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# Splenic T1-mapping: a novel quantitative method for assessing adenosine stress adequacy for cardiovascular magnetic resonance

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## Abstract

**Background:** Perfusion cardiovascular magnetic resonance (CMR) performed with inadequate adenosine stress leads to false-negative results and suboptimal clinical management. The recently proposed marker of adequate stress, the “splenic switch-off” sign, detects splenic blood flow attenuation during stress perfusion (spleen appears dark), but can only be assessed after gadolinium first-pass, when it is too late to optimize the stress response. Reduction in splenic blood volume during adenosine stress is expected to shorten native splenic T1, which may predict splenic switch-off without the need for gadolinium.

**Methods:** Two-hundred and twelve subjects underwent adenosine stress CMR: 1.5 T ( $n = 104$ ; 75 patients, 29 healthy controls); 3 T ( $n = 108$ ; 86 patients, 22 healthy controls). Native  $T1_{\text{spleen}}$  was assessed using heart-rate-independent ShMOLLI prototype sequence at rest and during adenosine stress (140  $\mu\text{g/kg/min}$ , 4 min, IV) in 3 short-axis slices (basal, mid-ventricular, apical). This was compared with changes in peak splenic perfusion signal intensity ( $\Delta SI_{\text{spleen}}$ ) and the “splenic switch-off” sign on conventional stress/rest gadolinium perfusion imaging.  $T1_{\text{spleen}}$  values were obtained blinded to perfusion  $\Delta SI_{\text{spleen}}$ , both were derived using regions of interest carefully placed to avoid artefacts and partial-volume effects.

**Results:** Normal resting splenic T1 values were  $1102 \pm 66$  ms (1.5 T) and  $1352 \pm 114$  ms (3 T), slightly higher than in patients ( $1083 \pm 59$  ms,  $p = 0.04$ ;  $1295 \pm 105$  ms,  $p = 0.01$ , respectively).  $T1_{\text{spleen}}$  decreased significantly during adenosine stress (mean  $\Delta T1_{\text{spleen}} \sim -40$  ms), independent of field strength, age, gender, and cardiovascular diseases. While  $\Delta T1_{\text{spleen}}$  correlated strongly with  $\Delta SI_{\text{spleen}}$  ( $\rho = 0.70$ ,  $p < 0.0001$ ); neither indices showed significant correlations with conventional hemodynamic markers (rate pressure product) during stress. By ROC analysis, a  $\Delta T1_{\text{spleen}}$  threshold of  $\geq -30$  ms during stress predicted the “splenic switch-off” sign (AUC 0.90,  $p < 0.0001$ ) with sensitivity (90%), specificity (88%), accuracy (90%), PPV (98%), NPV (42%).

**Conclusions:** Adenosine stress and rest splenic T1-mapping is a novel method for assessing stress responses, independent of conventional hemodynamic parameters. It enables prediction of the visual “splenic switch-off” sign without the need for gadolinium, and correlates well to changes in splenic signal intensity during stress/rest perfusion imaging.  $\Delta T1_{\text{spleen}}$  holds promise to facilitate optimization of stress responses before gadolinium first-pass perfusion CMR.

**Keywords:** Cardiovascular magnetic resonance, Adenosine stress, Splenic T1, Switch-off, ShMOLLI

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## Background

Adenosine stress perfusion cardiovascular magnetic resonance (CMR) accurately detects myocardial ischemia and guides clinical decision-making [1, 2]. However, perfusion CMR has a reported false-negative rate of between 5 and 16% [2–4], which may lead to suboptimal management strategies. In the absence of poor image quality, inadequate adenosine stress response is the commonest cause of false-negative perfusion scans [4], because conventional hemodynamic markers of stress response, such as heart rate and systolic blood pressure, are unreliable predictors of myocardial vasodilatation and the achievement of maximal hyperemia [5].

Recently, the “splenic switch-off” sign was proposed as a CMR marker of adequate adenosine stress. It describes visually reduced splenic perfusion during stress imaging (spleen appears dark) compared to rest imaging (spleen appears bright) [6], and in retrospective analyses, failed splenic switch-off was more commonly observed in false-negative perfusion scans than true-negatives [6]. The physiological basis for this phenomenon is that splenic blood volume reduces significantly during exercise, due to splanchnic blood redistribution [7, 8], and can manifest as splenic “disappearance” on nuclear imaging [9]. The degree of splenic blood volume reduction is proportional to exercise workload [7], independent of cardiac output [7], and is related to adenosine-mediated splenic vasoconstriction [10, 11]. More recently, splenic switch-off has been shown to relate to higher myocardial T2 values during dipyridamole stress, further suggesting a connection between splenic and myocardial vascular biology [12].

A key limitation of splenic switch-off is that it can only be assessed after gadolinium first-pass perfusion imaging [6], at which point it is too late to optimize stress adequacy [13]. Repetition of inadequately stressed images would require a wait-period (10–15 min) for gadolinium “wash-out” from the LV cavity to optimize myocardial-blood contrast during the subsequent (no longer first-pass) stress perfusion imaging, leading to longer scan durations, and exposes patients to additional adenosine and contrast agents [6]. Therefore, a method which can determine stress adequacy and offer opportunities for pre-emptive stress response optimization *before* gadolinium first-pass perfusion imaging is highly desirable.

Native T1-mapping enables quantitative characterization of tissue blood volumes without the need for gadolinium-based contrast agents (GBCA) [14–16], and offers the potential to assess stress responses before GBCA first-pass perfusion. T1 (proton spin-lattice) relaxation time is a magnetic property of tissues measured in milliseconds [14], and each tissue type, including the spleen, has its own normal range of T1 values [14]. T1 is sensitive to changes in tissue water content or blood volume [15–19],

and we recently showed that normal myocardial T1 increases by 6% during adenosine vasodilatory stress, due to expansions in myocardial blood volume [15, 16]. Furthermore, stress-T1 appears sensitive to changes in normal, ischemic and infarcted myocardium, without the need for GBCA [15]. Contrary to its vasodilatory effects in the myocardium, adenosine causes splenic *vasoconstriction*, reducing the splenic blood volume, and thus expected to lower the splenic T1 ( $T1_{\text{spleen}}$ ). Conveniently, the spleen is typically visible on stress perfusion CMR and can be inspected without additional planning.

This study sought to evaluate stress and rest  $T1_{\text{spleen}}$  as a gadolinium-free CMR marker of adenosine stress responses by comparing with the existing “splenic switch-off” sign and hemodynamic markers. We hypothesized that: (i)  $T1_{\text{spleen}}$  will decrease significantly from resting values during adenosine stress, due to splenic blood volume reductions and; (ii) stress-related changes in  $T1_{\text{spleen}}$  ( $\Delta T1_{\text{spleen}}$ ) correlate to changes in splenic perfusion on CMR (the “splenic switch-off” sign), but without the need for GBCA.

## Methods

All study procedures received favourable opinions from local ethics committees, and all subjects gave written informed consent.

### Study population

To establish the relationship between  $T1_{\text{spleen}}$  and splenic perfusion/switch-off, retrospective analysis was performed on CMR scans of 212 subjects; 104 subjects had CMR at 1.5 T (Magnetom Avanto, Siemens Healthcare, Erlangen, Germany) and 108 subjects had CMR at 3 T (Magnetom Trio a Tim system, Siemens Healthcare, Erlangen, Germany). The 1.5 T population had 75 patients ( $n = 36$  known coronary artery disease [CAD],  $n = 39$  Atrial Fibrillation [AF]) and 29 healthy controls; the 3 T population had 86 patients ( $n = 22$  known CAD,  $n = 23$  Type II Diabetes Mellitus [DM],  $n = 21$  Severe Aortic Stenosis [AS],  $n = 20$  Hypertrophic Cardiomyopathy [HCM]) and 22 healthy controls. Healthy controls had no history of cardiovascular disease, were not on regular medications, and had normal electrocardiograms.

### CMR protocol

All subjects avoided adenosine antagonizers (e.g. caffeine) for  $\geq 24$  h before CMR. T1-mapping was performed using the Shortened Modified Look-Locker Inversion recovery (ShMOLLI) prototype sequence (WIP 561 and 448C) with inline map generation, which uses 9-heartbeats breath-holds per T1-map acquisition and enables on-screen image reconstruction within 10 s [14].

Native T1-maps were acquired at rest and during peak adenosine stress (140  $\mu\text{g/kg/min}$ , 4 min, IV) in short-axis (basal, mid-ventricular, apical) slices, followed immediately

by first-pass perfusion imaging on matching slices during peak stress, with an IV bolus of GBCA (0.03 mmol/kg at 6 ml/s; Dotarem, Guerbet, Villepinte, France) and saline flush (15 ml at 6 ml/s) [15, 16]. Matching rest perfusion images were acquired >15 min after stress perfusion and adenosine discontinuation to allow sufficient time for contrast washout [15, 16].

### T1-mapping analysis

Separate data files containing all T1-maps were created and anonymized before analysis by an observer (>3 years of T1-mapping analysis experience) blinded to perfusion images and clinical information. T1-maps were excluded from analysis if the spleen was not clearly visible (2%), had respiratory-motion artefacts on raw Inversion-Recovery-weighted images (3%) or had suboptimal goodness-of-fit  $R^2$ -maps (2%) [17, 20]. Overall, 738 T1-maps were included in final analysis, using dedicated in-house software MC-ROI (programmed by S.K.P. in IDL, version 6.1, Exelis Visual Information Solutions, Boulder, Colorado) [14–18, 20]. To estimate mean native  $T1_{\text{spleen}}$ , regions of interest (ROIs) were manually placed on T1-maps to include as much splenic tissue as possible, avoiding partial volume effects from large splenic blood vessels and borders with neighbouring tissues (Fig. 1). ROIs

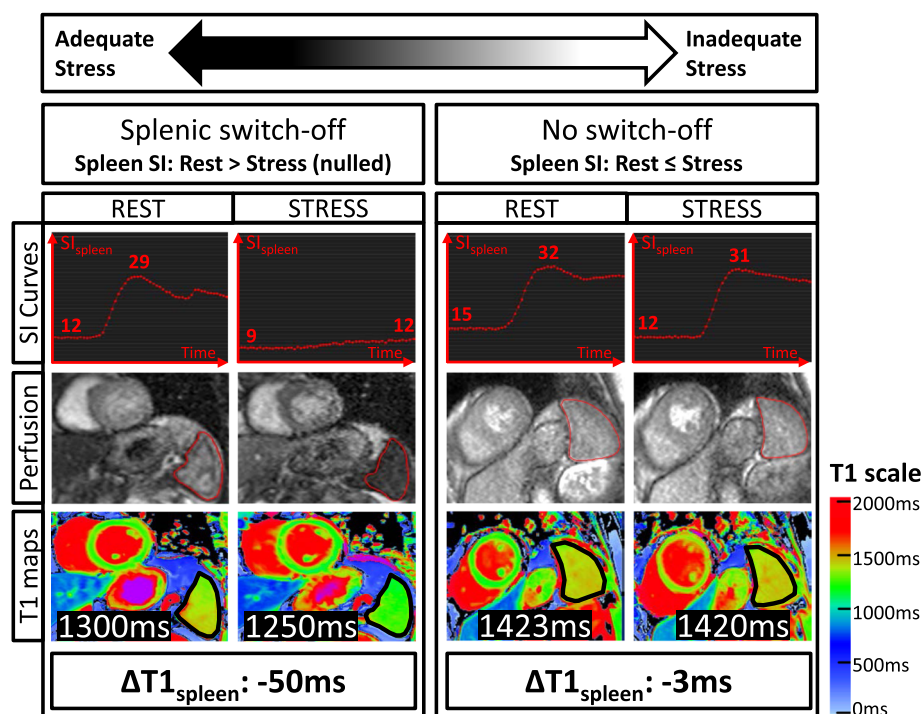
were quality checked against corresponding Inversion-Recovery-weighted images and  $R^2$ -maps. To derive thresholds suitable for direct application on the CMR console, splenic T1-reactivity to adenosine stress ( $\Delta T1_{\text{spleen}}$ ) was expressed in absolute terms:  $\Delta T1_{\text{spleen}}$  (ms) =  $\text{Stress}T1_{\text{spleen}} - \text{Rest}T1_{\text{spleen}}$ .

### T1<sub>spleen</sub> intra-scan variability assessments

Inter-slice variability in resting  $T1_{\text{spleen}}$ , stress  $T1_{\text{spleen}}$  and  $\Delta T1_{\text{spleen}}$  were assessed in cases where matching stress and rest T1-maps were performed in  $\geq 2$  different short-axis slice positions. To assess for intra-slice  $T1_{\text{spleen}}$  variability, we re-analyzed healthy-volunteers data from the original ShMOLLI methods paper, where T1-maps were repeated >15 min apart in the same short-axis slice within the same scan [14]. Intra-scan variability was calculated as the standard deviation of differences-from-the-mean in each individual.

### Splenic perfusion analysis

Splenic first-pass perfusion was analysed by an observer (>4 years of perfusion imaging analysis experience) blinded to T1-maps and clinical information, using CMR<sup>42</sup> software (Circle Cardiovascular Imaging Inc., Calgary, Canada). Splenic ROIs were placed on stress and rest perfusion images with frame-by-frame manual



**Fig. 1** Representative stress and rest splenic first-pass gadolinium perfusion and native T1-maps. Signal intensity (SI) curves represent splenic perfusion SI (y-axis, arbitrary units) over time (x-axis, 50–60 s). The maximum and minimum  $SI_{\text{spleen}}$  are as indicated. Splenic regions of interests on perfusion images and T1-maps are outlined in red and black, respectively. Mean native  $T1_{\text{spleen}}$  and stress changes ( $\Delta T1_{\text{spleen}}$ ) are as labelled. 3 T images were used for illustration (observed  $\Delta T1_{\text{spleen}}$  and  $\Delta SI_{\text{spleen}}$  are field strength independent)

correction for artefacts and respiratory motion, to generate curves showing mean splenic signal intensity (SI, arbitrary units) changes over time (50–60s). Peak splenic perfusion SI ( $SI_{\text{spleen}}$ ) was estimated as the numerical difference between baseline-SI and maximal-SI during splenic first-pass perfusion as previously described [6]. Adenosine-induced changes in  $SI_{\text{spleen}}$  compared to rest were expressed in percentages:  $\Delta SI_{\text{spleen}} (\%) = (StressSI_{\text{spleen}} - RestSI_{\text{spleen}}) \div RestSI_{\text{spleen}} \times 100\%$ .

Splenic switch-off on perfusion imaging was visually assessed by 2 independent observers (>3 years clinical CMR perfusion experience). In the 5/212 cases where the 2 observers disagreed, adjudication was sought from a 3<sup>rd</sup> independent observer (Fig. 1). Perfusion images were graded as previously described [6]: either displaying *splenic switch-off* (the spleen on rest imaging is clearly brighter than on stress imaging), or *no switch-off* (the spleen on rest imaging is of similar brightness compared to stress imaging).

### Statistical analysis

Data are reported as mean  $\pm$  SD, tests are 2-tailed and parametric, based on Kolmogorov-Smirnov normality-checks. Differences in individual characteristics were tested using Student's *t*-tests, paired within individuals (e.g. stress vs rest  $T1_{\text{spleen}}$ ) and unpaired between groups (e.g.  $\Delta T1_{\text{spleen}}$  in controls vs patients). Comparisons between  $\geq 3$  data groups were assessed using analysis of variance (ANOVA) with Bonferroni-corrected post-hoc method. Linear correlations were assessed using Pearson's correlation coefficient (*R*) and non-linear correlations were assessed using Spearman's rank correlation coefficient ( $\rho$ ). Intra-scan variability and inter-observer reproducibility of rest/stress  $T1_{\text{spleen}}$  and  $\Delta T1_{\text{spleen}}$  were assessed using the Intra-class correlation coefficient (ICC), reporting 95% confidence intervals. The performance of  $\Delta T1_{\text{spleen}}$  for replicating splenic switch-off was assessed using receiver-operating characteristics (ROC) curves [21], reporting area-under-the-curve (AUC  $\pm$  SEM), and also sensitivity, specificity, diagnostic accuracy, positive predictive values (PPV) and negative predictive values (NPV), with 95% confidence intervals (CI). All analyses were performed on single measures per-subject, using MedCalc 12.7.8 (MedCalc Software, Ostend, Belgium).  $P < 0.05$  denotes statistical significance.

## Results

### Subject characteristics

Subject characteristics are summarised in Table 1. All subjects experienced at least one adenosine-related symptoms (e.g. chest-tightness, dyspnoea, flushing) [13], and >10 bpm increase in heart rate (HR) during

adenosine stress, compared to rest. Significant blood pressure response (>10 mmHg SBP decrease during stress) was observed in 50% of subjects.

Mean stress HR was lower in 1.5 T patients compared to other subjects, despite similar resting HR, likely due to more frequent beta-blocker and non-dihydropyridine calcium channel antagonist administration in these patients (all AF/CAD, Table 1).

### Stress and rest $T1_{\text{spleen}}$ in controls and patients

In healthy controls, mean resting  $T1_{\text{spleen}}$  values were  $1102 \pm 66$  ms (1.5 T) and  $1352 \pm 114$  ms (3 T), which decreased significantly during adenosine stress at 1.5 T ( $\Delta T1_{\text{spleen}}$ :  $-40 \pm 25$  ms,  $p < 0.0001$ ) and 3 T ( $\Delta T1_{\text{spleen}}$ :  $-43 \pm 31$  ms,  $p < 0.0001$ ). Patients had slightly lower resting  $T1_{\text{spleen}}$  compared to controls at 1.5 T ( $1083 \pm 59$  ms vs.  $1102 \pm 66$  ms,  $p = 0.04$ ), and this pattern was more pronounced at 3 T ( $1295 \pm 105$  ms vs.  $1352 \pm 114$  ms,  $p = 0.01$ ). Despite these observed resting  $T1_{\text{spleen}}$  differences,  $\Delta T1_{\text{spleen}}$  was comparable between patients and controls, at 1.5 T ( $-44 \pm 21$  ms vs.  $-40 \pm 25$  ms,  $p = 0.43$ ) and 3 T ( $-44 \pm 26$  ms vs.  $-43 \pm 31$  ms,  $p = 0.93$ ; Table 2). In controls, there was a strong correlation between stress  $\Delta T1_{\text{spleen}}$  (mean  $-4.1 \pm 1.5\%$ ) and  $\Delta T1_{\text{myocardium}}$  (mean  $5.9 \pm 1.8\%$ ),  $r = -0.72$ ,  $p < 0.001$ . See Additional file 1 for more details.

In pooled analysis,  $\Delta T1_{\text{spleen}}$  did not appear to be significantly affected by field strength (1.5 T vs. 3 T:  $-43 \pm 22$  ms vs.  $-42 \pm 27$  ms,  $p = 0.89$ ), gender (male vs. female:  $-40 \pm 23$  ms vs.  $-47 \pm 28$  ms,  $p = 0.09$ ), age ( $R = 0.10$ ,  $p = 0.14$ , range 21–89 years) or the type of cardiovascular diseases (1.5 T CAD  $-42 \pm 20$  ms vs. 3 T CAD  $-40 \pm 25$  ms vs. AF  $-46 \pm 22$  ms vs. HCM  $-43 \pm 28$  ms vs. AS  $-43 \pm 21$  ms vs. DM  $-43 \pm 32$  ms,  $p = 0.54$ ). In addition,  $\Delta T1_{\text{spleen}}$  was not significantly affected by medication in patients (supplementary material in Additional file 2).

### $T1_{\text{spleen}}$ intra-scan variability

Inter-slice intra-scan variability (assessable in 96 subjects) was within  $\pm 19$  ms for resting  $T1_{\text{spleen}}$ ,  $\pm 18$  ms for stress  $T1_{\text{spleen}}$ , and  $\pm 10$  ms for  $\Delta T1_{\text{spleen}}$ . Re-analysis of the original ShMOLLI cohort (spleen visible in 9/10 cases), revealed an intra-slice intra-scan repeat variability of  $T1_{\text{spleen}}$  of  $\pm 9$  ms, ICC: 0.98 (95% confidence interval 0.93 to 0.99) [14].  $\Delta T1_{\text{spleen}}$  was derived by a second independent blinded observer in 45 subjects (20 controls; 25 patients: 5 CAD, 5 AF, 5 DM, 5 AS, 5 HCM), which yielded an ICC of 0.87 (95% confidence interval: 0.76 to 0.93). The Bland-Altman plot for inter-observer variability is shown in supplementary material (Additional file 3).

### Associations between splenic perfusion, T1 and rate pressure product (RPP)

By semi-quantitative analysis, peak splenic perfusion SI ( $SI_{\text{spleen}}$ ) decreased significantly with adenosine

**Table 1** Characteristics of study subjects: healthy controls and patients ( $n = 212$ )

	1.5 T Controls $n = 29$	1.5 T Patients $n = 75$	3 T Controls $n = 22$	3 T Patients $n = 86$
Age (years)	54 ± 17	65 ± 9*	43 ± 12	60 ± 14*
Men (%)	21 (72)	58 (77)	13 (59)	58 (67)
Body mass index (kg/m <sup>2</sup> )	25 ± 4	28 ± 5	26 ± 3	28 ± 4
Hematocrit	0.42 ± 0.03	0.43 ± 0.03	0.42 ± 0.04	0.42 ± 0.03
CMR hemodynamic data				
Rest HR (bpm)	66 ± 11	62 ± 15	62 ± 12	65 ± 10
Stress HR (bpm)	96 ± 15	79 ± 15*	95 ± 12	91 ± 14
Rest SBP (mmHg)	133 ± 21	139 ± 19	127 ± 14	136 ± 19
Stress SBP (mmHg)	127 ± 19	133 ± 19	122 ± 16	126 ± 19
Rest RPP (bpm.mmHg)	8800 ± 2200	8600 ± 2200	7600 ± 1700	8700 ± 2000
Stress RPP (bpm.mmHg)	12,200 ± 2600	10,500 ± 2500*	12,000 ± 2200	11,700 ± 2700
Adenosine symptoms	29 (100)	75 (100)	22 (100)	86 (100)
Co-morbidities				
Current smoker	3 (10)	2 (3)	2 (9)	12 (14)
Ex-smoker	3 (10)	21 (28)	3 (14)	21 (24)
Hypertension	-	28 (37)	-	24 (28)
Hyperlipidemia	-	23 (31)	-	23 (27)
Stroke/TIA	-	2 (3)	-	2 (2)
Medications				
Aspirin	-	36 (48)	-	35 (41)
Beta-blocker	-	34 (45) <sup>#</sup>	-	19 (22)
ACEi/ARB	-	36 (48)	-	39 (45)
Statin	-	31 (41) <sup>#</sup>	-	44 (51)
Nitrates	-	3 (4)	-	4 (5)
CCB (non-DHP)	-	13 (17) <sup>#</sup>	-	0 (0)
CCB (DHP)	-	4 (5)	-	7 (8)

Values are  $n$  (%) or mean ± SD

Abbreviations: RPP rate pressure product, TIA transient ischemic attack, ACEi angiotensin-converting enzyme inhibitors, ARB angiotensin receptor blockers, CCB calcium channel antagonist, DHP dihydropyridine

\* $p < 0.05$  compared to controls of corresponding field strength (1.5 T or 3 T). <sup>#</sup> $p < 0.05$  for comparisons between patient groups (1.5 T vs 3 T)

stress compared to rest, with no differences between controls and patients, or across field strengths (Table 3).  $\Delta SI_{\text{spleen}}$  correlated strongly with  $\Delta T1_{\text{spleen}}$  ( $\rho = 0.70$ ,  $p < 0.0001$ , Fig. 2). In contrast,  $\Delta SI_{\text{spleen}}$  and  $\Delta T1_{\text{spleen}}$  did not demonstrate significant correlations with stress-induced changes in RPP ( $R = 0.04$ ,  $p = 0.60$ ;  $R = 0.06$ ,  $p = 0.38$ , respectively).

#### Visual splenic switch-off assessment – relationships with perfusion, quantitative $T1_{\text{spleen}}$ and hemodynamic parameters

Subjects with visual splenic switch-off had greater stress  $\Delta SI_{\text{spleen}}$  and  $\Delta T1_{\text{spleen}}$  values compared to those with no switch-off (Table 4 and Fig. 3). In contrast, there were no significant differences in stress-related

**Table 2** Rest and stress splenic T1 in healthy controls and patients

	1.5 T Controls	1.5 T Patients	$p$ -value	3 T Controls	3 T Patients	$p$ -value
Rest $T1_{\text{spleen}}$ (ms)	1102 ± 66	1083 ± 59	0.04	1352 ± 114	1295 ± 105	0.01
Stress $T1_{\text{spleen}}$ (ms)	1061 ± 68	1039 ± 55	0.02	1308 ± 114	1253 ± 112	0.01
$\Delta T1_{\text{spleen}}$ (ms)	-40 ± 25	-44 ± 21	0.43	-43 ± 31	-44 ± 26	0.93

$\Delta T1_{\text{spleen}} = \text{Stress}T1_{\text{spleen}} - \text{Rest}T1_{\text{spleen}}$

**Table 3** Peak splenic perfusion signal intensity ( $SI_{\text{spleen}}$ ) at rest and during adenosine stress ( $\Delta SI_{\text{spleen}}$ ) in healthy controls and patients

	1.5 T Controls	1.5 T Patients	3 T Controls	3 T Patients	<i>p</i> -value
Rest $SI_{\text{spleen}}$ (au)	26 ± 8	24 ± 11	29 ± 13	27 ± 13	0.16
Stress $SI_{\text{spleen}}$ (au)	11 ± 5	11 ± 5	13 ± 12	12 ± 8	0.10
$\Delta SI_{\text{spleen}}$ (%)	-58 ± 23	-54 ± 22	-56 ± 28	-52 ± 30	0.51

Abbreviations: Au arbitrary units

*P*-values derived using ANOVA with Bonferroni post-hoc method

haemodynamic changes (HR, SBP, RPP) between subjects with splenic switch-off and no switch-off (Table 4 and Fig. 3).

#### ROC analysis of native $\Delta T1_{\text{spleen}}$ for replicating gadolinium-based “splenic switch-off”

ROC analysis using visual splenic switch-off as reference standard (true positives = splenic switch-off, true negatives = no switch-off) yielded an AUC of  $0.90 \pm 0.05$  ( $p < 0.0001$ , Fig. 4). A  $\Delta T1_{\text{spleen}}$  threshold of  $\geq -30$  ms during adenosine stress replicated visual splenic switch-off with a sensitivity of 90% (95% CI: 85–94%,  $p < 0.0001$ ), specificity 88% (95% CI: 62–98%,  $p < 0.0001$ ), diagnostic accuracy 90% (95% CI: 84–96%,  $p < 0.0001$ ), PPV 98% (95% CI: 96–100%,  $p < 0.0001$ ) and NPV 42% (95% CI: 26–61%,  $p < 0.0001$ ).

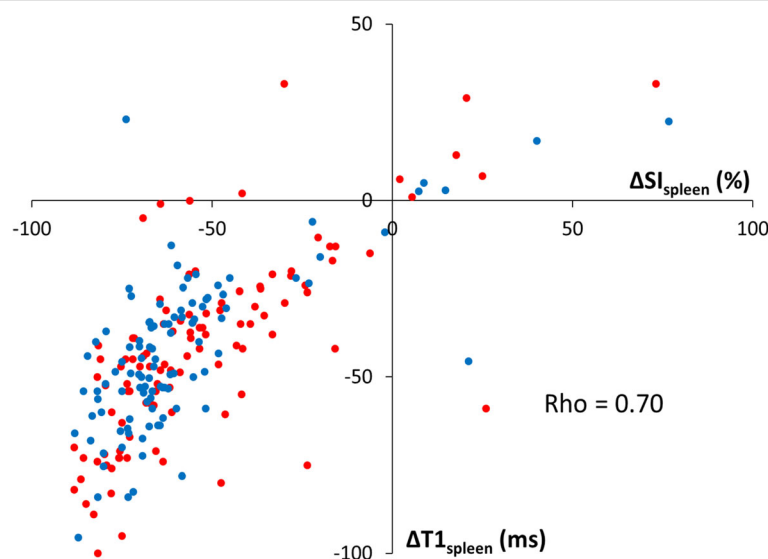
#### Discussion

This proof of principle study demonstrated that  $T1_{\text{spleen}}$  decreases significantly during adenosine stress compared

to baseline. The magnitude of the stress-induced  $T1_{\text{spleen}}$  response ( $\Delta T1_{\text{spleen}}$ ) is strongly correlated with splenic perfusion attenuation ( $\Delta SI_{\text{spleen}}$ ). From a clinical viewpoint, a native  $\Delta T1_{\text{spleen}}$  threshold of  $\geq -30$  ms accurately replicated the “splenic switch-off” sign with a high positive predictive value of 98% and offers the potential to assess adenosine stress adequacy before GBCA first-pass perfusion imaging. From a practical viewpoint, assessment of  $T1_{\text{spleen}}$  takes  $\sim 30$  s (Fig. 5), which means it can be repeated as necessary “on-the-fly”, to guide adenosine dosage up-titrations and optimize stress responses before injection of contrast agents (example protocol in Fig. 5). This pre-gadolinium approach may be advantageous over the retrospective and potentially gadolinium dose-sensitive splenic switch-off method for improving the quality of stress responses before first-pass perfusion imaging, which deserves further investigation in future studies to determine whether it decreases the number of false negative perfusion scans [6].

#### Stress/rest $T1_{\text{spleen}}$ as a marker of adenosine stress response

Patients had lower resting native  $T1_{\text{spleen}}$  values compared to controls. This may be related to the presence of co-morbidities in patients, such as hypertension and peripheral vascular disease, which may induce peripheral vasoconstriction, with expected reductions in resting organ blood volumes and  $T1_{\text{spleen}}$  values. This observation deserves further investigation in larger future studies. Native  $T1$ -relaxation times of tissues are prolonged by increased blood volume (i.e. water content) [14, 15, 22]. Adenosine causes splenic artery vasoconstriction and



**Fig. 2** Correlation between stress-induced reductions in peak splenic signal intensity ( $\Delta SI_{\text{spleen}}$ ) and splenic native  $T1$  ( $\Delta T1_{\text{spleen}}$ ). Pooled data of controls and patients at 1.5 T (blue) and 3 T (red), represented on per-subject basis ( $n = 212$ ). Spearman's rank correlation coefficient ( $Rho = 0.70$ ,  $p < 0.0001$ )

**Table 4** Stress-induced changes in peak splenic perfusion signal intensity ( $\Delta SI_{\text{spleen}}$ ), T1 ( $\Delta T1_{\text{spleen}}$ ) and hemodynamic parameters for visually assessed “splenic switch-off” sign

	Splenic Switch-off	No Switch-off	p-value
All subjects $n = 212$	196 (92)	16 (8)	-
Healthy volunteers $n = 51$	49 (96)	2 (4)	-
Patients $n = 161$	147 (91)	14 (9)	-
$\Delta SI_{\text{spleen}}$ (%)	$-62 \pm 17$	$17 \pm 29$	$<0.0001$
$\Delta T1_{\text{spleen}}$ (ms)	$-46 \pm 22$	$-2 \pm 25$	$<0.0001$
Stress hemodynamic changes			
$\Delta$ heart rate (bpm)	$19 \pm 9$	$20 \pm 12$	0.83
$\Delta$ SBP (mm. Hg)	$-8 \pm 22$	$-10 \pm 19$	0.76
$\Delta$ RPP (bpm.mmHg)	$2800 \pm 2100$	$2600 \pm 1700$	0.89

Values are  $n$  (%) or mean  $\pm$  SD

Abbreviations: *Bpm* beats per minute, *SBP* systolic blood pressure, *RPP* rate pressure product

reduced blood volume [6–11], which shortens splenic T1-relaxation times. This is supported by our observation of significantly lower  $T1_{\text{spleen}}$  during adenosine stress compared to rest, in both controls and patients. The stress  $\Delta T1_{\text{spleen}}$  was not significantly affected by different field strengths, age, gender and cardiovascular diseases, likely reflecting reproducible T1-estimations in this study [14, 15, 22].

The correlation between stress  $\Delta T1_{\text{spleen}}$  and  $\Delta T1_{\text{myocardium}}$  in normal controls suggests the vasoconstrictor effect of adenosine on the spleen is associated with vasodilatory effects in the myocardium. For the relationship between myocardial and splenic stress T1 in patients with cardiovascular disease, larger ongoing studies will offer reference ranges for  $\Delta T1$  in disease, and resolve

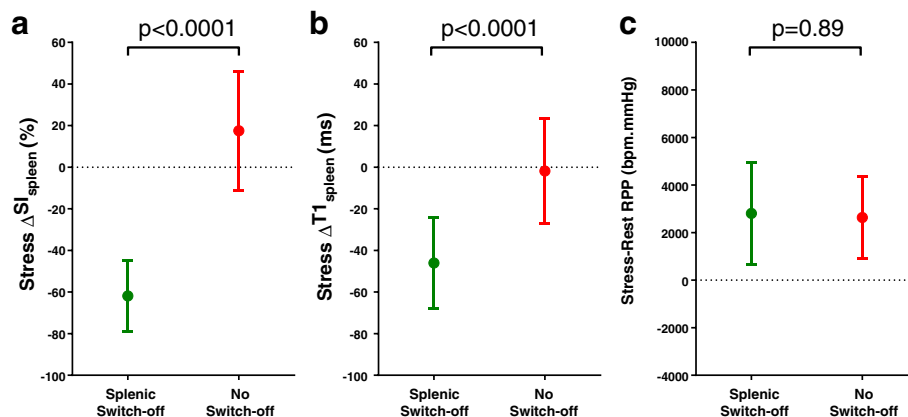
the separate effects of regional myocardial differences and medication on stress T1 reactivity.

The observed strong correlation between  $\Delta T1_{\text{spleen}}$  and  $\Delta SI_{\text{spleen}}$  suggests that stress-induced changes in splenic blood volume are related to blood flow, which is regulated by alterations in the adenosine-mediated splenic arterial tone [10, 11]. The lack of significant correlation between  $\Delta SI_{\text{spleen}}$  or  $\Delta T1_{\text{spleen}}$  with rate pressure product is consistent with existing evidence showing dissociation between imaging and hemodynamic markers of stress response [5, 6], and further suggests that stress responses during clinical CMR cannot be reliably assessed using hemodynamic observations alone [5]. This deserves further investigation.

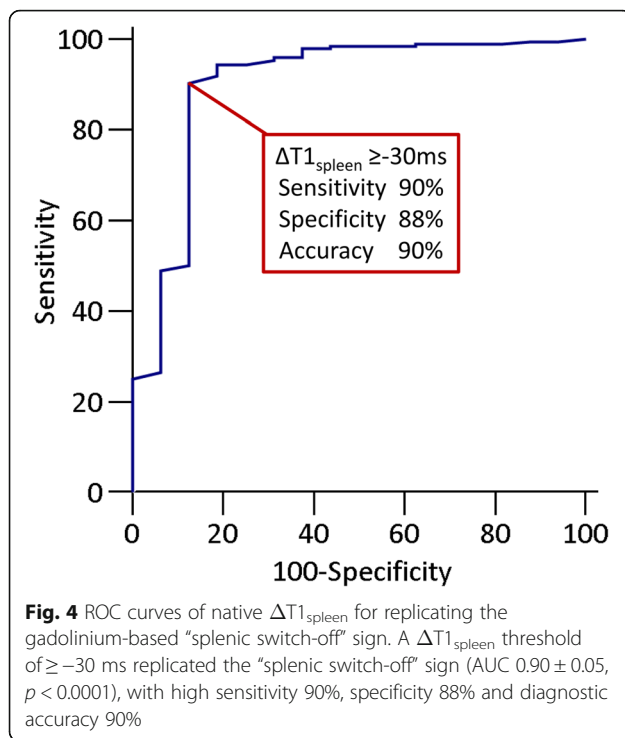
A threshold of  $\geq 30$  ms decrease in  $T1_{\text{spleen}}$  replicated complete splenic switch-off with a high positive predictive value of 98%. The intra-scan variability in  $T1_{\text{spleen}}$  (inter-slice:  $\pm 10$  ms; intra-slice:  $\pm 9$  ms) was 3-times less than this proposed threshold  $\geq 30$  ms drop, with excellent T1-fit as evident on quality control  $R^2$ -maps, despite the lack of dedicated image optimization (e.g. shimming) over the spleen. For stress  $T1_{\text{spleen}}$  responses  $< 30$  ms, further work is needed to determine whether adenosine dose-increments or waiting longer with the same infusion rate may improve the confidence of stress responses, and impact on diagnostic performance of stress CMR for the diagnosis of ischemia.

#### Limitations and future directions

This proof-of-concept study is based on ShMOLLI  $T1_{\text{spleen}}$  values derived from short-axis slices planned for myocardial perfusion CMR imaging; the spleen was not visible in a small proportion of T1-maps ( $\sim 2\%$ ), and future applications of splenic T1-mapping may benefit



**Fig. 3** Relations between different markers of stress adequacy. Subjects with the “splenic switch-off” sign had greater stress-induced reductions in **a** gadolinium-based splenic perfusion ( $\Delta SI_{\text{spleen}}$ , same technique) and **b** gadolinium-free splenic T1 ( $\Delta T1_{\text{spleen}}$ , different technique) compared to subjects with no switch-off. There was no difference in stress-induced **c** hemodynamic changes in rate pressure product (RPP) between the splenic switch-off and the no switch-off subjects. Data are mean  $\pm$  1SD

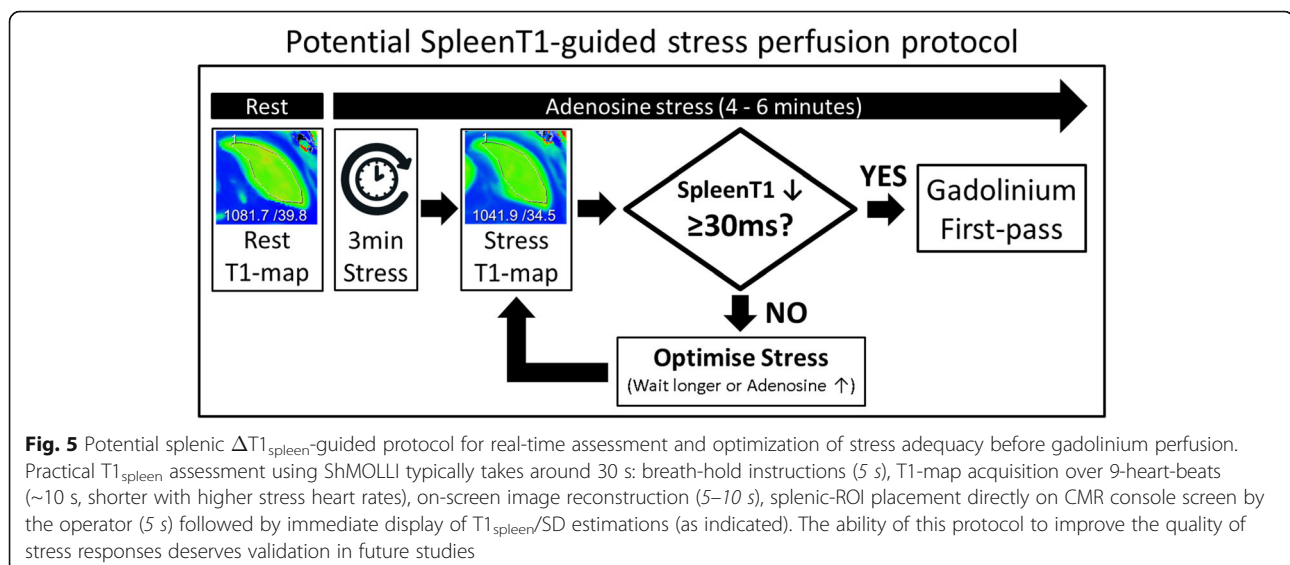


from a dedicated image planned through the spleen. Rapid on-scanner T1-map reconstructions, with the immediate availability of goodness-of-fit measures (such as  $R^2$ -maps), are imperative to enable practical “on-the-fly” repetition of reliable  $T1_{\text{spleen}}$  estimations to guide stress response optimization (Fig. 5). Given the overall excellent  $R^2$ -maps over the spleen and the narrow  $T1_{\text{spleen}}$  ranges obtained, data in this study suggest that stress/rest splenic T1-mapping can be feasibly included in CMR protocols without major technical adjustments. Practical in-vivo T1-estimations are method-dependent, and demonstrate

increasingly discrepant heart rate dependencies at longer T1-values [23]. Therefore, results achieved with ShMOLLI, in particular the splenic T1-thresholds replicating splenic switch-off, should be interpreted with care before directly translating to other T1-mapping techniques. Choosing methods that can withstand dynamic HR-variations and tachycardia without significant HR-dependencies is therefore paramount when performing stress-T1 studies. The gadolinium-based splenic switch-off sign is only seen with non-selective adenosine receptor agonists (dipyridamole and adenosine), but was absent with cardio-selective vasodilators (e.g. regadenoson) or inotropic agents (e.g. dobutamine) [6]. Further work is needed to elucidate stress  $T1_{\text{spleen}}$  responses using pharmacological agents other than adenosine and during physical exercise. Patients in this study were unselected for diseases known to affect splenic blood volumes, e.g. venous portal hypertension, hematological malignancies and systemic inflammation; thus, further studies to characterize the effects of these diseases on  $T1_{\text{spleen}}$  will help to determine the general applicability of this technique. While we identified a cut-off of  $\geq 30$  ms drop in  $T1_{\text{spleen}}$  during stress for replicating complete splenic switch-off, the clinical utility of this threshold for detecting *true* stress adequacy needs to be validated against false-negative perfusion scans, determined by comparison to invasive coronary angiography and pressure-wire based assessments of functional ischemia, such as fractional flow reserve. This is topic of ongoing work.

## Conclusions

Adenosine stress and rest splenic T1-mapping is a novel method for assessing stress responses, independent of conventional hemodynamic parameters. It



enables prediction of the visual “splenic switch-off” sign without the need for gadolinium, and correlates well to changes in splenic signal intensity during stress/rest perfusion imaging.  $\Delta T1_{\text{spleen}}$  holds promise to facilitate optimization of stress responses before gadolinium first-pass perfusion CMR.

## Additional files

**Additional file 1: Figure S1.** Description of data: Correlation between adenosine stress  $\Delta T1_{\text{spleen}}$  and  $\Delta T1_{\text{myocardium}}$  in 51 healthy controls. Data are presented per-subject. (DOCX 26 kb)

**Additional file 2: Table S1.** Description of data: Effect of medication on  $\Delta T1_{\text{spleen}}$  in patients with cardiovascular disease. ACE: angiotensin converting enzyme; ARB: angiotensin receptor blocker; CCB: calcium channel blockers; DHP: dihydropyridine. (DOCX 13 kb)

**Additional file 3: Figure S2.** Description of data: Bland Altman plot of  $\Delta T1_{\text{spleen}}$  estimation by 2 independent blinded observers. (DOCX 38 kb)

## Abbreviations

CAD: Coronary artery disease; CMR: Cardiovascular magnetic resonance; ECG: Electrocardiogram; GBCA: Gadolinium-based contrast agents; ROC: Receiver-operating characteristics; ShMOLLI: Shortened modified look-locker inversion recovery;  $SI_{\text{spleen}}$ : Peak splenic signal intensity;  $T1_{\text{spleen}}$ : Mean native splenic T1 values

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## Availability of data and materials

The datasets during and/or analysed during the current study available from the corresponding author on reasonable request.

## Authors' contributions

All authors made appropriate contributions according to the ICMJE guidance, and as such have read and approved the final manuscript. All authors take public responsibility for appropriate portions of the manuscript content; and agree to be accountable in ensuring that questions related to the accuracy or integrity of the work are appropriately investigated and resolved. In addition, author specific contributions to the study are listed below: AL contributed to study conception and design, subject recruitment, data acquisition, analysis and interpretation; drafting of manuscript and revisions. RSW contributed to subject recruitment, data acquisition, analysis, interpretation and manuscript revisions. RA, MM, EL contributed to subject recruitment, data acquisition and manuscript revisions. AG contributed to giving technical advice on T1-mapping and manuscript revisions. MP, GK, JCF, RKK, KMC contributed to subject recruitment and manuscript revisions. SN contributed to study conception and design, data interpretation, study supervision and manuscript revisions. SKP contributed to study conception and design, data analysis, interpretation, study supervision and manuscript revisions. VMF contributed to study conception and design, subject recruitment, data acquisition, analysis, interpretation, study supervision and manuscript revisions.

## Competing interests

SKP has patent authorship rights for U.S. patent 9285446 B2. Systems and methods for shortened look locker inversion recovery (Sh-MOLLI) cardiac gated mapping of T1. Granted March 15, 2016. All rights transferred to Siemens Medical.

All other authors have no relationships relevant to the contents of this paper to disclose.

## Consent for publication

All subjects gave written informed consent for publication.

## Ethics approval and consent to participate

This study was approved by the South Central Oxford A Health Research Authority (formerly known as Oxfordshire Research Ethics Committee A) based at Bristol HRA Centre, Level 3, Block B, Whitefriars, Lewins Mead, Bristol, BS1 2NT, UK. All subjects gave written informed consent.

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