



**HAL**  
open science

## Improved ultrasound attenuation measurement method for the non-invasive evaluation of hepatic steatosis using FibroScan ®

Stéphane Audière, Aymeric Labourdette, Véronique Miette, Céline Fournier, Redouane Ternifi, Salem Boussida, Philippe Pouletaut, Fabrice Charleux, Sabine Bensamoun, Stephen A Harrison, et al.

### ► To cite this version:

Stéphane Audière, Aymeric Labourdette, Véronique Miette, Céline Fournier, Redouane Ternifi, et al.. Improved ultrasound attenuation measurement method for the non-invasive evaluation of hepatic steatosis using FibroScan ®. *Ultrasound in Medicine & Biology*, 2021, 47 (11), pp.3181-3195. 10.1016/j.ultrasmedbio.2021.07.007 . hal-03290919

**HAL Id: hal-03290919**

**<https://hal.utc.fr/hal-03290919>**

Submitted on 19 Jul 2021

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

# Improved ultrasound attenuation measurement method for the non-invasive evaluation of hepatic steatosis using FibroScan<sup>®</sup>

Stéphane Audière<sup>a</sup>, Aymeric Labourdette<sup>a</sup>, Véronique Miette<sup>a</sup>, Céline Fournier<sup>a</sup>, Redouane Ternifi<sup>b,1</sup>, Salem Boussida<sup>b,2</sup>, Philippe Pouletaut<sup>b</sup>, Fabrice Charleux<sup>c</sup>, Sabine F.Bensamoun<sup>b</sup>, Stephen A.Harrison<sup>d</sup> and Laurent Sandrin<sup>a</sup>

<sup>a</sup> Echosens, Paris, France;

<sup>b</sup> Université de technologie de Compiègne, CNRS, Biomechanics and Bioengineering, Centre de recherche Royallieu, CS 60319, 60203 Compiègne Cedex, France;

<sup>c</sup> ACRIM-Polyclinique Saint Côme, Medical Radiology, Compiègne, France;

<sup>d</sup> Radcliffe Department of Medicine, University of Oxford, UK;

Corresponding author:

Stéphane Audière

Postal address: 6 rue Ferrus, 75014 Paris, France

Telephone number: +33 1 44 82 78 50

E-mail address: stephane.audiere@echosens.com

In preparation for *Ultrasound in Medicine and Biology*

---

<sup>1</sup> Present Address: Mayo Clinic Rochester, Rochester, Minnesota, USA

<sup>2</sup> Present Address: Centre hospitalier universitaire Amiens-Picardie, France

## 1 **Abstract**

2 Controlled Attenuation Parameter (CAP) is a measurement of ultrasound attenuation used to  
3 assess liver steatosis non-invasively. However, the standard method has some limitations. We  
4 aimed to assess the performance of a new CAP method by ex vivo and in vivo assessments.  
5 The major difference with the new method is that it uses ultrasound data continuously acquired  
6 during the imaging phase of the FibroScan examination. Seven reference tissue-mimicking  
7 phantoms were used to test the performances. In vivo performances were assessed on two  
8 cohorts (in total 195 patients) of patients using magnetic resonance imaging proton density fat  
9 fraction (MRI-PDF) as a reference. The precision of CAP was improved by more than 50% on  
10 tissue-mimicking phantoms and between 22% and 41% in the in vivo cohort studies. The  
11 agreement between both methods was excellent and the correlation between CAP and MRI-  
12 PDFF improved in both studies (0.71 to 0.74, 0.70 to 0.76). Using MRI-PDFF as a reference,  
13 the diagnostic performance of the new method was at least equal or superior (area under the  
14 receiver operating curve 0.889 to 0.900, 0.835 to 0.873). This study suggests that the new  
15 continuous CAP method can significantly improve the precision of CAP measurements ex vivo  
16 and in vivo.

## 17 **Keywords**

18 Ultrasound attenuation, Controlled Attenuation Parameter (CAP), Liver, Steatosis,  
19 Elastography, Vibration-Controlled Transient Elastography (VCTE), FibroScan, Proton  
20 Density Fat Fraction (PDFF).

# 1 **Introduction**

2 Nonalcoholic fatty liver disease (NAFLD) is a one of the leading causes of liver disease, found  
3 in 20-25% of adults in the developed world (Younossi, et al. 2016). About half of NAFLD  
4 patients are obese, with the prevalence of NAFLD in nonobese or lean patients at 15.7% and  
5 10.2%, respectively (Harrison, et al. 2021). It has also been reported that 26% of American  
6 obese children have NAFLD (Elizabeth, et al. 2019). Thus, it has major health and economic  
7 burdens. The severity is worsened as NAFLD can go undetected for some time, increasing the  
8 risk of it developing into the more advance and progressive form of nonalcoholic steatohepatitis  
9 (NASH), which can manifested further into cirrhosis and liver cancer. A recent study (Harrison,  
10 et al. 2021) reported the overall prevalence of NAFLD was 38% and 14% for NASH confirmed  
11 by biopsy in a cohort of asymptomatic middle-aged American adults. NAFLD is defined as  
12 hepatic steatosis, which is the accumulation of triglycerides within hepatocytes that exceeds 5%  
13 of liver weight. Liver biopsy is the gold standard method to assess liver steatosis (Bravo, et al.  
14 2001), but it carries strong limitations due to its invasiveness and potential sampling error  
15 (Ratziu, et al. 2005, Shahin Merat, et al. 2012).

16 Due to the increasing prevalence of NAFLD and the limitations of liver biopsy for diagnosis,  
17 there is a strong need for non-invasive tests to accurately detect hepatic steatosis. Magnetic  
18 resonance imaging derived proton density fat fraction (MRI-PDFF) has emerged as a leading  
19 non-invasive modality for the assessment of hepatic steatosis (Reeder, et al. 2012, Tang, et al.  
20 2013), and as a reliable alternative to the histological assessment of hepatic steatosis in patients  
21 with NAFLD (Tang, et al. 2014). However, a wide application of this modality is impaired by  
22 its cost and availability. Ultrasound (US) techniques have been proposed since fatty liver is  
23 associated with increased US attenuation (Lu, et al. 1999, Gaitini, et al. 2004). In fact, lipid  
24 droplets can greatly contribute to energy absorption during US propagation due to their typical  
25 dimension in liver tissue (Kanayama, et al. 2013).

1 US attenuation measurement using controlled attenuation parameter (CAP) (Sasso, et al. 2010,  
2 Sasso, et al. 2012, Sasso, et al. 2016) (Echosens, Paris, France) was introduced in 2010 to assess  
3 liver steatosis non-invasively. CAP measures the attenuation of the US beam that travels  
4 through the liver tissue, usually at a frequency of 3.5 MHz. It is available on the FibroScan (FS)  
5 device which concomitantly assesses CAP and liver stiffness using vibration-controlled  
6 transient elastography (VCTE) (Sandrin, et al. 2003, Tapper, et al. 2015). The operation of  
7 FibroScan does not require special skills in US. CAP can be measured using both M and XL  
8 probes. The CAP final results are the median and the interquartile range (IQR) of several  
9 (typically 10) manually triggered sequential measurements of US attenuation. CAP is expressed  
10 in dB/m and ranges from 100 to 400.

11 Several US scanner manufacturers have recently introduced alternative methods to assess US  
12 attenuation using B-mode US: ATT (Hitachi Ltd, Japan) (Iijima 2018), ATI (Canon Medical  
13 Systems, Japan) (Tamaki, et al. 2018, Koizumi, et al. 2019), and UGAP (GE Healthcare, USA)  
14 (Fujiwara, et al. 2018, Bende, et al. 2020), UDFE (Siemens Healthineers, Germany) (Labyed  
15 and Milkowski 2020). However, US scanners require that the operator be skilled in US as they  
16 must select manually a region of interest on the US image to assess the liver tissue. This can be  
17 an advantage in the presence of a liver lesion which would not be detected using FS due to the  
18 lack of 2D US imaging.

19 Despite CAP being a good surrogate marker of hepatic steatosis (De Lédinghen, et al. 2014,  
20 Cardoso, et al. 2016, De Lédinghen, et al. 2016, De Lédinghen, et al. 2017, Karlas, et al. 2017,  
21 Naveau, et al. 2017, Thiele, et al. 2018) and has the ability to monitor the improvement of  
22 hepatic steatosis in patients with NAFLD (Paul, et al. 2018, Eddowes, et al. 2019) it has several  
23 limitations with its current measurement method. CAP accuracy for detecting fatty liver  
24 declines in the presence of high variability (Wong, et al. 2017). In addition, studies on the inter-  
25 observer concordance in the CAP values have shown that the mean difference in CAP values  
26 between two observers is up to 20 dB/m (Recio, et al. 2013, Ferraioli, et al. 2014). Hence, this

1 difference should be considered in longitudinal follow-up of patients. Lower performances for  
2 detecting fatty liver were reported when the IQR is superior to 40 dB/m (Wong, et al. 2017,  
3 Mendes, et al. 2018). Whereas another study shows that CAP variability seems to have no  
4 influence on diagnostic performance (Naveau, et al. 2017). The main cause of variability is the  
5 sensitivity of CAP to the presence of heterogeneities from blood vessels and nodules in the US  
6 signal (Audière, et al. 2013). Furthermore, the distribution of steatosis in the liver may be  
7 heterogeneous (Qayyum, et al. 2012, Bannas, et al. 2015). [Recent studies \(Fujiwara, et al. 2018,](#)  
8 [Ferraioli, et al. 2019, Ferraioli, et al. 2020\)](#) report that US attenuation measurements using B-  
9 mode techniques perform significantly better than CAP for steatosis detection with MRI-PDFF  
10 as a reference. This increased performance may be attributed to the manual selection of the ROI  
11 in the B-mode image which allows the exclusion of structures that may affect the measured  
12 values and to the larger 2D ROI which is used to assess ultrasound attenuation. The lack of  
13 robustness and precision of the CAP by the standard method is a possible source of suboptimal  
14 performances (Petroff, et al. 2021).

15 We have developed an alternative method for CAP measurement, which uses continuous CAP  
16 during the imaging phase of the examination with the FS device that we propose will improve  
17 the precision of CAP measurement using FS. The aim of this study was to validate the  
18 performances of the new method on tissue-mimicking phantoms and retrospectively in cohorts  
19 of patients using MRI-PDFF as a reference.

## 20 **Material and method**

### 21 **Standard CAP method**

22 CAP measures the US attenuation using US signals collected with the single element US  
23 transducer located at tip of the probe of the FS device. Using the standard CAP method, the  
24 final CAP results include the median and the IQR (expressed in dB/m) of the individual US  
25 attenuation measurements performed during the examination with the FS device. [Individual US](#)

1 attenuation measurements are estimated by processing the US data collected for the shear wave  
2 speed measurements, triggered manually by the operator, from which stiffness measurements  
3 are derived (Sandrin, et al. 2003). The recommended number of US attenuation measurements  
4 is typically 10 since it is recommended that operators perform 10 valid stiffness measurements.  
5 Given that a shear wave speed measurement lasts 80 ms, the US attenuation measurements are  
6 typically collected during a cumulative duration of less than one second. These individual  
7 measurements are distributed over the entire duration of the examination. FS can be used with  
8 three different probes (S, M and XL). The standard CAP method is only compatible with M and  
9 XL probes (Sasso, et al. 2016). The measurement depths and center frequency with M and XL  
10 probes are 25-65 mm and 3.5 MHz, 35-75 mm and 2.5 MHz, respectively. Although the US  
11 transducer center frequency is different on the M probe and XL probe, the US attenuation is  
12 computed at the same US frequency of 3.5 MHz, leveraging the large bandwidth of US  
13 transducers. The lower and upper limits for CAP measurement are 100 dB/m and 400 dB/m,  
14 respectively.

## 15 Principle of the new method (continuous CAP)

16 The main difference between the current and the new method is that the new method uses US  
17 signals acquired continuously during the imaging phase of the examination with the FS device.  
18 For clarity reasons, the new method will therefore be named ‘continuous CAP method’ in this  
19 document. The continuous CAP method is as automated as the initial method. The influence on  
20 the operation of the device is very limited. Moreover, the proprietary algorithm (Sasso, et al.  
21 2010, Sasso, et al. 2012, Sasso, et al. 2016) used to compute the individual US attenuation  
22 measurements using the US signal is identical in both methods. Contrarily to the standard CAP  
23 method, the continuous CAP method is compatible with all probes including the S probe  
24 (Ferraioli, et al. 2012), which uses a center frequency of 5.0 MHz. The US attenuation is  
25 computed at the same US frequency of 3.5 MHz whatever the probe model being used.

1 Three major improvements are implemented in the continuous CAP method. First, the number  
2 of individual US attenuation measurements from which CAP is calculated is higher and  
3 corresponds to a larger volume of liver tissue sampled. Second, the US signals are qualified  
4 using a dedicated validity criteria. And third, the measurement depths are adapted  
5 automatically.

#### 6 Increased volume sampling

7 The volume of liver tissue sampled with the standard and continuous CAP methods are  
8 schematically represented in figure 1. Using the continuous CAP method, the CAP value is  
9 derived from US attenuation measurements which are computed from the US signals acquired  
10 continuously in real-time during the imaging phase of the FS examination (i.e., FS imaging  
11 mode between stiffness measurements) at a repetition frequency of 20 Hz. At least 200 US  
12 attenuation measurements are recommended with the continuous CAP method, which  
13 correspond to at least 10 seconds of US acquisition; much larger than the less than 1 second  
14 acquisition used with the standard CAP method. Furthermore, during this acquisition time, the  
15 liver moves in front of the probe due to breathing with a frequency of approximately 0.5 Hz,  
16 and an amplitude of 20 mm, mainly in the cranio-caudal direction (Bussels, et al. 2003).

#### 17 Selection of US signals

18 The selection of US signals used to compute the final CAP value is different in both methods.  
19 In the standard CAP method, US signals are selected based on the validity of the shear wave  
20 propagation induced when the operator presses on the probe button. In the continuous CAP  
21 method, US signals are selected based on the US characteristics of the signals. The selection of  
22 US signals is performed automatically using the Liver Targeting Tool (LTT) (Audière, et al.  
23 2013), which assesses the absence of heterogeneities in the US signal. LTT is displayed in real-  
24 time on the screen of the FS during the examination to assist the operators in finding an optimal  
25 measurement site.

#### 26 Measurement depths automated adjustment



1 The continuous CAP method includes an automated adaptation of the measurements depths  
2 based on the probe to liver capsule distance (PCD) (Audière, et al. 2010). The purpose is to  
3 avoid biases of US attenuation measurements in the presence of subcutaneous tissues in the  
4 region of measurements. Several depth ranges are available depending on the probe model: 25-  
5 65 mm or 30-70 mm with the M probe, 35-75 mm, 40-80 mm or 45-85 mm with the XL probe.  
6 With the S probe, depth ranges are selected manually at the beginning of the exam and  
7 correspond to the stiffness measurement depths (S1: 15-40 mm, S2: 20-50 mm).

#### 8 Examination outputs

9 Given the large number of individual measurements collected with the continuous CAP method,  
10 a normal distribution of measurements is observed. The final CAP results are expressed as the  
11 mean and standard deviation (SD).

#### 12 Test on tissue-mimicking phantoms

##### 13 Tissue-mimicking phantoms characteristics

14 The standard CAP and continuous CAP methods were tested on seven custom reference tissue-  
15 mimicking phantoms (Gammex INC, Middleton, WI, USA), which US attenuations are 95, 142,  
16 207, 249, 339, 403 and 475 dB/m, with an uncertainty  $\pm 20$  dB/m. Reference US attenuation  
17 values were measured at the Wisconsin Institute of Medical Research (Madison, WI 53705,  
18 USA) for each phantom using a sample of each batch material. A standard narrowband through-  
19 transmission substitution technique (Madsen, et al. 1982) was used.

##### 20 Test method

21 The continuous CAP method was evaluated with S, M and XL probes while the standard CAP  
22 method was only evaluated with M and XL probes given that the S probe is not compatible. US  
23 acquisitions measurements were performed with a FS device acquisition platform connected to  
24 a standard computer running Matlab (Mathworks, Natick, MA, USA). The shear wave

1 generation was disabled since the reference US attenuation phantoms were too stiff to obtain  
2 valid stiffness measurements. Each set of measurements consisted in scanning a phantom by  
3 moving the probe in the center (20 x 20 mm<sup>2</sup>) of the phantom surface. A motorized linear  
4 translation stage (Newport Corporation, Irvine, CA, USA) was used to move the probes at the  
5 surface of the phantom, with a displacement speed of 20 mm/s. Acquisitions for the assessment  
6 of the continuous CAP method were performed while the probe was moving at the surface of  
7 the phantom to mimic the liver movement during the FS imaging mode, allowing to achieve  
8 200 US attenuation measurements. Acquisitions for the assessment of the standard CAP method  
9 were performed with a step by step displacement of the probe at the surface of the phantom to  
10 mimic the acquisition of US signals during the FS sequential stiffness measurements. Ten  
11 individual measurements were performed at 10 different locations within the same region of  
12 interest used with the continuous CAP method. Acquisitions for both methods were performed  
13 at the same depths. Due to the large distribution of the reference US attenuation of tissue-  
14 mimicking phantoms, the lower and upper limits for CAP measurement were set to 50 dB/m  
15 and 500 dB/m, respectively.

#### 16 Statistical analysis

17 The Shapiro-Wilk test was used to assess the normal distribution of the measurements  
18 performed with both methods. For comparison purposes, the standard CAP results were  
19 expressed as the mean (instead of median for the standard CAP method) of all performed  
20 measurements. The CAP precision is expressed as the SD. The significance of the precision  
21 difference between the two methods is achieved through a Student's-test.

22 The intra-class coefficient (ICC) was used to assess the agreement between the two methods  
23 and the reproducibility on phantoms, as suggested in (Raunig, et al. 2015). Two other probes  
24 of each type were used to estimate the reproducibility of both methods. Agreement was  
25 classified as poor (ICC = 0.00-0.20), fair to good (ICC = 0.40-0.75) or excellent (ICC > 0.75)  
26 (Fleiss, et al. 2013).

## 1 In vivo studies

2 The comparison between both CAP methods was performed using clinical data collected during  
3 two different studies (study cohorts A and B). MRI-PDFF was used as a reference to identify  
4 the ability of both CAP methods to identify patients with a MRI-PDFF of more than 5%. The  
5 study protocols conformed to the ethical guidelines of the 1975 Declaration of Helsinki and was  
6 approved by the local Ethic committee. Patients were enrolled after written informed consent  
7 was obtained. For both study cohorts, CAP measurements were performed with a FibroScan  
8 502 Touch configured to record the raw US radio-frequency signals collected during the whole  
9 FS examinations. The operators were following the training requirements relative to the  
10 standard measurements of liver stiffness by VCTE and CAP. The continuous CAP method was  
11 evaluated retrospectively by reprocessing the raw data recorded in examination files of the  
12 standard CAP method.

### 13 Study cohort A

14 Patients referred for a routine liver screening with no prior history of liver disease or alcohol  
15 abuse were offered to participate in this prospective prevalence study. All patients who  
16 underwent CAP and MRI-PDFF examinations at the Brooke Army Medical Center (San  
17 Antonio, Texas, USA) between January 2016 and December 2016 were eligible. MRI-PDFF  
18 results were reprocessed with Liver MultiScan IDEAL algorithm (Hardy and Mcpherson 2017)  
19 (liver MultiScan™, Perspectum Diagnostics, Oxford, England), which assesses hepatic  
20 steatosis, fibrosis and iron content. Region of interest (ROI) was positioned by the clinician on  
21 the MRI-PDFF parametric image, taking the most representative area. This area is positioned  
22 without taking into account the position of the FS measurement.

23 A total of 201 consecutive patients fulfilled the inclusion criteria (male and female patients, age  
24 18 to 80), and consisted of retired and active military personnel and their dependents. Among  
25 them, 74 were excluded because the MRI-PDFF could not be performed and 14 because the

1 delay between FS and MRI-PDFF examinations was superior to 100 days. Finally, the study A  
2 included 113 patients.

### 3 Study cohort B

4 Patients referred for a liver MRI examination were offered participation in a prospective  
5 prevalence study. All patients who underwent CAP and MRI-PDFF examinations at the  
6 ACRIM-Polyclinique Saint-Côme (Compiègne, France) between February 2017 and October  
7 2018 were eligible. MRI-PDFF ROI in liver were placed at nearly the same location as the  
8 ROI of the FS examination (Bensamoun, et al. 2008, Ternifi, et al. 2018)

9 A total of 90 consecutive patients fulfilled the inclusion criteria (male and female patients, age  
10 18 to 80). Among them, five were excluded because the CAP files with raw US data were  
11 unusable (corrupted files) and two patients because the matching between FS and MRI  
12 examinations was not possible. One patient was also excluded because MRI-PDFF was not  
13 performed. Finally, the study B included 82 patients.

14 The two study cohorts were not pulled as they involved different MRI devices associated with  
15 different algorithms for the reconstruction of MRI-PDFF maps. Furthermore, the ROI  
16 placements were performed by two different teams, with two different techniques as explained  
17 above.

### 18 Magnetic Resonance Imaging

19 MRI-PDFF magnetic resonance imaging-based phenotyping was performed using 1.5T MRI  
20 devices (Avanto, Siemens, and Signa HDx, General Electric). Patients were in the supine  
21 position. MRI-PDFF results were estimated using the Dixon 3 points method (Ma 2008) in both  
22 studies.

### 23 Statistical analysis

24 CAP results were expressed as the median and the mean of all valid measurements for the  
25 standard CAP method and the continuous CAP method, respectively. SD was used to assess the

1 precision of both methods to ease the comparison. The significance of the precision difference  
2 between the two methods is achieved through a Student's-test.

3 The ICC were used to assess the agreement between the two methods. [A Bland Altman analysis](#)  
4 [was used to determine the bias between the two methods.](#)

5 Data collected in vivo were used to assess the performance of both CAP methods versus MRI-  
6 PDFF. Pearson's rank correlations were used to evaluate the relationship between CAP values  
7 and log MRI-PDFF. Correlation obtained with both CAP methods were compared using the  
8 Hittner's test. The test is significant if the p-value is  $<0.05$ . The performance of both CAP  
9 methods for identifying patients with hepatic steatosis defined by MRI-PDFF of more than 5%  
10 was assessed using a Receiver Operating Characteristic (ROC) analysis. The diagnostic  
11 performance was evaluated in terms of area under the receiver operating curve (AUROC).  
12 AUROC obtained with both CAP methods were compared using the Delong test (difference  
13 test and non-inferiority test with a 0.02 margin). The test is significant if the p-value is  $<0.05$ .  
14 CAP cut-off values were estimated by maximizing sensitivity and specificity (Youden's index).  
15 A linear fit was performed between CAP (in dB/m) and the logarithmic transformation of MRI-  
16 PDFF (in percentage). All statistical analyses were performed using the R software (The R  
17 Foundation for Statistical Computing, Vienna, Austria) and the graphics, stats, pROC, ggplot2,  
18 IRR, BlandAltmanLeh and RVAideMemoire packages.

## 19 **Results**

### 20 Validation on tissue-mimicking phantoms

21 We assessed the two CAP methods with seven tissue-mimicking phantoms. The comparison  
22 and precision of both CAP methods versus reference US attenuations are presented in figure 2.  
23 The precision of both CAP methods for each scan is represented by an error bar ( $\pm$ SD). The  
24 results of the linear regressions between CAP and reference US attenuations are provided on  
25 each graph. The relationship is closer to identity with the continuous CAP method than with

1 the standard CAP. As shown in figure 3, the precision (in term of SD) was improved by 57%  
2 and 63% for M and XL probes, respectively. The ICC between the standard and continuous  
3 CAP methods was 0.996 [0.978; 0.999] and 0.988 [0.935; 0.998], with the M probe and XL  
4 probe respectively, showing excellent agreement. In terms of reproducibility, the ICC for both  
5 methods are all above 0.98.

## 6 In vivo validation

### 7 Patients and examinations characteristics

8 As described above, two study cohorts were formed of 113 patients for cohort A and 82 patients  
9 for cohort B. The characteristics of the two study cohorts are provided in table 1. For studies A  
10 and B, respectively 83% and 94% of CAP measurements were performed at the same depth for  
11 both methods. In study A, 88% of FS examinations were performed by the same expert operator.  
12 In study B, FS examinations were equitably distributed between two novice operators.

### 13 Comparison versus MRI-PDFF

14 The comparison of both CAP methods versus MRI-PDFF in both study cohorts is given in table  
15 2. The relationships between MRI-PDFF and CAP are shown figure 4. ROC curves for a MRI-  
16 PDFF  $\geq 5\%$  are shown figure 5.

### 17 Agreement between standard and continuous methods, bias, precisions

18 The ICC between the standard and continuous CAP methods was 0.901 and 0.940 for study  
19 cohort A and B, respectively. A Bland Altman analysis is presented in figure 6. Compared to  
20 the standard method, the CAP continuous method is on average 8.6 dB/m and 5.6 dB/m lower  
21 for study cohort A and B, respectively.

22 The precisions of both CAP methods are presented in figure 7. Box-and-whisker plots were  
23 used to appraise the precision of both CAP methods. In study cohort A, the precision was  
24 improved by 41% and 33% using the continuous CAP method with the M probe and XL probe,

1 respectively. In study cohort B, the precision was improved by 38% and 22% using the  
2 continuous CAP method with the M probe and XL probe, respectively.

### 3 **Discussion**

4 This study demonstrates that the precision of CAP measurement can be improved using the new  
5 continuous CAP method. The main difference between the standard and the continuous CAP  
6 method was the collection and selection of the US signals used to compute the US attenuation.  
7 The continuous CAP method relies on the collection of US signals collected during the imaging  
8 mode of the examination with a FS device. Furthermore, only US signals of sufficient quality  
9 in terms of homogeneity are automatically selected for computation. The performances of the  
10 standard and new methods were studied in phantoms as well as in vivo.

11 The tests on homogeneous tissue-mimicking phantoms of known reference US attenuations  
12 demonstrated that both the standard and continuous CAP methods are accurate although the  
13 precision with the continuous CAP method was improved by 57% and 63% for M probe and  
14 XL probe, respectively. ICC between the two methods and reproducibility tests showed a  
15 perfect agreement. The improved precision of CAP measurements using the continuous CAP  
16 method was also observed in vivo in the two study cohorts. The precision of CAP was on  
17 average improved by 34% with the new method.

18 Given that US signals obtained on tissue-mimicking phantoms are free of artefacts, the  
19 improved precision on phantoms may be attributed only to the larger number of US attenuation  
20 measurements collected with the new method.

21 The improvement of precision with the continuous CAP method is obtained by collecting the  
22 US signals during the imaging mode of the examination with the FS device instead of relying  
23 on the US signals collected for liver stiffness measurements. In study cohort A and study cohort  
24 B, the number of US attenuation measurements collected during the imaging mode and used  
25 for CAP measurement is on average 900 and 1555, respectively. These numbers are much

1 higher than the 10 US attenuation values usually obtained with the standard CAP method.  
2 Furthermore, given that these values are collected at a maximum pulse-repetition frequency of  
3 20 Hz, they translate into equivalent durations of 45 seconds and 778 seconds, respectively.  
4 These durations are much higher than the equivalent duration using the standard CAP method,  
5 which is about one second. The longer acquisition time associated with liver movement due to  
6 breathing significantly increases the volume of liver tissue sampled with the new method and  
7 therefore the spatial averaging. Consequently, the precision of the final CAP value with the  
8 continuous CAP method is better than with the standard method. Although different US signals  
9 are used for the assessment of CAP and liver stiffness measurements using the continuous CAP  
10 method. Both values still reflect the same portion of the liver as they are acquired during the  
11 same examination, which combines imaging sequences during which US attenuation values are  
12 collected and elastography measurement sequences during which shear wave speed and  
13 stiffness values are measured.

14 The continuous CAP method demonstrated superior performances (although not significantly)  
15 in terms of AUROCs and correlation with MRI-PDFF in both study cohorts, which suggests a  
16 diagnostic performance at least as good as the standard CAP method. The DeLong test with a  
17 0.02 AUROC margin demonstrated a significant non-inferiority of the continuous CAP method  
18 when compared to standard CAP method. This improvement may be attributed to the improved  
19 precision and the enhanced selection of US signals used in both methods. As a matter of fact,  
20 in the standard CAP method the selection of US signals is based on the validity of stiffness  
21 measurement which may be suboptimal. Indeed, the US attenuation estimate is influenced by  
22 the presence of heterogeneities in the US signal which may not affect the shear wave  
23 propagation map and the associated shear wave speed assessment. The presence of  
24 subcutaneous tissues (fat, muscle, etc.) or blood vessels within the ROI (Shen, et al. 2015)  
25 usually result in hyperechogenic artefacts in the US signals, which may - depending on their  
26 position in the ROI - give overestimations or underestimations and associated false positive or



1 false negative. The enhanced selection of US signals based on their US characteristics may  
2 contribute to decreases in the influence of US artefacts.

3 The ICC between the two CAP methods showed a perfect in vivo agreement, which confirms  
4 that the two methods measure the same US attenuation parameter. [The Bland Altman results](#)  
5 [show that the new CAP method is on average slightly lower than CAP with standard method.](#)  
6 [The automatic ROI selection according to the subcutaneous thickness and the selection of the](#)  
7 [valid US signals may explain this trend.](#)

8 The linear regression fits between the standard and the new method versus MRI-PDFF are  
9 slightly different in the two study cohorts. This variation may be attributed to differences in the  
10 MRI-PDFF measurement methods used in the two study cohorts and to the significantly  
11 different BMI distributions in the two study cohorts. The good agreement found between the  
12 standard CAP method and the continuous CAP method is supported by using the exact same  
13 core algorithm to compute the individual US attenuation values. A good agreement between  
14 both methods is important to ensure that the published CAP cut-offs obtained with the standard  
15 CAP method can be used with the continuous CAP method. Interestingly, as the relationship  
16 between CAP expressed in dB/m and MRI-PDFF expressed in percentage is logarithmic, a  
17 relative decrease in MRI-PDFF would result in an absolute decrease in CAP value. For an  
18 example, given the coefficients obtained using the regression (table 2), a relative decrease of  
19 30% in MRI-PDFF corresponds to a decrease of 14 to 18 dB/m in CAP value.

20 A limitation of this study is that the new method was evaluated retrospectively by reprocessing  
21 the raw data recorded during examinations which were performed using a FS device with the  
22 standard CAP method. Future studies will help assess the performances of the new method  
23 when the operator is provided with the actual real-time information relative to the collection of  
24 US attenuation measurements. Another limitation is the use of MRI-PDFF as a reference instead  
25 of histopathology. Indeed, MRI-PDFF results may be influenced by several factors including  
26 the MRI device model, the calculation method (Liver MultiScan for study cohort A and clinical

1 procedure for study cohort B), the size of the ROI and the manual selection of the MRI-PDFF  
2 ROI by the radiologist (Campo, et al. 2017). Some studies assessed the accuracy and  
3 reproducibility of MRI-PDFF for multivendor MRI on phantoms (Hernando, et al. 2017,  
4 Hayashi, et al. 2018). With a standard reconstruction algorithm, the relative error is inversely  
5 proportional to the true fat fraction of phantom. The error is about 20% for low fat fraction and  
6 about 5% for high fat fraction. For study cohort A, the delay between FS examination and MRI-  
7 PDFF examination can reach several weeks and may compromise the relevance of the results  
8 because of the progression of steatosis. With the uncertainties on the etiology of the steatosis,  
9 these MRI-PDFF limitations could explain the inconsistency of MRI-PDFF distribution  
10 between the two study cohorts. Lastly, in this study no artificial body wall between the FS and  
11 the phantom material has been included as we used a single element US transducer with a small  
12 aperture of approximately 1 cm<sup>2</sup>. However, it will be of interest to analyze the impact of the  
13 artificial body wall on the CAP measurements.

## 14 **Conclusion**

15 In this study, a new method for CAP measurement was successfully validated on US attenuation  
16 reference phantoms and retrospectively on in vivo data from two study cohorts. The continuous  
17 CAP method significantly improves the precision of US attenuation assessment. Furthermore,  
18 the new method demonstrated higher performances in terms of hepatic steatosis quantification  
19 when using MRI-PDFF as a reference and a better correlation with MRI-PDFF. The new  
20 method is implemented in FS devices with only minor changes in the operation of the  
21 examination. The differences include the introduction of a specific gage to reflect the number  
22 of US attenuation measurements, and the use of the mean and the SD instead of median and  
23 IQR, respectively. A perfect agreement was found between both methods, indicating that the  
24 cut-offs defined for CAP in the literature are applicable to CAP measurements performed with  
25 the new method. CAP measured using the continuous CAP method is a promising tool and a  
26 reliable alternative or complement the standard CAP method for diagnosing and monitoring

1 hepatic steatosis during longitudinal follow-up of patients with chronic liver disease. These  
2 preliminary results should be confirmed in a larger prospective study.

### 3 **Acknowledgements**

4 The authors are very grateful to the research biomedical engineer Jennifer Whitehead, as well  
5 as the expert pathologist Dr. Angelo H. Paredes, from Brooke army medical center.

6 The authors would like to thank Laura A. Kehoe for her help in improving the English of this  
7 paper.

### 8 **Conflicts of interest**

9 Stéphane Audière, Aymeric Labourdette, Céline Fournier, Laurent Sandrin and Véronique  
10 Miette are Echosens employees. Stephen A. Harrison is a consultant of Echosens. Redouane  
11 Ternifi and Salem Boussida received support from Echosens. All other authors declare no  
12 conflicts of interest.

13

14

## 1 **Reference List**

- 2 Audière S, Charbit M, Angelini ED, Oudry J, Sandrin L. 2010 Measurement of the skin-liver capsule  
3 distance on ultrasound RF data for 1D transient elastography. *International Conference on*  
4 *Medical Image Computing and Computer-Assisted Intervention: Springer*, 34-41.
- 5 Audière S, Clet M, Sasso M, Sandrin L, Miette V. 2013 Influence of heterogeneities on ultrasound  
6 attenuation for liver steatosis evaluation (CAP™): relevance of a liver guidance tool. *IEEE*  
7 *International Ultrasonics Symposium (IUS): IEEE*, 401-04.
- 8 Bannas P, Kramer H, Hernando D, Agni R, Cunningham AM, Mandal R, Motosugi U, Sharma SD,  
9 Munoz del Rio A, Fernandez L, Reeder SB. Quantitative magnetic resonance imaging of hepatic  
10 steatosis: Validation in ex vivo human livers. *Hepatology* 2015;62:1444-55.
- 11 Bende F, Sporea I, Şirli R, Bâldea V, Lazăr A, Lupuşoru R, Fofiu R, Popescu A. Ultrasound-Guided  
12 Attenuation Parameter (UGAP) for the quantification of liver steatosis using the Controlled  
13 Attenuation Parameter (CAP) as the reference method. *Medical Ultrasonography* 2020;23:7-14.
- 14 Bensamoun SF, Wang L, Robert L, Charleux F, Latrive JP, Ho Ba Tho MC. Measurement of liver  
15 stiffness with two imaging techniques: magnetic resonance elastography and ultrasound  
16 elastometry. *Journal of Magnetic Resonance Imaging: An Official Journal of the International*  
17 *Society for Magnetic Resonance in Medicine* 2008;28:1287-92.
- 18 Bravo AA, Sheth SG, Chopra S. Liver biopsy. *New England Journal of Medicine* 2001;344:495-500.
- 19 Bussels B, Goethals L, Feron M, Bielen D, Dymarkowski S, Suetens P, Haustermans K. Respiration-  
20 induced movement of the upper abdominal organs: a pitfall for the three-dimensional conformal  
21 radiation treatment of pancreatic cancer. *Radiother Oncol* 2003;68:69-74.
- 22 Campo CA, Hernando D, Schubert T, Bookwalter CA, Pay AJV, Reeder SB. Standardized approach for  
23 ROI-based measurements of proton density fat fraction and R2\* in the liver. *American Journal*  
24 *of Roentgenology* 2017;209:592-603.

1 Cardoso AC, Beaugrand M, de Ledinghen V, Douvin C, Poupon R, Trinchet J-C, Ziol M, Bedossa P,  
2 Marcellin P. Diagnostic performance of controlled attenuation parameter for predicting steatosis  
3 grade in chronic hepatitis B. *Annals of hepatology* 2016;14:826-36.

4 de Lédighen V, Hiriart J-B, Vergniol J, Merrouche W, Bedossa P, Paradis V. Controlled attenuation  
5 parameter (CAP) with the XL probe of the Fibroscan®: a comparative study with the M probe  
6 and liver biopsy. *Digestive diseases and sciences* 2017;62:2569-77.

7 de Lédighen V, Vergniol J, Capdepon M, Chermak F, Hiriart J-B, Cassinotto C, Merrouche W,  
8 Foucher J. Controlled attenuation parameter (CAP) for the diagnosis of steatosis: a prospective  
9 study of 5323 examinations. *Journal of hepatology* 2014;60:1026-31.

10 de Lédighen V, Wong GLH, Vergniol J, Chan HLY, Hiriart JB, Chan AWH, Chermak F, Choi PCL,  
11 Foucher J, Chan CKM. Controlled attenuation parameter for the diagnosis of steatosis in non-  
12 alcoholic fatty liver disease. *Journal of gastroenterology and hepatology* 2016;31:848-55.

13 Eddowes PJ, Sasso M, Allison M, Tsochatzis E, Anstee QM, Sheridan D, Guha IN, Cobbold JF, Deeks  
14 JJ, Paradis V. Accuracy of FibroScan controlled attenuation parameter and liver stiffness  
15 measurement in assessing steatosis and fibrosis in patients with nonalcoholic fatty liver disease.  
16 *Gastroenterology* 2019;156:1717-30.

17 Elizabeth LY, Golshan S, Harlow KE, Angeles JE, Durelle J, Goyal NP, Newton KP, Sawh MC, Hooker  
18 J, Sy EZ. Prevalence of nonalcoholic fatty liver disease in children with obesity. *The Journal of*  
19 *pediatrics* 2019;207:64-70.

20 Ferraioli G, Lissandrin R, Zicchetti M, Filice C. Assessment of liver stiffness with transient elastography  
21 by using S and M probes in healthy children. *European journal of pediatrics* 2012;171:1415-15.

22 Ferraioli G, Maiocchi L, Raciti MV, Tinelli C, De Silvestri A, Nichetti M, De Cata P, Rondanelli M,  
23 Chiovato L, Calliada F. Detection of liver steatosis with a novel ultrasound-based technique: a  
24 pilot study using MRI-derived proton density fat fraction as the gold standard. *Clinical and*  
25 *translational gastroenterology* 2019;10:1-8.

1 Ferraioli G, Maiocchi L, Savietto G, Tinelli C, Nichetti M, Rondanelli M, Calliada F, Preda L, Filice C.  
2 Performance of the Attenuation Imaging Technology in the Detection of Liver Steatosis. *Journal*  
3 *of Ultrasound in Medicine* 2020;40:1325-32.

4 Ferraioli G, Tinelli C, Lissandrin R, Zicchetti M, Rondanelli M, Perani G, Bernuzzi S, Salvaneschi L,  
5 Filice C. Interobserver reproducibility of the controlled attenuation parameter (CAP) for  
6 quantifying liver steatosis. *Hepatology international* 2014;8:576-81.

7 Fleiss JL, Levin B, Paik MC. *Statistical methods for rates and proportions*: John Wiley & Sons, 2013.

8 Fujiwara Y, Kuroda H, Abe T, Ishida K, Oguri T, Noguchi S, Sugai T, Kamiyama N, Takikawa Y. The  
9 B-Mode Image-Guided Ultrasound Attenuation Parameter Accurately Detects Hepatic Steatosis  
10 in Chronic Liver Disease. *Ultrasound Med Biol* 2018;44:2223-32.

11 Gaitini D, Baruch Y, Ghersin E, Veitsman E, Kerner H, Shalem B, Yaniv G, Sarfaty C, Azhari H.  
12 Feasibility study of ultrasonic fatty liver biopsy: texture vs. attenuation and backscatter.  
13 *Ultrasound Med Biol* 2004;30:1321-7.

14 Hardy T, McPherson S. Imaging-based assessment of steatosis, inflammation and fibrosis in NAFLD.  
15 *Current Hepatology Reports* 2017;16:298-307.

16 Harrison SA, Gawrieh S, Roberts K, Lisanti CJ, Schwoppe RB, Cebe KM, Paradis V, Bedossa P, Aldridge  
17 Whitehead JM, Labourdette A, Miette V, Neubauer S, Fournier C, Paredes AH, Alkhoury N.  
18 Prospective evaluation of the prevalence of non-alcoholic fatty liver disease and steatohepatitis  
19 in a large middle-aged US cohort. *J Hepatol* 2021.

20 Hayashi T, Fukuzawa K, Yamazaki H, Konno T, Miyati T, Kotoku Ji, Oba H, Kondo H, Toyoda K,  
21 Saitoh S. Multicenter, multivendor phantom study to validate proton density fat fraction and  
22 T2\* values calculated using vendor-provided 6-point DIXON methods. *Clinical imaging*  
23 2018;51:38-42.

24 Hernando D, Sharma SD, Aliyari Ghasabeh M, Alvis BD, Arora SS, Hamilton G, Pan L, Shaffer JM,  
25 Sofue K, Szeverenyi NM. Multisite, multivendor validation of the accuracy and reproducibility  
26 of proton- density fat- fraction quantification at 1.5 T and 3T using a fat–water phantom.  
27 *Magnetic resonance in medicine* 2017;77:1516-24.

- 1 Iijima H. 2018 Assessment of non-alcoholic fatty liver disease with Attenuation Imaging (ATI).  
2 *VISIONS n°30: CANON MEDICAL SYSTEMS // ULEU170064.*
- 3 Kanayama Y, Kamiyama N, Maruyama K, Sumino Y. Real-time ultrasound attenuation imaging of  
4 diffuse fatty liver disease. *Ultrasound Med Biol* 2013;39:692-705.
- 5 Karlas T, Petroff D, Sasso M, Fan JG, Mi YQ, de Ledinghen V, Kumar M, Lupsor-Platon M, Han KH,  
6 Cardoso AC, Ferraioli G, Chan WK, Wong VW, Myers RP, Chayama K, Friedrich-Rust M,  
7 Beaugrand M, Shen F, Hiriart JB, Sarin SK, Badea R, Jung KS, Marcellin P, Filice C, Mahadeva  
8 S, Wong GL, Crotty P, Masaki K, Bojunga J, Bedossa P, Keim V, Wiegand J. Individual patient  
9 data meta-analysis of controlled attenuation parameter (CAP) technology for assessing steatosis.  
10 *J Hepatol* 2017;66:1022-30.
- 11 Koizumi Y, Hirooka M, Tamaki N, Yada N, Nakashima O, Izumi N, Kudo M, Hiasa Y. New diagnostic  
12 technique to evaluate hepatic steatosis using the attenuation coefficient on ultrasound B mode.  
13 *PloS one* 2019;14:e0221548.
- 14 Labyed Y, Milkowski A. Novel Method for Ultrasound- Derived Fat Fraction Using an Integrated  
15 Phantom. *Journal of Ultrasound in Medicine* 2020;39:2427-38.
- 16 Lu ZF, Zagzebski JA, Lee FT. Ultrasound backscatter and attenuation in human liver with diffuse  
17 disease. *Ultrasound Med Biol* 1999;25:1047-54.
- 18 Ma J. Dixon techniques for water and fat imaging. *J Magn Reson Imaging* 2008;28:543-58.
- 19 Madsen EL, Zagzebski JA, Frank GR. Oil-in-gelatin dispersions for use as ultrasonically tissue-  
20 mimicking materials. *Ultrasound in Medicine and Biology* 1982;8:277-87.
- 21 Mendes LC, Ferreira PA, Miotto N, Zanaga L, Lazarini MS, Goncales ESL, Pedro MN, Goncales FLJ,  
22 Stucchi RSB, Vigani AG. Controlled attenuation parameter for steatosis grading in chronic  
23 hepatitis C compared with digital morphometric analysis of liver biopsy: impact of individual  
24 elastography measurement quality. *Eur J Gastroenterol Hepatol* 2018;30:959-66.
- 25 Naveau S, Voican CS, Lebrun A, Gaillard M, Lamouri K, Njike-Nakseu M, Courie R, Tranchart H,  
26 Balian A, Prevot S, Dagher I, Perlemuter G. Controlled attenuation parameter for diagnosing

1 steatosis in bariatric surgery candidates with suspected nonalcoholic fatty liver disease. *Eur J*  
2 *Gastroenterol Hepatol* 2017;29:1022-30.

3 Paul J, Venugopal RV, Peter L, Shetty KNK, Shetti MP. Measurement of Controlled Attenuation  
4 Parameter: A Surrogate Marker of Hepatic Steatosis in Patients of Nonalcoholic Fatty Liver  
5 Disease on Lifestyle Modification - a Prospective Follow-up Study. *Arq Gastroenterol*  
6 2018;55:7-13.

7 Petroff D, Blank V, Newsome PN, Voican CS, Thiele M, de Lédinghen V, Baumeler S, Chan WK,  
8 Perlemuter G, Cardoso A-C. Assessment of hepatic steatosis by controlled attenuation  
9 parameter using the M and XL probes: an individual patient data meta-analysis. *The Lancet*  
10 *Gastroenterology & Hepatology* 2021;6:185-98.

11 Qayyum A, Nystrom M, Noworolski SM, Chu P, Mohanty A, Merriman R. MRI steatosis grading:  
12 development and initial validation of a color mapping system. *AJR Am J Roentgenol*  
13 2012;198:582-8.

14 Ratziu V, Charlotte F, Heurtier A, Gombert S, Giral P, Bruckert E, Grimaldi A, Capron F, Poynard T,  
15 Group LS. Sampling variability of liver biopsy in nonalcoholic fatty liver disease.  
16 *Gastroenterology* 2005;128:1898-906.

17 Raunig DL, McShane LM, Pennello G, Gatsonis C, Carson PL, Voyvodic JT, Wahl RL, Kurland BF,  
18 Schwarz AJ, Gonen M, Zahlmann G, Kondratovich MV, O'Donnell K, Petrick N, Cole PE,  
19 Garra B, Sullivan DC, Group QTPW. Quantitative imaging biomarkers: a review of statistical  
20 methods for technical performance assessment. *Stat Methods Med Res* 2015;24:27-67.

21 Recio E, Cifuentes C, Macías J, Mira JA, Parra-Sánchez M, Rivero-Juárez A, Almeida C, Pineda JA,  
22 Neukam K. Interobserver concordance in controlled attenuation parameter measurement, a  
23 novel tool for the assessment of hepatic steatosis on the basis of transient elastography.  
24 *European Journal of Gastroenterology & Hepatology* 2013;25:905-11.

25 Reeder SB, Hu HH, Sirlin CB. Proton density fat-fraction: a standardized MR-based biomarker of tissue  
26 fat concentration. *J Magn Reson Imaging* 2012;36:1011-4.



- 1 Sandrin L, Fourquet B, Hasquenoph JM, Yon S, Fournier C, Mal F, Christidis C, Ziol M, Poulet B,  
2 Kazemi F, Beaugrand M, Palau R. Transient elastography: a new noninvasive method for  
3 assessment of hepatic fibrosis. *Ultrasound Med Biol* 2003;29:1705-13.
- 4 Sasso M, Audiere S, Kemgang A, Gaouar F, Corpechot C, Chazouilleres O, Fournier C, Golsztejn O,  
5 Prince S, Menu Y, Sandrin L, Miette V. Liver Steatosis Assessed by Controlled Attenuation  
6 Parameter (CAP) Measured with the XL Probe of the FibroScan: A Pilot Study Assessing  
7 Diagnostic Accuracy. *Ultrasound Med Biol* 2016;42:92-103.
- 8 Sasso M, Beaugrand M, De Ledinghen V, Douvin C, Marcellin P, Poupon R, Sandrin L, Miette V.  
9 Controlled attenuation parameter (CAP): a novel VCTE™ guided ultrasonic attenuation  
10 measurement for the evaluation of hepatic steatosis: preliminary study and validation in a cohort  
11 of patients with chronic liver disease from various causes. *Ultrasound in medicine & biology*  
12 2010;36:1825-35.
- 13 Sasso M, Miette V, Sandrin L, Beaugrand M. The controlled attenuation parameter (CAP): A novel tool  
14 for the non-invasive evaluation of steatosis using Fibroscan®. *Clinics and research in*  
15 *hepatology and gastroenterology* 2012;36:13-20.
- 16 Shahin Merat M, Rasoul Sotoudehmanesh M, Mehdi Nouraie M, Masoumeh Peikan-Heirati M, Sadaf  
17 G, Reza Malekzadeh M, Sotoudeh M. Sampling error in histopathology findings of nonalcoholic  
18 fatty liver disease: a post mortem liver histology study. *Archives of Iranian medicine*  
19 2012;15:418.
- 20 Shen F, Zheng RD, Shi JP, Mi YQ, Chen GF, Hu X, Liu YG, Wang XY, Pan Q, Chen GY, Chen JN,  
21 Xu L, Zhang RN, Xu LM, Fan JG. Impact of skin capsular distance on the performance of  
22 controlled attenuation parameter in patients with chronic liver disease. *Liver Int* 2015;35:2392-  
23 400.
- 24 Tamaki N, Koizumi Y, Hirooka M, Yada N, Takada H, Nakashima O, Kudo M, Hiasa Y, Izumi N.  
25 Novel quantitative assessment system of liver steatosis using a newly developed attenuation  
26 measurement method. *Hepatology Research* 2018;48:812-28.

1 Tang A, Desai A, Hamilton G, Wolfson T, Gamst A, Lam J, Clark L, Hooker J, Chavez T, Ang BD.  
2 Accuracy of MR imaging–estimated proton density fat fraction for classification of  
3 dichotomized histologic steatosis grades in nonalcoholic fatty liver disease. *Radiology*  
4 2014;274:416-25.

5 Tang A, Tan J, Sun M, Hamilton G, Bydder M, Wolfson T, Gamst AC, Middleton M, Brunt EM,  
6 Loomba R, Lavine JE, Schwimmer JB, Sirlin CB. Nonalcoholic fatty liver disease: MR imaging  
7 of liver proton density fat fraction to assess hepatic steatosis. *Radiology* 2013;267:422-31.

8 Tapper EB, Castera L, Afdhal NH. FibroScan (vibration-controlled transient elastography): where does  
9 it stand in the United States practice. *Clinical Gastroenterology and Hepatology* 2015;13:27-36.

10 Ternifi R, Pouletaut P, Sasso M, Miette V, Charleux F, Bensamoun S. Improvements of Liver MR  
11 Imaging Clinical Protocols to Simultaneously Quantify Steatosis and Iron Overload. *IRBM*  
12 2018;39:219-25.

13 Thiele M, Rausch V, Fluhr G, Kjaergaard M, Piecha F, Mueller J, Straub BK, Lupsor-Platon M, De-  
14 Ledinghen V, Seitz HK, Detlefsen S, Madsen B, Krag A, Mueller S. Controlled attenuation  
15 parameter and alcoholic hepatic steatosis: Diagnostic accuracy and role of alcohol  
16 detoxification. *J Hepatol* 2018;68:1025-32.

17 Wong VW, Petta S, Hiriart JB, Camma C, Wong GL, Marra F, Vergniol J, Chan AW, Tuttolomondo A,  
18 Merrouche W, Chan HL, Le Bail B, Arena U, Craxi A, de Ledinghen V. Validity criteria for the  
19 diagnosis of fatty liver by M probe-based controlled attenuation parameter. *J Hepatol*  
20 2017;67:577-84.

21 Younossi ZM, Koenig AB, Abdelatif D, Fazel Y, Henry L, Wymer M. Global epidemiology of  
22 nonalcoholic fatty liver disease—meta- analytic assessment of prevalence, incidence, and  
23 outcomes. *Hepatology* 2016;64:73-84.

24  
25

1 **Figure Caption list**

2 **Figure 1.** Schematic representation of liver tissues sampled with (a) the standard CAP method  
3 and (b) the continuous CAP method. PCD = Probe to liver Capsule Distance.

4 **Figure 2.** Comparison of CAP measurements obtained with both the standard and continuous  
5 CAP methods with reference attenuation values on seven tissue-mimicking phantoms. The  
6 precision is represented by an error bar ( $\pm$ SD). Dash lines and equations represent the linear fit  
7 between the reference attenuation and the CAP measurement.  $R^2$  is the coefficient of  
8 determination of the fit.

9 **Figure 3.** Distribution of CAP precision (SD) obtained with the standard and continuous CAP  
10 methods using the M, S and XL probes. The significant test of the precision difference is  
11 performed with a t-test (\*\*\*) =  $p < 0.001$ ). In the box plots, the boundary of the box closest to  
12 zero indicates the 25<sup>th</sup> percentile, a black line within the box marks the median, and the  
13 boundary of the box farthest from zero indicates the 75<sup>th</sup> percentile. The length of the vertical  
14 lines above and below the box is 1.5 times the IQR. Outlier: any patient result lying outside the  
15 upper or lower whiskers.

16 **Figure 4.** CAP versus MRI-PDFF for study cohort A (a) and study cohort B (b) using the  
17 standard and continuous CAP methods. Dash lines represent the linear fit between logarithmic  
18 PDFF and CAP.  $R^2$  is the coefficient of determination of the fit. Pearson values are the results  
19 of the Pearson correlations.

20 **Figure 5.** Receiver operating curves (ROC) analysis of CAP for the detection of patients with  
21 hepatic steatosis defined by MRI-PDFF  $\geq 5\%$ . Standard CAP method (dash lines) and  
22 continuous CAP method (plain line) for study cohort A (a) and study cohort B (b).

23 **Figure 6.** Bland Altman plot of differences between standard CAP method and continuous CAP  
24 method measurements vs. the mean of the two measurements. For study cohort A (a) and study

1 cohort B (b), the mean difference (8.6 dB/m, 5.6 dB/m) and the lower and upper limits of  
2 agreement (-32.9 and 50.1 dB/m, -31.9 and 43.1 dB/m) are represented as dashed lines.

3 **Figure 7.** Distribution of CAP precision obtained with the standard and continuous CAP  
4 methods using the M and XL probes for study cohort A (a) and study cohort B (b). The IQR of  
5 the standard CAP method provided for indication only. The significant test of the precision  
6 improvement is performed with a t-test (\*\*  $p < 0.01$ , \*\*\*\*  $p < 0.0001$ ). In the box plots, the  
7 boundary of the box closest to zero indicates the 25<sup>th</sup> percentile, a black line within the box  
8 marks the median, and the boundary of the box farthest from zero indicates the 75<sup>th</sup> percentile.  
9 The length of the vertical lines above and below the box is 1.5 times the IQR. Outlier: any  
10 patient result lying outside the upper or lower whiskers.

11

12

1 **Table 1.** Characteristics of the two study cohorts.

Study cohort	A	B
<b>Patients</b>		
<b>Number of patients</b>	113	82
<b>Male</b>	44%	56%
<b>Age Mean ± SD (years)</b>	53 ± 8	55 ± 19
<b>BMI Mean ± SD (kg/m<sup>2</sup>)</b>	30 ± 5	27 ± 5
<b>Normal weight / Overweight / Obese</b>	16% / 38% / 46%	38% / 30% / 21%
<b>FS examinations</b>		
<b>M probe / XL probe</b>	69% / 31%	73% / 27%
<b>At least 10 valid measurements</b>	100%	96%
<b>At least recommended number of US attenuation measurements for continuous CAP method</b>	94%	100%
<b>PCD M &amp; XL probes Mean ± SD (mm)</b>	M: 17.4 ± 2.9 XL: 24.0 ± 4.4	M: 16.1 ± 2.9 XL: 21.8 ± 2.6
<b>Standard CAP method values Mean ± SD (dB/m)</b>	275.3 ± 52.3	264.9 ± 57.9
<b>Continuous CAP method values Mean ± SD (dB/m)</b>	266.8 ± 49.3	259.6 ± 56.4
<b>Number of US attenuation measurements Standard CAP method Mean ± SD</b>	10.2 ± 0.8	11.0 ± 3.4
<b>Number of US attenuation measurements Continuous CAP method Mean ± SD</b>	900 ± 599	1555 ± 834
<b>MRI-PDFF examinations</b>		
<b>MRI-PDFF Median [IQR] (%)</b>	2.7 [5.1]	5.83 [9.7]
<b>Delay between MRI-PDFF &amp; FS Median [IQR] (days)</b>	22 [30]	0 [0]

2 SD = standard deviation; PCD = probe to liver capsule distance; US = ultrasound; MRI-PDFF  
3 = magnetic resonance imaging - proton density fat fraction; FS = FibroScan; IQR = interquartile  
4 range.

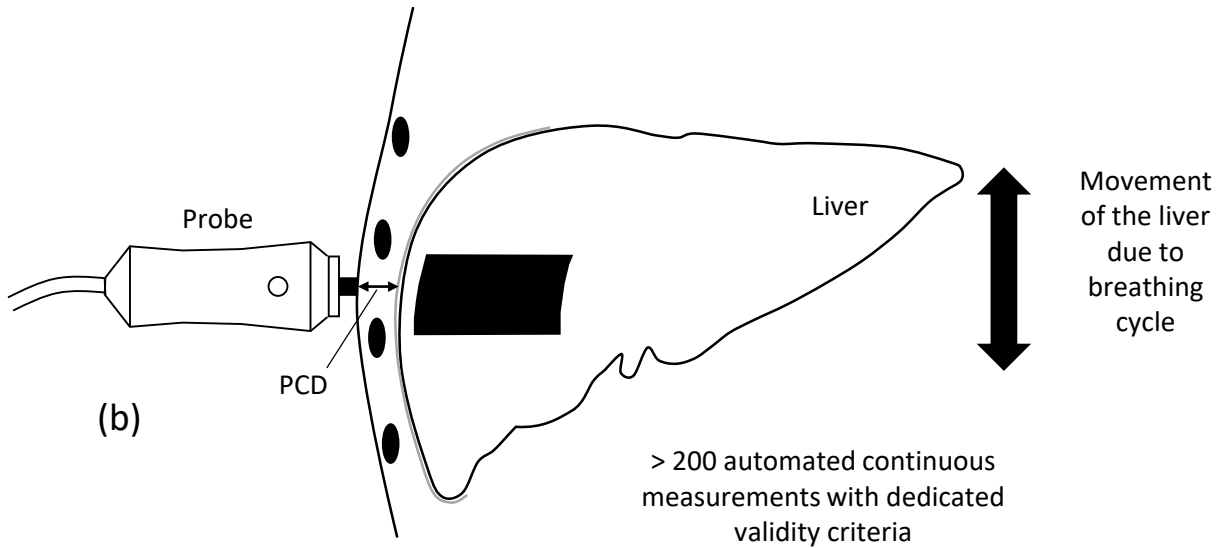
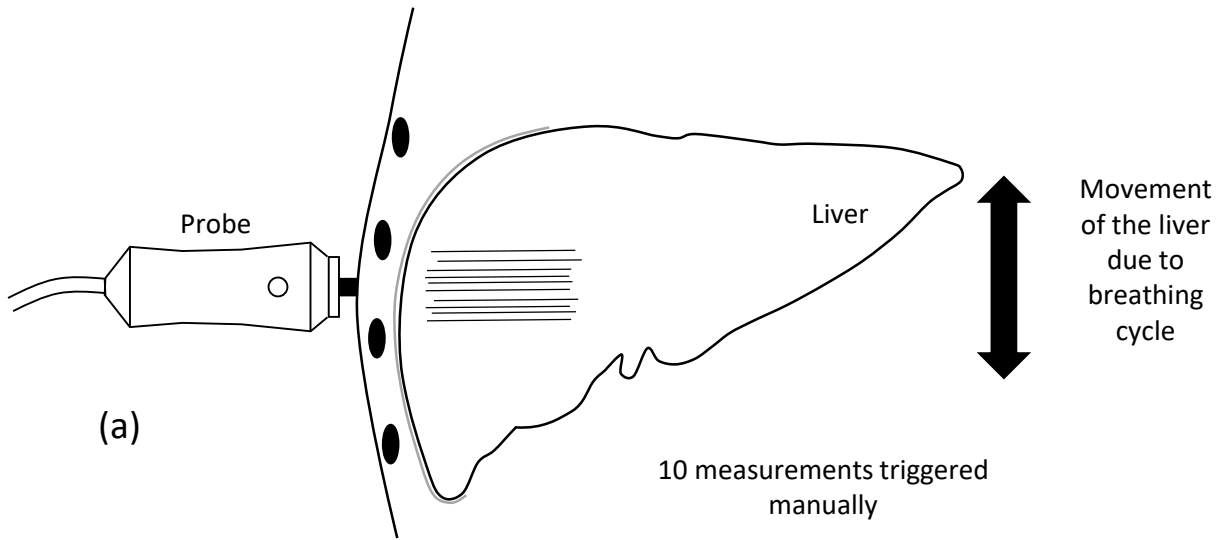
5

1 **Table 2.** Comparison of both CAP methods versus MRI-PDFF in the two study cohorts.

Study cohort		A	B
<b>Pearson correlations CAP vs log<sub>10</sub>(PDFF)</b>	<i>Standard CAP method</i>	0.71 [0.60-0.79]	0.71 [0.58-0.80]
	<i>Continuous CAP method</i>	0.74 [0.65-0.81]	0.76 [0.66-0.84]
	<i>Hittner test p-value</i>	0.22	0.02
<b>Regression CAP = a x log<sub>10</sub>(PDFF) + b</b>	<i>Standard CAP method</i>	a = 88 b = 231 (R <sup>2</sup> = 0.50)	a = 109 b = 172 (R <sup>2</sup> = 0.50)
	<i>Continuous CAP method</i>	a = 87 b = 223 (R <sup>2</sup> = 0.55)	a = 115 b = 161 (R <sup>2</sup> = 0.59)
<b>AUROC PDFF &gt; 5%</b>	<i>Prevalence</i>	32%	59%
	<i>Standard CAP method</i>	0.889 [0.827-0.953] Cutoff (Youden) = 273 dB/m	0.835 [0.745-0.924] Cutoff (Youden) = 264 dB/m
	<i>Continuous CAP method</i>	0.900 [0.838-0.961] Cutoff (Youden) = 286 dB/m	0.873 [0.798 – 0.949] Cutoff (Youden) = 256 dB/m
	<i>Delong test p-value (difference)</i>	0.55	0.06
	<i>Delong test p-value (non-inferiority with a 0.02 margin)</i>	0.02	<0.01

2 PDFF = proton density fat fraction; AUROC = area under the receiver operating characteristic,

3 R<sup>2</sup> = coefficient of determination



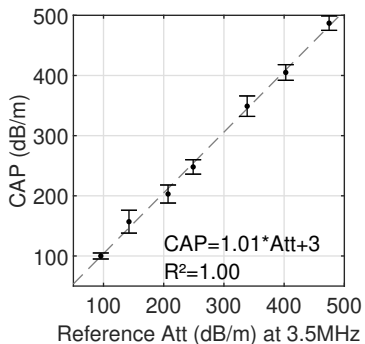
# Standard CAP method

Click here to access/continuous CAP method

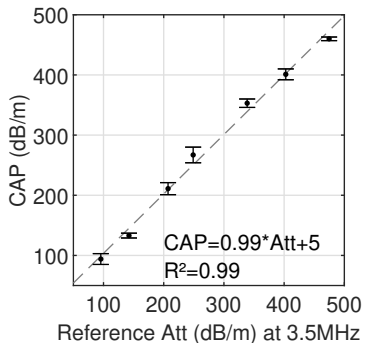
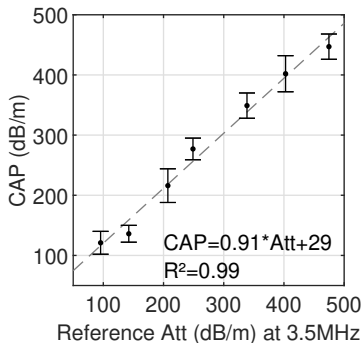


**S  
Probe**

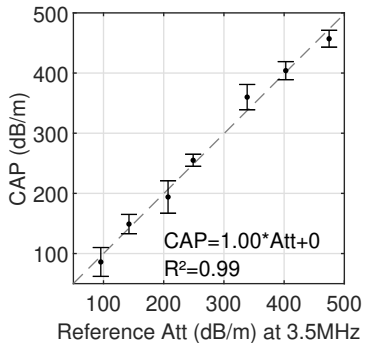
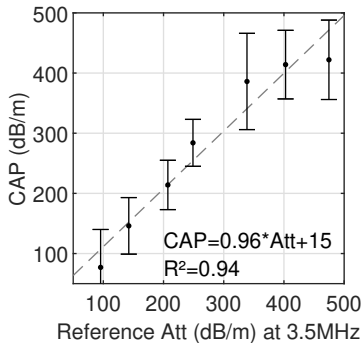
**No data**



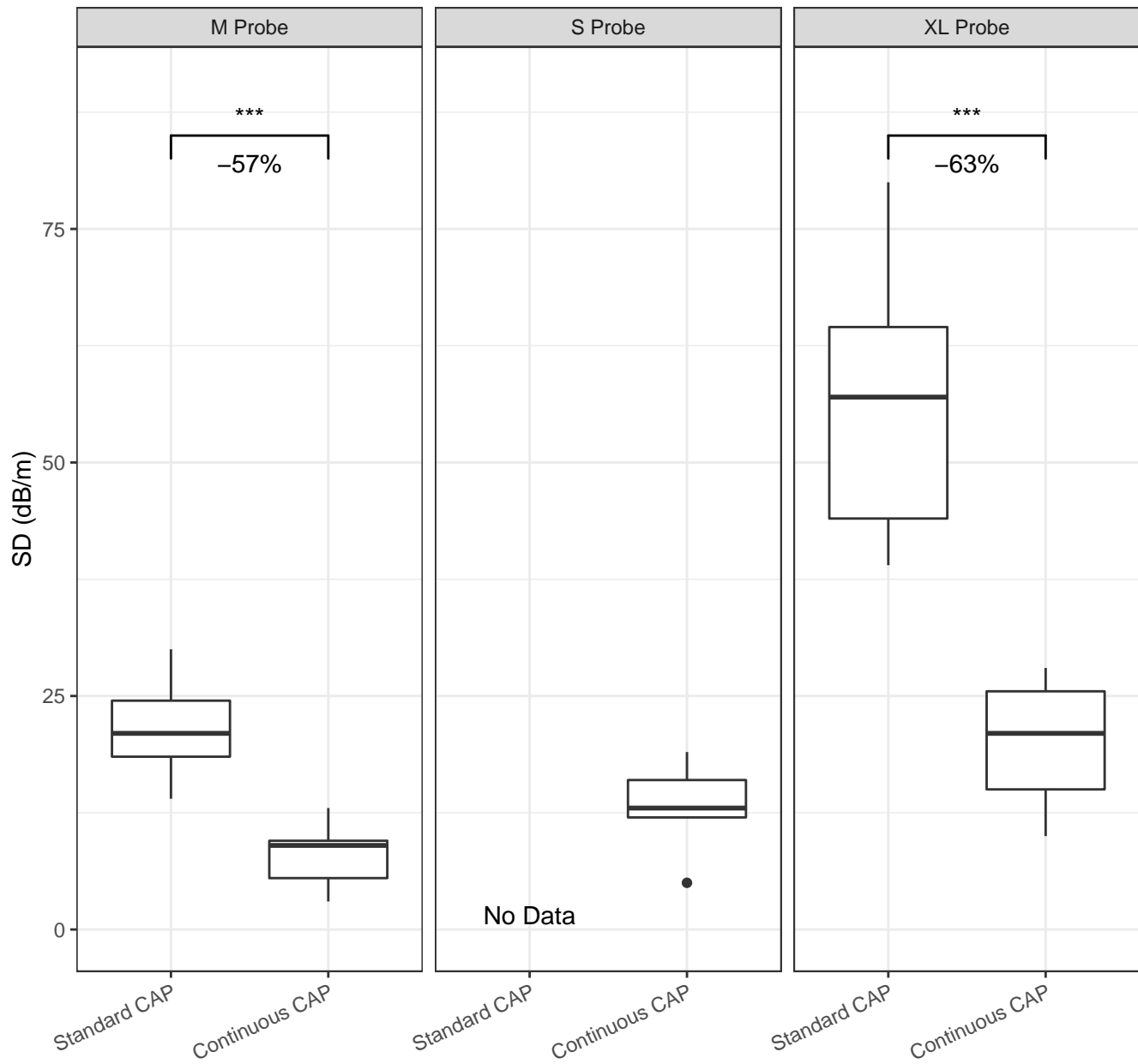
**M  
Probe**



**XL  
Probe**

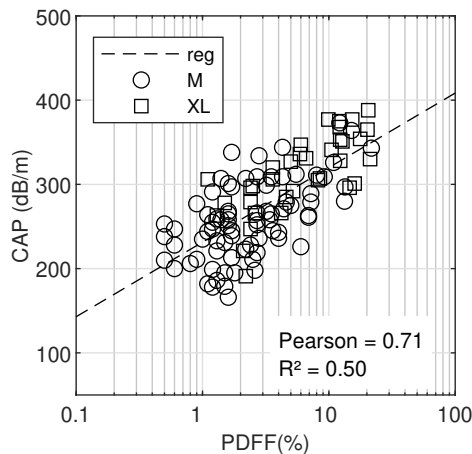






## Standard CAP method

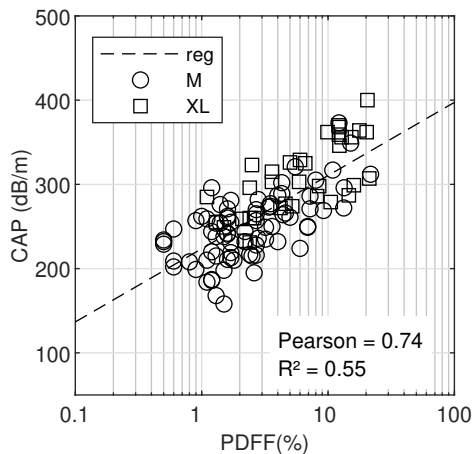
(a)



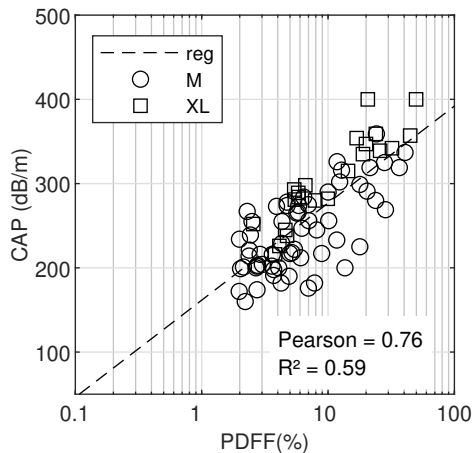
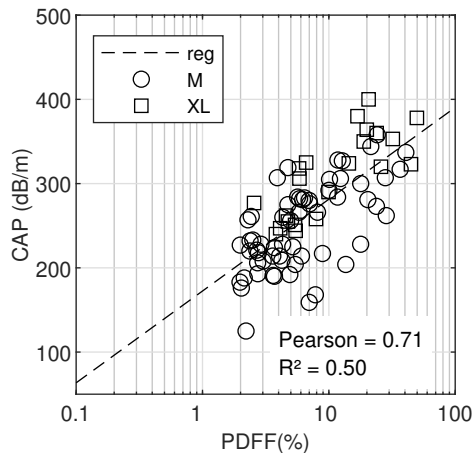
Click here to  
access/download;figure;figure\_4.pdf

## Continuous CAP method

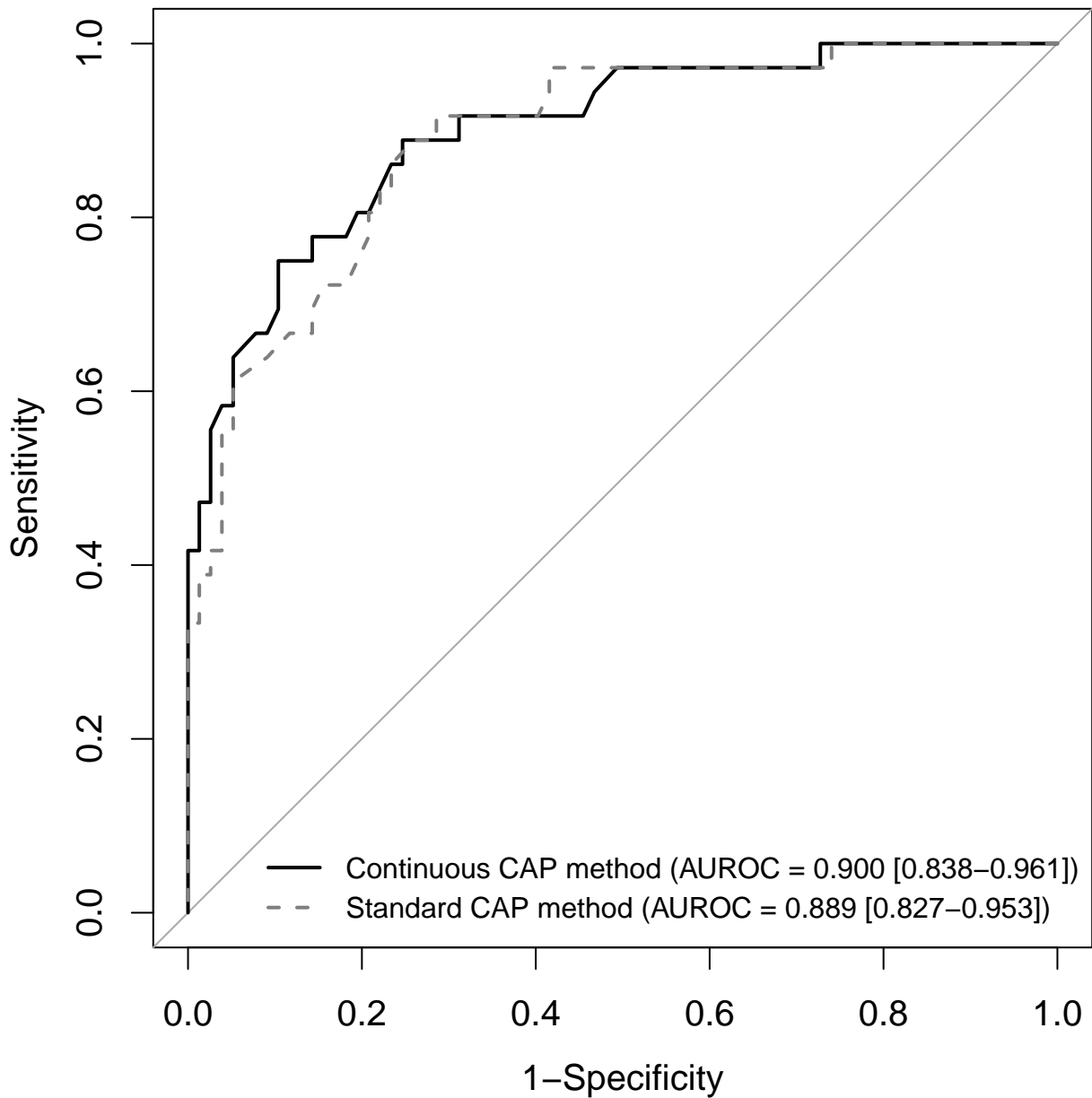
(a)



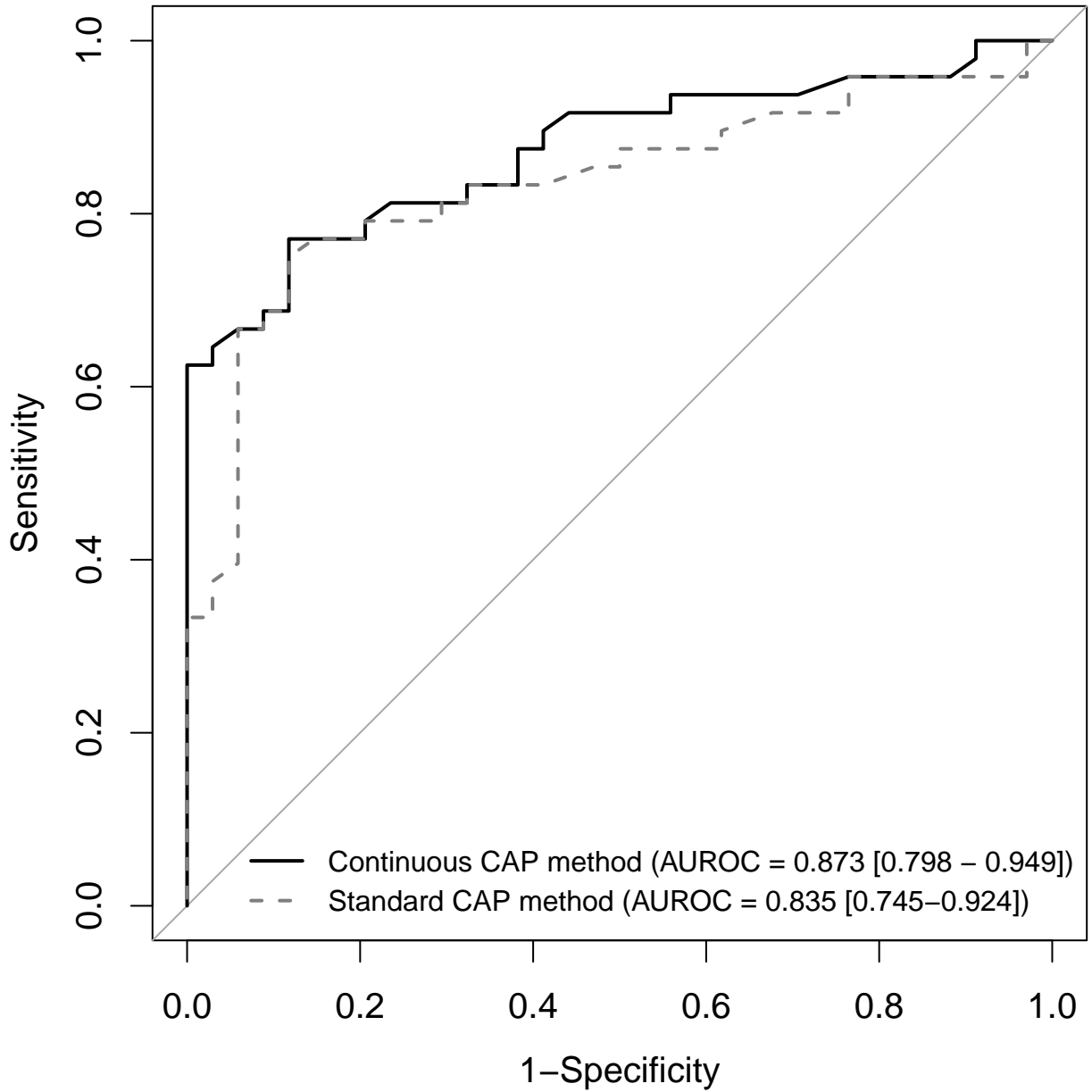
(b)



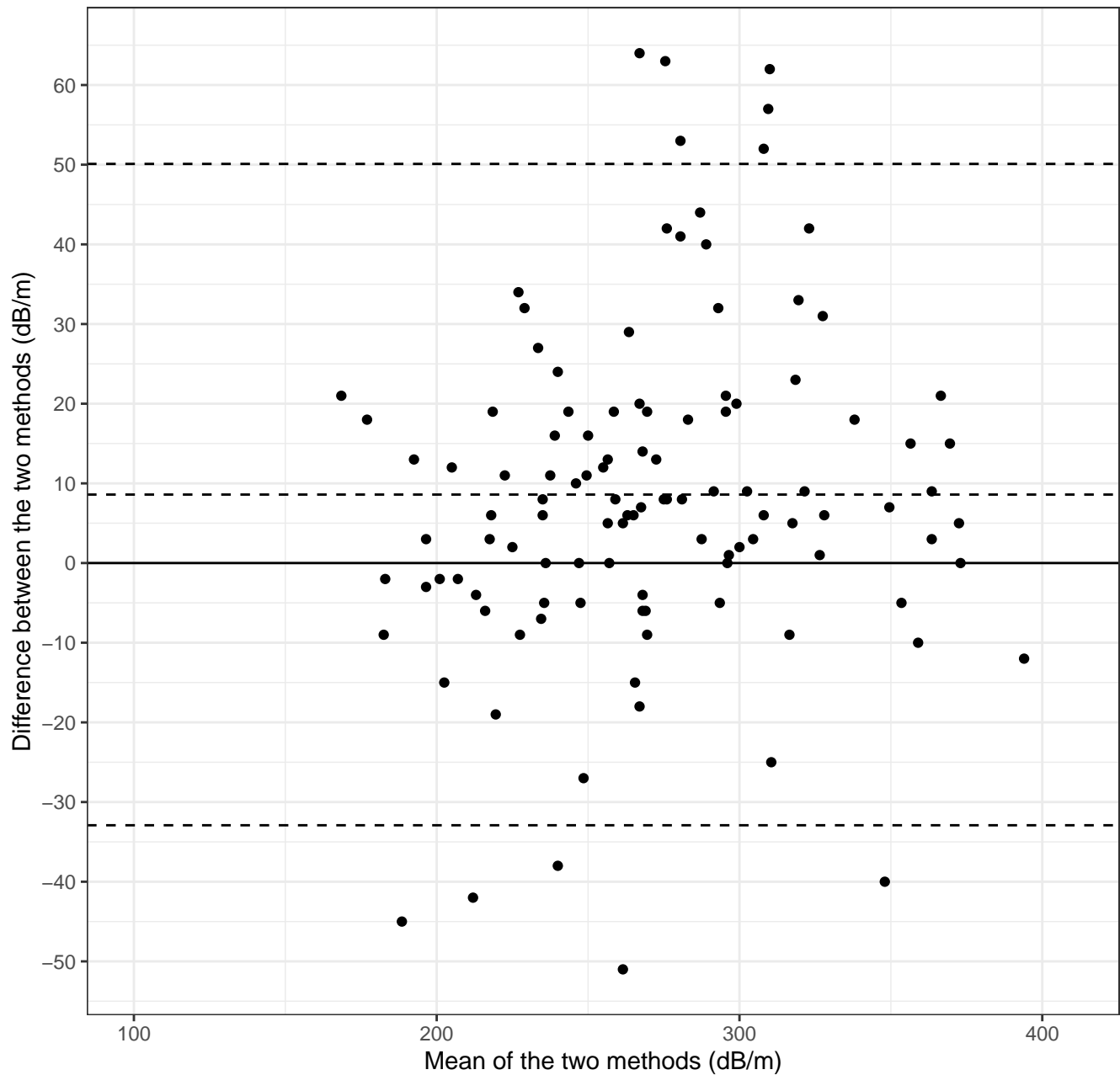
**(a)**

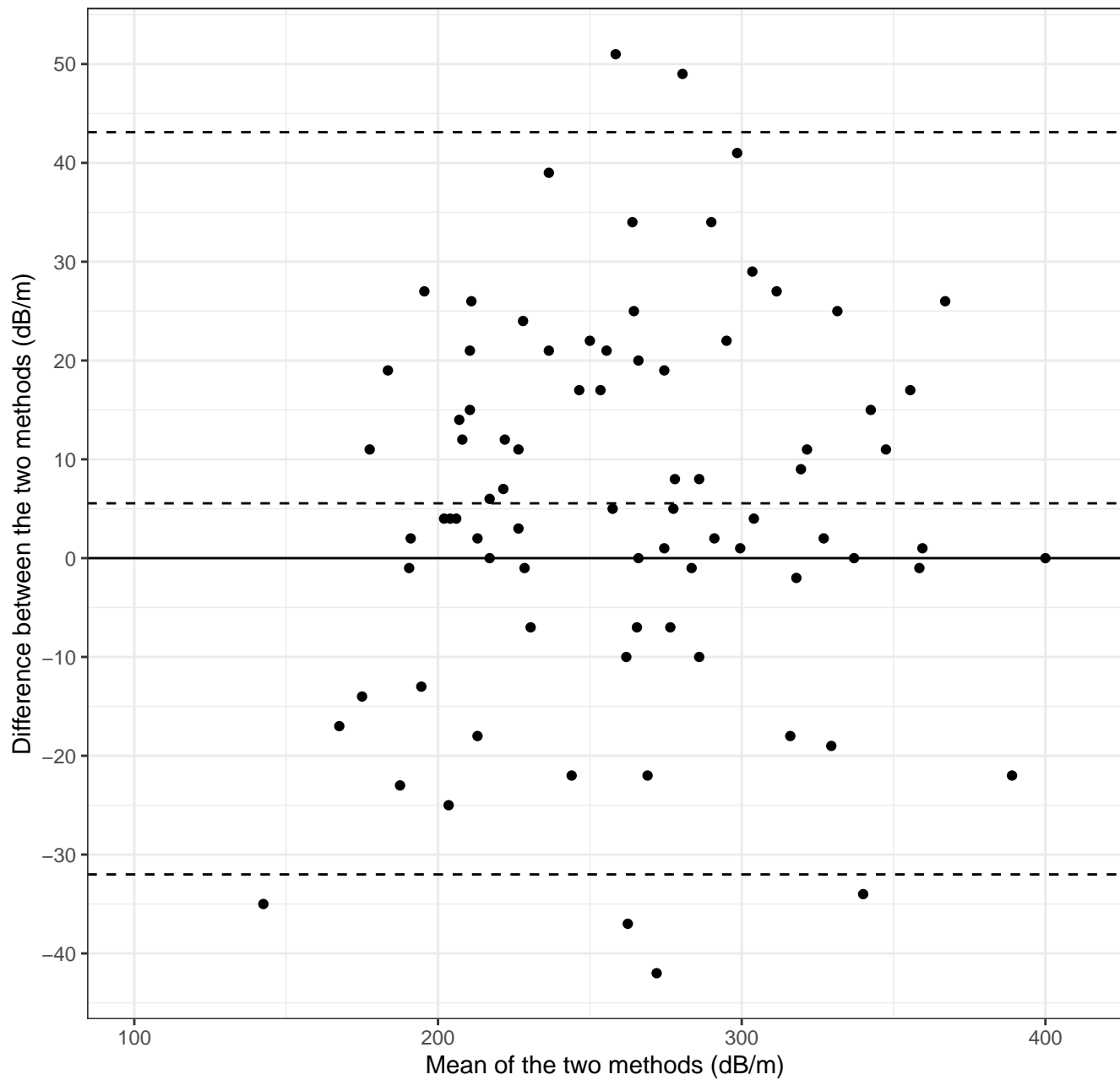


**(b)**



(a)





(a)

