



Quantification of Pulmonary Inflammation after Segmental Allergen Challenge Using Turbo-Inversion Recovery-Magnitude Magnetic Resonance Imaging

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Abstract

Rationale: There is a need to develop novel noninvasive imaging biomarkers that help to evaluate antiinflammatory asthma treatments.

Objectives: To investigate whether the extent of the segmental lung edema measured noninvasively using turbo-inversion recovery-magnitude magnetic resonance imaging (TIRM MRI) corresponds to the severity of the regional allergic reaction determined by the percentage of eosinophils in bronchoalveolar lavage fluid (BAL) 24 hours after segmental allergen challenge in patients with asthma compared with normal control subjects.

Methods: Eleven volunteers with allergic asthma and five healthy volunteers underwent segmental challenges with different allergen doses by two bronchoscopies 24 hours apart. They had lung MRI at baseline and 6 and 24 hours after segmental challenge. MRI TIRM scores were correlated with the eosinophilic response at 24 hours.

Measurements and Main Results: In patients with asthma, there were significant differences of eosinophil percentages in BAL at 24 hours from segments given standard-dose, low-dose, or no allergen (saline) ($P < 0.001$). Correspondingly significant differences between the TIRM score in allergen standard-dose, low-dose, and saline-treated segments were observed at 24 hours post-challenge ($P < 0.001$). With increasing TIRM score at 24 hours the percent eosinophils per segment 24 hours post-challenge also increased accordingly ($P < 0.001$). There was interobserver agreement for TIRM score grading ($\kappa = 0.72$ for 24-h time point).

Conclusions: The MRI-based noninvasive TIRM score is a promising biomarker for the noninvasive detection of the inflammatory response after segmental allergen challenge in patients with asthma and may serve to monitor the therapeutic effectiveness of novel antiinflammatory drugs in future human trials.

Keywords: magnetic resonance imaging; asthma; segmental bronchial allergen challenge; bronchoscopy

At a Glance Commentary

Scientific Knowledge on the Subject: The extent of the segmental lung infiltrate measured noninvasively using magnetic resonance imaging–derived turbo-inversion recovery-magnitude score corresponds to the severity of the regional allergic reaction determined by the percentage of eosinophils in bronchoalveolar lavage fluid 24 hours after allergen challenge in patients with asthma and normal control subjects.

What This Study Adds to the Field: The presented magnetic resonance imaging–based noninvasive turbo-inversion recovery-magnitude score is a promising biomarker for the noninvasive detection of the inflammatory response after segmental allergen challenge in patients with asthma and may serve to monitor the therapeutic effectiveness of novel antiinflammatory drugs in future human trials.

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Asthma is estimated to affect more than 300 million patients worldwide with still significant mortality (1, 2). Although considerable progress has been made in the understanding of the pathogenesis of asthma (3), development of novel antiinflammatory treatments has been hampered by the seeming lack of effectiveness of investigational new drugs tested in human clinical trials that seemed promising in animal models (4, 5). Although the spirometric assessment of the late asthmatic response following inhaled allergen challenge has an accepted predictive value in proof-of-concept clinical trials (6), the value of inflammatory endpoints to predict clinical effectiveness of antiasthma drugs is less clear. Moreover, assessment of inflammatory endpoints from the lung compartment may necessitate invasive approaches, which limits repetitive measurements. Therefore, there is a clear need to develop novel noninvasive biomarkers in patients with asthma that help to evaluate antiinflammatory asthma treatments.

Segmental allergen challenge has been introduced in studies on the pathophysiology of asthma and for efficacy studies in the process of drug development (7–12). In response to a segmental challenge with allergen, airways in patients with asthma develop eosinophilic inflammation with localized edema and increased mucus production. The major advantage of this technique is the induction of a profound inflammatory response that is circumscribed and safe (13). However, the number of bronchoscopies to collect lavage fluid is limited, which hampers sequential assessments of the inflammatory response. Moreover, the bronchoscopy with lavage can induce an inflammatory response by itself reducing specificity of the method (14). Therefore, the development of less-invasive methods to repetitively assess the inflammatory response following allergen challenge is desirable.

Recently, functional pulmonary magnetic resonance imaging (MRI) has evolved into a promising noninvasive clinical tool to detect and quantify the regional inflammatory response (15–17). It has been used to monitor the inflammatory response to allergen challenge in animal models (18–22). However, up-to-date lung MRI in humans for quantification of inflammation has not been evaluated.

Table 1: Demographics

	Patients with Asthma	Control Group	P Value
N	11	5	
Number of excluded participants	0	1	
Reason for exclusion		Traffic accident, not related to the study	
Male/female	9/2	4/0	
Age, yr (median, quartile range)	41 (36–48)	44 (36–50)	0.48
IgE, IU/ml (median, quartile range)	173 (97–353)	23 (7.8–87.8)	0.03
PC ₂₀ , mg/ml (median, quartile range)	3.2 (1.1–13.4)	>16 for all control subjects	0.02

Definition of abbreviation: PC₂₀ = concentration at which methacholine causes a 20% decrease in FEV₁.

The two-dimensional short tau inversion recovery sequence has been shown to accurately detect lung fluid (23). Therefore, this study focuses on the evaluation of a turbo-inversion recovery-magnitude (TIRM) MRI sequence, which reconstructs the magnitude of the signal compared with short tau inversion

recovery, for visualization and quantification of bronchial inflammatory response following segmental allergen challenge (24). The hypothesis of this study was, that the extent of the segmental lung edema measured noninvasively using TIRM MRI corresponds to the severity of the regional allergic reaction determined by the

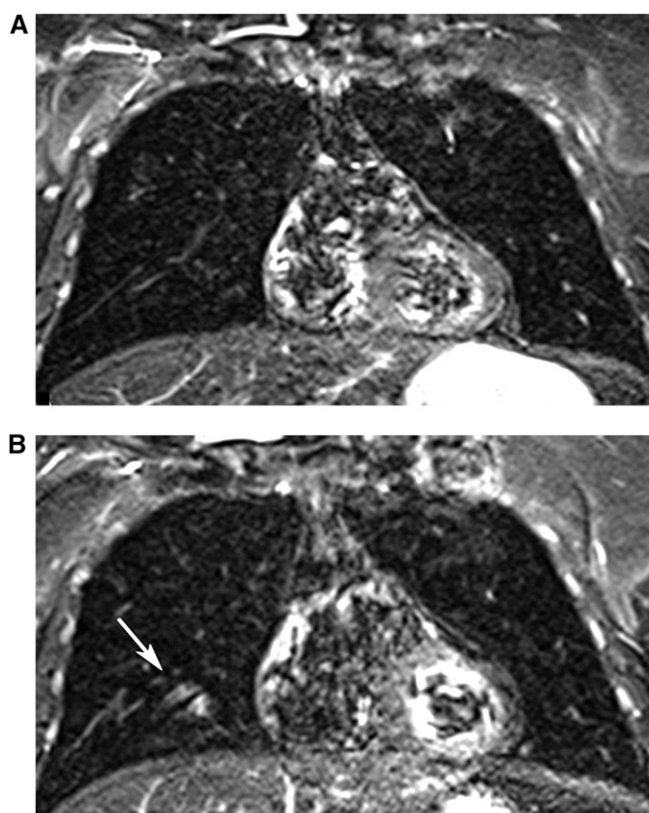


Figure 1. Coronal turbo-inversion recovery-magnitude magnetic resonance images of a patient with asthma at prechallenge (A) and 24 hours post-allergen challenge (B). At 24 hours, small peribronchial edema (arrow, turbo-inversion recovery-magnitude score 1) was noted in segment 4 of the right lung, which was not present before allergen challenge. The bronchoalveolar lavage at 24 hours in segment 4 of the right lung showed 2.25% eosinophils, whereas prechallenge eosinophil percentage was 0.5%.

number and percentage of eosinophils in the bronchoalveolar lavage fluid 24 hours after allergen challenge in patients with asthma compared with normal control subjects.

Some of the results of these studies have been previously reported in the form of an abstract (25).

Methods

Study Population

This prospective study was approved by the institutional ethics committee. Written informed consent was obtained from all volunteers. The study was conducted according to the principles of the Declaration of Helsinki.

Eleven volunteers with allergic asthma (nine male, age 41 [36–48]) and five healthy volunteers (four male, age 44 [36–50]) were included in this study (for subject demographics see Table 1). All participants were nonsmokers for at least 5 years and had an overall smoking history of less than 1 year.

Patients with asthma had a physician diagnosis of mild asthma according to the Global Initiative on Asthma guidelines (26) and a positive skin prick test as well as a positive response to the inhaled allergen (either grass mix or house dust mite). Bronchial responsiveness to methacholine was determined but not regarded as an inclusion criterion.

Healthy control subjects had no clinical evidence of allergy or respiratory disease. All revealed a negative skin prick test for both allergens and a negative response to an incremental methacholine challenge.

Upper or lower respiratory infection in the 4 weeks before enrollment into the study was regarded as an exclusion criterion. One healthy volunteer did not finish the protocol because of a car accident unrelated to the study.

Study Protocol

Subjects underwent two bronchoscopies 24 hours apart. The preparation, conduct, and observation of the bronchoscopy with segmental allergen challenge have been described in detail elsewhere (7). At first bronchoscopy, all subjects underwent a baseline bronchoalveolar lavage in the left lower lobe. Patients with asthma were instilled with the standard dose of allergen (SA) determined as described previously (27) in two segments of contralateral lungs (i.e., right middle lobe and left lingula, respectively) to study intraindividual

reproducibility. Moreover, another segment in the right upper lobe was instilled with one-tenth of the SA (low allergen dose, LA). Additionally, one segment in the middle lobe unaffected by allergen was instilled with 10 ml of normal saline (S) to control for specificity of the procedure.

Healthy subjects only had three instillations receiving only one instillation with SA because reproducibility was not a question. In contrast, one additional segment in the right middle lobe was lavaged with 150 ml of saline at the first bronchoscopy to increase the total number of recovered cells for separate *in vitro* experimentation.

After 24 hours, all instilled segments were lavaged and cells were counted and differentiated as described previously (7).

All subjects underwent three MRI scans. The first MRI scan was performed at baseline (0 h) on the day before first bronchoscopy. Subsequent MRI scans were performed at 6 and 24 hours after segmental challenge. The second bronchoscopy at 24 hours was completed within 1–2 hours after the final MRI scan. Pulmonary function was assessed before (0 and 24 h) and 4 hours (4 and 28 h) after each bronchoscopy by spirometry in the study center and two-hourly by

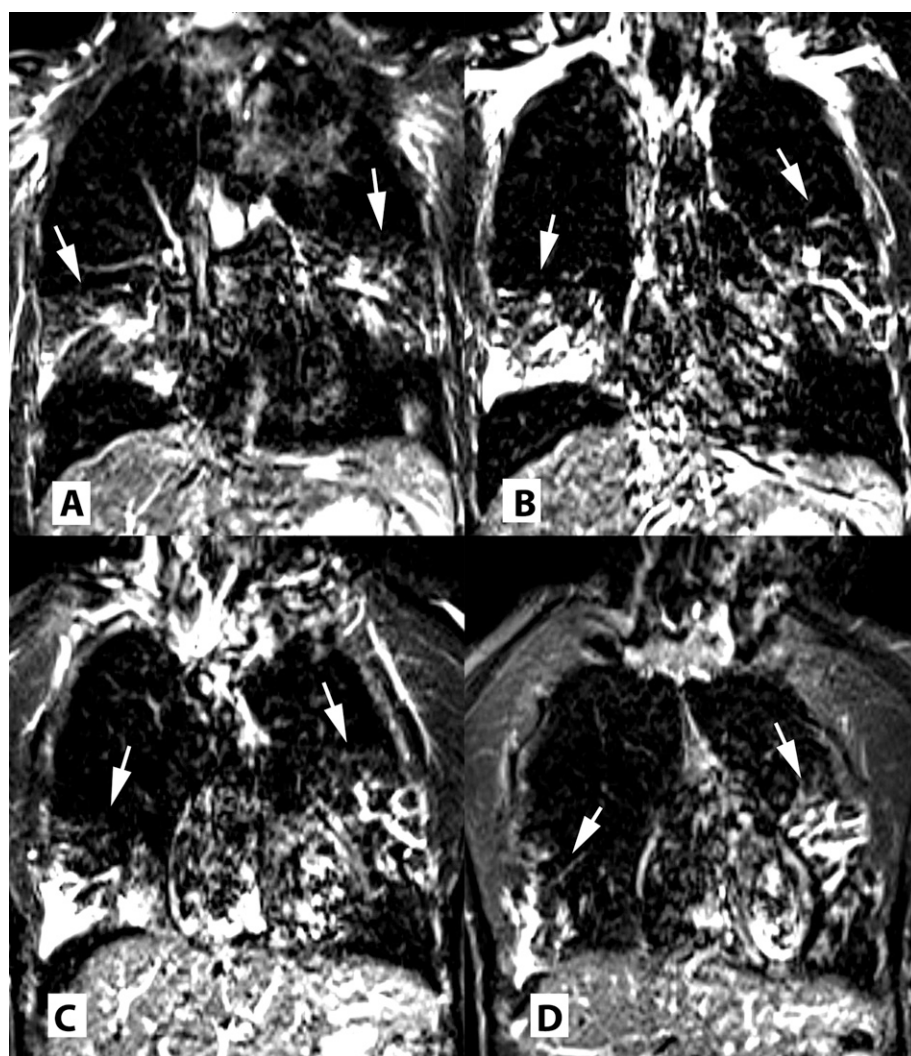


Figure 2. Representative coronal turbo-inversion recovery-magnitude (TIRM) images (A–D, dorsal to ventral) 24 hours post-allergen challenge. Large areas of increased signal corresponding to fluid and edema are noted in the challenged segments (arrows, segment 4 right lung with a TIRM score of 4 and segment 4 left lung with a TIRM score of 4). The extent of the affected area within a segment influenced the TIRM score and not signal intensity. The bronchoalveolar lavage at 24 hours in segment 4 of the right lung showed 81.5% eosinophils and in segment 4 of the left lung 78.8% eosinophils.

Table 2: Lung Function

	0 Hours (Baseline)		4 Hours		24 Hours		P Values	
	Median (25–75)	%	Median (25–75)	% of Baseline	Median (25–75)	% of Baseline	Absolute Kruskal-Wallis	Relative Kruskal-Wallis
Asthma (n = 11)								
FEV ₁ , L	3.8 (3.4–4.0)	100	3.4 (2.9–3.6)*	84.4 (82.8–92.5)*	3.3 (3.2–3.6)	90.2 (87.4–94.2)*	0.025	<0.0001
FEV ₁ , % predicted	93.4 (89.6–111)	100	85.8 (77.5–96)	84.5 (82.8–92.7)*	87.4 (79.9–98.4)	90.19 (87.3–94.4)*	0.08	<0.0001
FVC, L	4.9 (4.5–5.5)	100	4.6 (3.9–4.9)	90.7 (84.6–92.70)*	4.96 (4.3–5.2)	95.4 (93.8–100.2)*	0.24	0.001
FEV ₁ /FVC, %	74.7 (69.1–83.7)	100	73.8 (68.5–77.9)	96.8 (93.0–99.9)*	71.9 (65.6–79.5)	95.0 (92.1–96.6)*	0.59	0.002
Control (n = 4)								
FEV ₁ , L	4.6 (4.3–4.9)	100	4.2 (4.0–4.3)	91.5 (85.9–97.4)	4.4 (4.2–4.7)	96.5 (95.6–96.9)	0.55	0.11
FEV ₁ , % predicted	115.5 (108.0–122.8)	100	98.5 (94.6–105.1)	89.7 (80.9–97.4)	110.4 (105.6–113.3)	95 (92.3–97.1)	0.25	0.11
FVC, L	6.1 (5.9–6.4)	100	5.6 (5.2–5.9)	88.1 (85.4–91.9)*	6 (5.8–6.1)	97 (95.6–97.7)*	0.25	0.01
FEV ₁ /FVC, %	76.5 (74.2–77.3)	100	77.8 (75.8–79.4)	103 (100.8–105.2)	76.7 (73.7–78.0)	100.3 (99.3–100.8)	0.69	0.3

**P* < 0.05 compared with baseline, Wilcoxon test.

a handheld device until bedtime after discharge.

MRI

MRI was performed at 1.5 T (Avanto; Siemens Healthcare, Erlangen, Germany) using a TIRM sequence (TE/TR = 73/3,450 ms; TI = 160 ms; field of view = 36 × 48 cm²; matrix 180 × 320; bandwidth 252 Hz/pixel; averages, 1; slice thickness 6 mm with 0.6-mm gap; turbo factor 33) and an eight-channel torso phased-array coil. The total lung was imaged using a stack of consecutive coronal slices within four to five breathholds. The subjects were instructed to breathe normally and stop breathing at the end of a normal inspiration.

Image Post-processing and Data Analysis

One radiologist (J.V.-C., 12 yr of experience) blinded to the bronchoalveolar lavage fluid results scored visually the extent of segmental lung infiltrates at all three time points using the following semiquantitative TIRM scoring system: TIRM0 = no infiltrate, TIRM1 = small peribronchial infiltrate, TIRM2 = infiltrate less than 50% of lung segment volume, TIRM3 = infiltrate 50–75% of lung segment volume, and TIRM4 = infiltrate more than 75% of lung segment volume (Figures 1 and 2). For intraobserver and interobserver variability assessment of the score, the data were reanalyzed 3 months after the first analysis by the same radiologist and by an additional radiologist (J.R., 2 yr of experience).

Statistical Analysis

Statistical analysis was performed using nonparametric Kruskal-Wallis and Wilcoxon tests. Results are given as median

and 25–75% quartile. For intraobserver and interobserver agreement kappa was calculated. For calculations JMP Pro 10 (SAS Institute GmbH, Böblingen, Germany) software package was used. Graphs were designed using Microsoft Excel 2010 (Microsoft Deutschland, Unterschleißheim, Germany).

Results

Spirometric measurements of both groups are shown in Table 2. There was a slight intermittent reduction in pulmonary function variables that was statistically significant in the asthma group but not in healthy subjects. The magnitude of deterioration was mild and in line with previous reports (28–30), although the number of challenged segments and therefore invasiveness was higher in our study.

Expectedly, instillation of allergen significantly increased eosinophil percentages in the asthma group but not in the control group (Figure 3A). In patients with asthma, there were significant differences between the percent of eosinophils in SA-, LA-, and S-treated segments at 24 hours post-challenge bronchoscopy (*P* < 0.001). Correspondingly significant differences between the TIRM score in SA-, LA-, and S-treated segments were observed at 24 hours post-challenge (*P* < 0.001) (Figure 3B). With increasing TIRM score at 6 and 24 hours the eosinophils per milliliter and the percent eosinophils per segment 24 hours post-challenge increased accordingly (*P* < 0.0001) (Table 3, Figure 4). The percent of macrophages and monocytes and percent of lymphocytes 24 hours post-challenge decreased with increasing TIRM score at 24 hours (percent macrophages,

P = 0.0002; percent lymphocytes, *P* = 0.029) (Table 3). The total cell count per milliliter 24 hours post-challenge significantly increased with increasing TIRM scores at 24 hours (*P* = 0.04) (Table 3). Neutrophils did not significantly change with increasing TIRM scores. Total protein in the lavage fluid 24 hours post-challenge increased significantly with increasing TIRM scores at 6 hours (*P* = 0.002) and 24 hours post-allergen challenge (*P* < 0.0001) (Table 3).

In healthy control subjects, the percent eosinophils per segment 24 hours post-challenge and the 24 hours TIRM score did not differ significantly between SA- (0.2 [0.0–0.5] percent eosinophils, TIRM score 0 [0–0]), LA- (0.5 [0.0–1.9] percent eosinophils, TIRM score 0 [0–0]) and S-treated (0.0 [0.0–0.4] percent eosinophils, TIRM score 0 [0–0]) segments (percent eosinophils *P* = 0.62; TIRM score *P* = 1.0).

TIRM Score Time Course

There were significant changes in TIRM score during the observation period in the SA and LA of the asthma group (both *P* < 0.001), whereas TIRM score did not change in the S segments (*P* = 0.37). Unexpectedly, TIRM score changed in the S segments of the control group (*P* = 0.003), whereas no change was observed in allergen-challenged segments (LA, *P* = 0.37; SA, *P* = 1.0). In patients with asthma, the TIRM score at baseline prechallenge was 0 (0–0) in all segments and increased significantly in the LA- and SA-treated segments but not in the S-treated segments at 6 and 24 hours (*P* < 0.001). In the control group, the TIRM score at baseline prechallenge was 0 (0–0) in all segments and increased significantly only in the S-treated segments at 6 hours (2 [0–2]; *P* = 0.01) but not at 24 hours (Figure 5).

Agreement of TIRM Score Readings

There was good intraobserver and interobserver agreement for TIRM score grading (intraobserver kappa = 0.66 for all time points and 0.63 for 24 h time point only; interobserver kappa = 0.66 for all time points and 0.72 for 24 h time point only).

Discussion

This study provides noninvasive quantitative measures of allergic airway inflammation by applying MRI to subjects undergoing segmental allergen challenge. The extent of the segmental lung infiltrate

measured noninvasively using MRI-derived TIRM score corresponds to the severity of the regional allergic reaction determined by the number and percentage of eosinophils in bronchoalveolar lavage fluid 24 hours after allergen challenge in patients with asthma and normal control subjects.

TIRM MRI score at 24 hours post-allergen challenge represents a specific biomarker for quantification and visualization of the regional allergen-induced inflammatory response. Although bright signal of TIRM MRI in general shows lung fluid and edema and is not specific for eosinophilic inflammatory response, in this controlled study setting it

was specific for the delayed phase response at 24 hours after segmental allergen challenge in patients with asthma. Importantly, TIRM MRI at 6 hours post-challenge revealed unspecific signals with elevated TIRM scores in healthy subjects. However, this may be because healthy control subjects underwent a slightly different protocol with lavage of two segments at the first bronchoscopy, which very likely impacts on the TIRM MRI signal at 6 hours post-bronchoscopy. Thus, residual saline lavage fluid from bronchoscopy might not have been fully reabsorbed in all lung segments. Second, at the 6-hour time point there may be

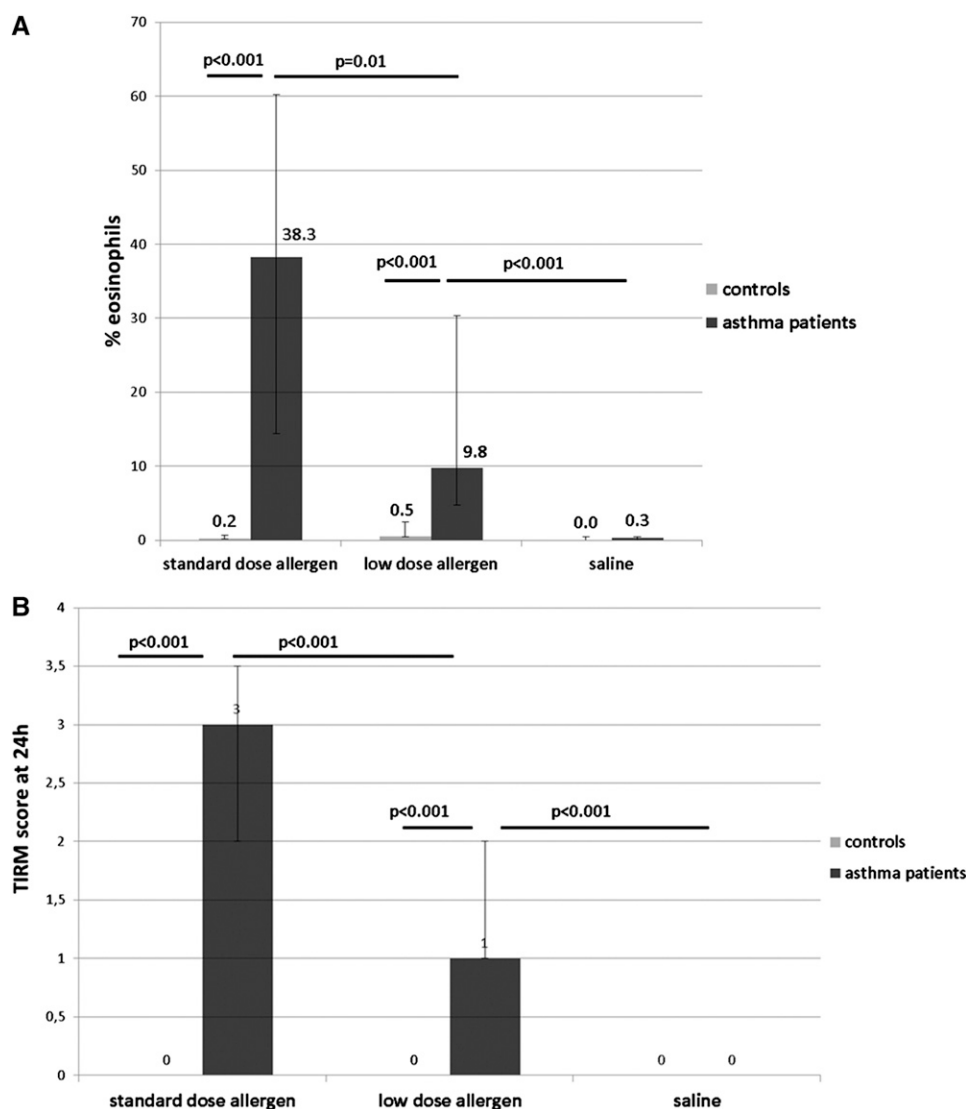


Figure 3. (A) Percentage of eosinophils (% eosinophils) in bronchoalveolar lavage at 24 hours in standard-dose allergen, low-dose allergen, and saline-treated segments in the asthma group compared with the control group. Data are presented as medians, 25–75th percentile error bars. (B) Turbo-inversion recovery-magnitude (TIRM) score at 24 hours in standard-dose allergen, low-dose allergen, and saline-treated segments in the asthma group compared with the control group. Data are presented as medians, 25–75th percentile error bars.

Table 3: Segmental Bronchial Lavage Fluid 24 Hours after Challenge Compared with MRI-derived TIRM Score 6 and 24 Hours after Challenge in Volunteers with Asthma

	Recovery (ml)	Total Cell Count × 10 ⁶ /ml	Eosinophils (%)	Eosinophils × 10 ⁶ /ml	Neutrophils (%)	Neutrophils × 10 ⁶ /ml	Macrophages (%)	Macrophages × 10 ⁶ /ml	Lymphocytes (%)	Lymphocytes × 10 ⁶ /ml	Total Protein (μg/ml)
6 Hours											
TIRM0	55 (51–66)	8.0 (4.2–14.9)	0.3 (0.0–1.3)	0.0 (0–0.1)	4 (1.8–23.3)	0.2 (0.1–2.4)	86.8 (66.5–91)	7.3 (3.9–13.2)	5 (3.5–8.5)	0.4 (0.2–1.4)	86 (72–111)
TIRM1	54 (48–70)	9.5 (7.3–19.9)	9.8 (5.5–37.3)	1.6 (0.4–4)	2 (1.3–6)	0.2 (0.1–1.0)	77.8 (53.8–83.8)	7.2 (5.6–10.7)	2.25 (2.0–4.8)	0.4 (0.1–0.6)	122 (82–166)
TIRM2	57 (46–66)	12.5 (9.0–31.3)	20.4 (4.1–32.3)	2.2 (0.4–8.6)	4.6 (1.7–24.5)	1.1 (0.2–5.0)	58.2 (43.3–80.2)	8.1 (6.4–13.0)	4.125 (3.4–6.5)	0.5 (0.4–1.0)	126 (103–168)
TIRM3	56 (41–63)	23.1 (8.4–45.2)	38.5 (19.6–66.9)	7.8 (2.2–28.1)	6.5 (2.6–18.1)	1.5 (0.6–5.1)	37.5 (24.8–58.3)	6.3 (4.3–11.7)	2.75 (1.9–4.5)	0.6 (0.2–1.6)	261 (144–523)
TIRM4	31 (30–32)	21.1 (18.3–23.8)	65 (56.7–73.3)	13.5 (13.4–13.5)	8.9 (7.5–10.3)	1.9 (1.4–2.5)	24 (18–30)	5.3 (3.3–7.1)	2.13 (1.3–3.0)	0.5 (0.2–0.7)	235 (110–359)
	<i>P</i> = 0.32	<i>P</i> = 0.10	<i>P</i> < 0.0001	<i>P</i> < 0.0001	<i>P</i> = 0.47	<i>P</i> = 0.36	<i>P</i> = 0.0008	<i>P</i> = 0.7	<i>P</i> = 0.07	<i>P</i> = 0.58	<i>P</i> = 0.002
24 Hours											
TIRM0	56 (51–68)	10.3 (4.4–14.7)	0.3 (0–1.3)	0.1 (0–0.1)	3.9 (1.4–19.9)	0.2 (0.1–1.3)	87.6 (71.1–92.9)	8.5 (4.1–12.9)	5 (3.3–8.2)	0.4 (0.2–1.2)	83 (73–111)
TIRM1	54 (49–68)	10.7 (7.3–18.3)	6.4 (3.4–9.3)	0.7 (0.4–1.4)	2.5 (1.4–18.6)	0.2 (0.1–3.4)	80.8 (70.4–84.3)	7.2 (6.4–9.6)	3.9 (2.4–6.1)	0.5 (0.4–0.6)	101 (83–133)
TIRM2	47.5 (40–61)	14.9 (9.2–22.5)	33.8 (25.9–48.9)	5.7 (2.7–12.1)	5.5 (3.1–7.6)	0.8 (0.3–1.5)	53.8 (27.3–60.8)	6.1 (4.2–9.3)	2.5 (1.2–4.8)	0.3 (0.2–0.8)	130 (116–162)
TIRM3	58 (55–63)	24.1 (18–49.8)	37.4 (17.3–50)	10.6 (2.3–24.8)	7.9 (2.1–23.4)	3.0 (0.6–7.4)	37.8 (33.7–59.7)	11.3 (7.4–19.7)	4.5 (3.4–5.4)	1.1 (0.7–2.0)	434 (211–525)
TIRM4	38 (31–67)	23.1 (6.7–52.3)	63.8 (24.4–80.2)	14.7 (1.8–42.2)	5.5 (2.6–17.1)	1.3 (0.7–1.8)	27.5 (15.2–52.4)	6.1 (3.5–7.7)	2.3 (0.9–2.6)	0.5 (0.1–1.2)	261 (146–1162)
	<i>P</i> = 0.49	<i>P</i> = 0.04	<i>P</i> < 0.0001	<i>P</i> < 0.0001	<i>P</i> = 0.87	<i>P</i> = 0.37	<i>P</i> = 0.0002	<i>P</i> = 0.2	<i>P</i> = 0.029	<i>P</i> = 0.11	<i>P</i> < 0.0001

Definition of abbreviations: MRI = magnetic resonance imaging; TIRM = turbo-inversion recovery-magnitude.

P Kruskal-Wallis test.

still a transient nonspecific reaction to the mechanical interactions of the bronchoscopic lavage procedure causing edema and thus an increased TIRM score. Interestingly, unspecific elevation of TIRM scores in non-allergen-treated lung segments was only present in healthy subjects, which favors the former explanation.

Blé and coworkers (18) demonstrated the feasibility of proton MRI to detect extravascular and luminal fluid in a mouse model of allergen-induced airway inflammation using a gradient echo sequence. Twenty-four hours after the fourth administration of ovalbumin, intense and continuous fluid signals were detected in proximal lung regions. At 72 hours after the fourth administration of ovalbumin, remaining signals had a discontinuous texture. Histologic analysis revealed edema and secreted mucus at 24 hours, whereas only mucus was encountered at 72 hours. In their study the volume of fluid signal intensity in the lungs detected with MRI significantly correlated with the total protein level and the number of eosinophils quantified in the bronchoalveolar lavage fluid samples. Recently, using the same animal model, Bianchi and coworkers (31) used an ultrashort-TE MRI sequence (TE 276 μs at 4.7 T) with high spatial resolution and good signal-to-noise ratio to improve image quality for noninvasive quantification of peribronchial eosinophilic inflammation. These findings correspond well to the increasing extent of the observed infiltrate/TIRM score with increasing percentages of eosinophils at 24 hours post-challenge in

the human subjects with asthma in the present study.

Edema was also measured noninvasively in a longitudinal rat asthma animal model post-allergen challenge by Beckmann and coworkers (22) using a gradient echo MRI sequence. They showed that the volume of edema in the lung parenchyma post-allergen challenge resolved faster in animals treated with glucocorticosteroids compared with nontreated rats. Although this finding is important because it highlights the potential to detect treatment effects on airway inflammation by noninvasive means of MRI, only a few studies of segmental allergen provocation to test antiasthma therapies have been reported (32, 33).

Recently, positron emission tomography in six patients with asthma showed decreased ventilation and perfusion and an increased relative fluorodeoxyglucose uptake rate in challenged lung segments 10 hours after

provocation (34). The authors could also show that the ratio of eosinophil count (allergen to diluent) at 24 hours was linearly related to the ratio of the fluorodeoxyglucose uptake rate at 10 hours post-allergen challenge. However, in contrast to MRI, positron emission tomography exposes the human volunteers to a considerable amount of ionizing radiation, which is a notable limitation for serial positron emission tomography measurements.

In the present study the TIRM MRI sequence was chosen because the inflammatory lung fluid and edema were best depicted and visually quantified on TIRM MRI compared with half-Fourier-acquisition single-shot turbo spin-echo, ECG-gated steady-state free precession or volume interpolated gradient echo MRI sequences in our experience (data not shown). The strength using the TIRM MRI sequence was that the unaffected lung tissue showed only very little signal and no signal

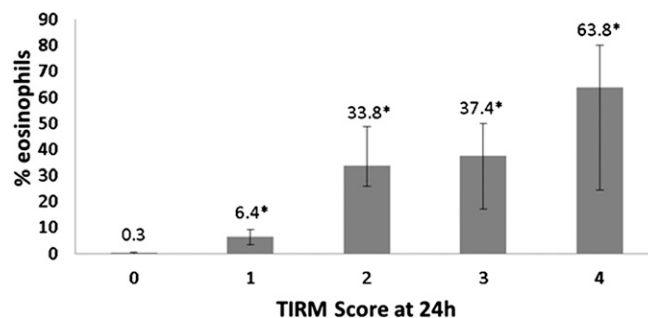


Figure 4. Percentage of eosinophils (% eosinophils) in bronchoalveolar lavage at 24 hours post-challenge from lung segments according to turbo-inversion recovery-magnitude (TIRM) signal. Data are presented as medians, 25–75% percentile error bars. **P* < 0.001 compared with TIRM 0 group.

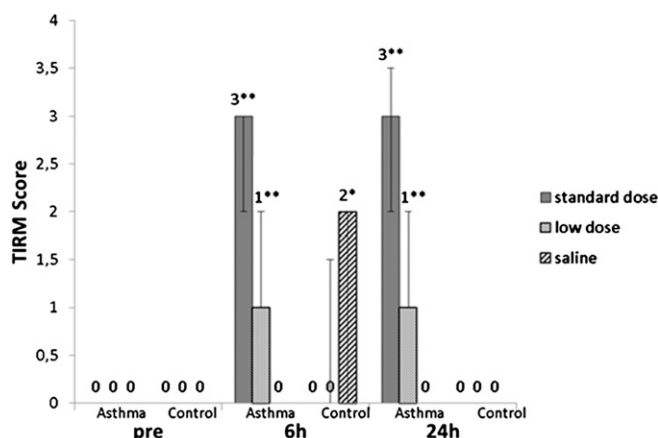


Figure 5. Turbo-inversion recovery-magnitude (TIRM) score before challenge (pre) and during post-challenge observation period (6 and 24 h) per study group. Median TIRM score in both groups is 0 pretreatment with significant increases in patients with asthma at 6 and 24 hours. In the control group, intermittent elevation of median TIRM score is present at 6 hours. Data are presented as medians, 25–75% percentile error bars. ** $P < 0.001$, * $P < 0.01$ compared with pretreatment values.

was seen at 24 hours in any healthy volunteer in the challenged segments. Moreover, the TIRM MRI sequence was quite sensitive showing small amounts of peribronchial edema at 24 hours in cases with mild eosinophilic response to allergen challenge in this study. The segmental allergen challenge is usually of higher dose compared with techniques using whole-lung allergen challenge. From the presented data, it can therefore not be concluded that the TIRM MRI method can also detect or determine the degree of

inflammation in “naturally” occurring exacerbation or following whole-lung allergen challenge.

Because of the lack of ECG gating cardiac pulsation artifacts were noted in the phase-encoding direction. Because the phase encode was in the craniocaudal direction the cardiac pulsation artifacts remained mainly within the mediastinum and did not affect the challenged lung segments.

In the present study we could show that the presented TIRM score at 24 hours is fast

and easy to acquire without the need of time-consuming image post-processing and sophisticated analysis tools. Although it is semiquantitative we could show an association of the TIRM score with the severity of the eosinophilic inflammatory response in volunteers with asthma and good intraobserver and interobserver agreement. At 24 hours kappa was 0.72 for interobserver agreement TIRM score. At this time point, no interobserver difference occurred of 3 or more TIRM score units. In only 6.6% of the evaluated segments a difference of 2 TIRM score units was noted, all between the lower TIRM scores of 0 and 2. Thus, a trained radiologist with expertise in MRI should perform the analysis to achieve good interobserver agreement.

In conclusion, the presented MRI-based noninvasive TIRM score is a promising biomarker for the detection of the inflammatory response after segmental allergen challenge in patients with asthma and may serve to monitor the therapeutic effectiveness of novel antiinflammatory drugs in future human trials. ■

Author disclosures are available with the text of this article at www.atsjournals.org.

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