1/-2	1.87 (1.60, 2.13)		<0.001	1.62 (1.29, 1.95)		<0.001	0.385	
Day 1 vs. Day -1/-2	0.24 (-0.02, 0.51)		0.031	0.27 (-0.05, 0.60)		0.095	0.818	
Day 2/3 vs. Day -1/-2	0.11 (-0.11, 0.33)		0.293	0.07 (-0.26, 0.39)		0.671	0.402	
	Estimated change maternal liver	R2* (s ⁻¹)	<i>P</i> value	Estimated change maternal liver	R2* (s ⁻¹)	<i>P</i> value	<i>P</i> value ^a	<i>P</i> value ^a
	Estimated change maternal liver	R2* (s ⁻¹)	<i>P</i> value	Estimated change maternal liver	R2* (s ⁻¹)	<i>P</i> value	<i>P</i> value ^a	<i>P</i> value ^a
Day 0-pre vs. Day -1/-2	3.5 (-211.9, 218.8)		0.972	24.6 (-317.9, 367.1)		0.879	0.591	
Day 0-post vs. Day -1/-2	429.8 (214.5, 645.2)		0.001	598.9 (179.5, 1018.4)		0.009	0.534	
Day 1 vs. Day -1/-2	1261.4 (1046.0, 1476.7)		<0.002	1910.3 (1567.8, 2252.8)		<0.001	0.028	
Day 2/3 vs. Day -1/-2	1046.9 (831.6, 1262.3)		<0.001	1331.4 (988.9, 1673.9)		<0.001	0.320	

Data in parentheses are 95% confidence intervals, IL-1 β = interleukin 1beta, vs. = versus, SC = saline control, IT = IL-1 β treated.

^aData were compared by using the Student's t-test with conservative Bonferroni correction for multiple comparison. Threshold for statistical significance was set to $P < 0.0125$ after correction.