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RESEARCH PAPER

Reference values and associated factors of controlled attenuation parameter and liver stiffness in adults: A cross-sectional study

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Abstract *Background & aims:* The utilization of non-invasive techniques for liver fibrosis and steatosis assessment has gained acceptance as a viable substitute for liver biopsy in clinical practice. This study aimed to establish normative data for the controlled attenuation parameter (CAP) and liver stiffness measurement (LSM) by age and gender, as well as to explore the relationship between anthropometric measures, clinical status, and biochemical profile according to the 90th percentile cut-off values for CAP/LSM in a U.S. adult population.
Methods and results: In this cross-sectional analysis, 7.522 US adults aged 20–80 years from the National Health and Nutrition Examination Survey (NHANES 2017–2020) were included. CAP and LSM were quantified using the FibroScan® 502-v2 device. A comprehensive range of data was collected, including sociodemographic, anthropometric, biochemical, lifestyle, and clinical conditions. Participants were segmented by sex and age. The median ± standard deviation (SD) for CAP was significantly lower in women (258.27 ± 61.02 dB/m) than in men (273.43 ± 63.56 dB/m), as was the median ± SD for LSM (women: 5.50 ± 4.12 kPa, men: 6.36 ± 5.63 kPa). Although median CAP and LSM values displayed an upward trend with age, statistical significance was not achieved. Notably, higher liver CAP values (above the 90th percentile) correlated with more pronounced clinical and biochemical profile differences compared to lower CAP values (below the 90th percentile) ($p < 0.001$).
Conclusions: Our study provides age- and sex-stratified standard values for CAP and LSM in a sizeable, nationally representative cohort of adults. The evidence of sex-specific variations in TE test results from our study sets the stage for future research to further corroborate these findings.

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1. Introduction

Noncirrhotic nonalcoholic fatty liver disease (NAFLD) stands as a principal contributor to hepatic morbidity globally [1]. The spectrum of NAFLD spans from benign hepatic steatosis to more severe forms, including steatohepatitis, cirrhosis, and hepatocellular carcinoma, which may culminate in liver transplantation or fatality [2]. Affecting approximately 25% of the global adult population [3], its prevalence exhibits geographical variability, documented at 10–30% across North America [4], Europe [5] and Asia [6] and 30% in South America [3]. Notably, its incidence escalates dramatically in individuals with obesity (41–61%), type 2 diabetes (18–28%), metabolic syndrome (30–56%) or hyperlipidemia (50–83%) [3]. This uptrend in liver disorders correlates strongly with the pervasive incidences of excessive weight, suboptimal dietary habits, and sedentary lifestyles, potentially progressing to hepatopathies [7]. Furthermore, NAFLD imposes significant clinical, economic, and quality-of-life burdens [8].

While Liver biopsy remains the diagnostic benchmark for NAFLD and fibrosis, its invasiveness limits its application [9]. Ultrasound energy is dissipated more rapidly in a steatotic liver. Transient elastography (TE), have emerged as favored alternatives [10]. The controlled attenuation parameter (CAP), derived via a TE probe (FibroScan®), captures the decline in the amplitude of ultrasound waves in the liver parenchyma to estimate the degree of hepatic steatosis [11]. CAP, alongside liver stiffness measurement (LSM)—indicative of fibrosis—are assessed concurrently, enhancing the diagnostic process. TE's diagnostic prowess is reflected in its high accuracy, with receiver operating characteristic (ROC) curve areas signifying its effectiveness in fibrosis detection [12,13]. Moreover, CAP's correlation with steatosis, benchmarked against biopsy, reinforces its diagnostic value [14–16]. Recent individual patient data meta-analysis of CAP technology support its used for assessing steatosis [17,18].

Ethnic variations in hepatic steatosis prevalence underscore the importance of demographic-specific interpretations [19]. Accurate steatosis and fibrosis assessments necessitate age and sex-specific benchmarks for healthy liver tissue, acknowledging the need to obviate previous selection biases and steatosis as a confounding factor [20–22]. This study's objective is to establish normative CAP and LSM data for a diverse, US cohort, devoid of liver disease, as well as to explore the relationship between anthropometric measures, clinical status, and biochemical profile according to the 90th percentile cut-off values for CAP/LSM, utilizing the National Health and Nutrition Examination Surveys (NHANES) from 2017 to 2020.

2. Methods

2.1. Design and study population

This cross-sectional analysis leveraged data from the National Health and Nutrition Examination Survey (NHANES) encompassing the years 2017 to March 2020. Field operations for the NHANES program were suspended in March 2020 due to the COVID-19 pandemic. Consequently, data from the start of 2017 up to March 2020 were amalgamated with the 2017–2020 NHANES cycle to compile a pre-pandemic, nationally representative sample, hereafter referred to as NHANES 2017-March 2020. The study secured ethical approval from the National Center for Health Statistics Research Ethics Review Board (CDC, 2016). Given the de-identified nature of the data, no further ethical clearance was required. The study methodologies were in strict adherence to the principles of the 1975 Helsinki Declaration, as revised in 2013, and written informed consent was obtained from all participants.

2.2. Participants

From the 10,409 individuals enrolled in the NHANES 2017–2020 cohort, spanning ages 0–80 years, a subset of 7522 adults aged over 20 years old was selected for percentile calculation. Exclusions applied to participants with current illness or significant medical histories at the time of the assessment (n = 89), those testing positive for HBsAg, IgG anti-HBc, anti-HCV, anti-HIV antibodies, or other similar conditions (n = 354), and participants with incomplete or partial transient elastography (TE) data (n = 282). The data management process is outlined in Fig. 1.

2.3. Instrumentation and measurements

Adhering to NHANES published guidelines and protocols [https://wwwn.cdc.gov/nchs/nhanes/], the liver tissue examination was integrated to procure comprehensive data on fatty liver prevalence in the U.S. populace and to inform public health intervention strategies. TE was conducted in the NHANES Mobile Examination Center (MEC), employing the FDA-approved FibroScan® model 502 V2 Touch, with a medium (M) or extra-large (XL) wand. Criteria for exclusion included the inability to recline on the examination table, pregnancy or uncertain pregnancy status without a confirmatory urine test, possession of an implanted electronic medical device, or the presence of a bandage or lesions on the abdomen's right side where measurements were to be taken. Assessments were conducted with participants in a supine position, estimating LSM in kilopascals (kPa) and hepatic steatosis in decibels per meter (dB/m). Only those participants who had fasted for a minimum of 3 h, produced ten or more viable LSM readings, and exhibited an interquartile range of less than 30% of the median LSM value were included in the final analysis. The inter-rater reliability for the LSM and CAP measurements, comparing health technicians with reference examiners (n = 32), was recorded at 0.86 and 0.94, respectively. To assure consistency over time within and between FibroScan® devices and probes, NHANES employed four shear wave liver fibrosis phantoms (CIRS Model 039). The interrater reliability between health technicians and expert FibroScan® technicians, tested on 32 subjects, yielded a reliability of 0.86 for stiffness (mean difference: 0.44 ± 1.3 kPa) and 0.94

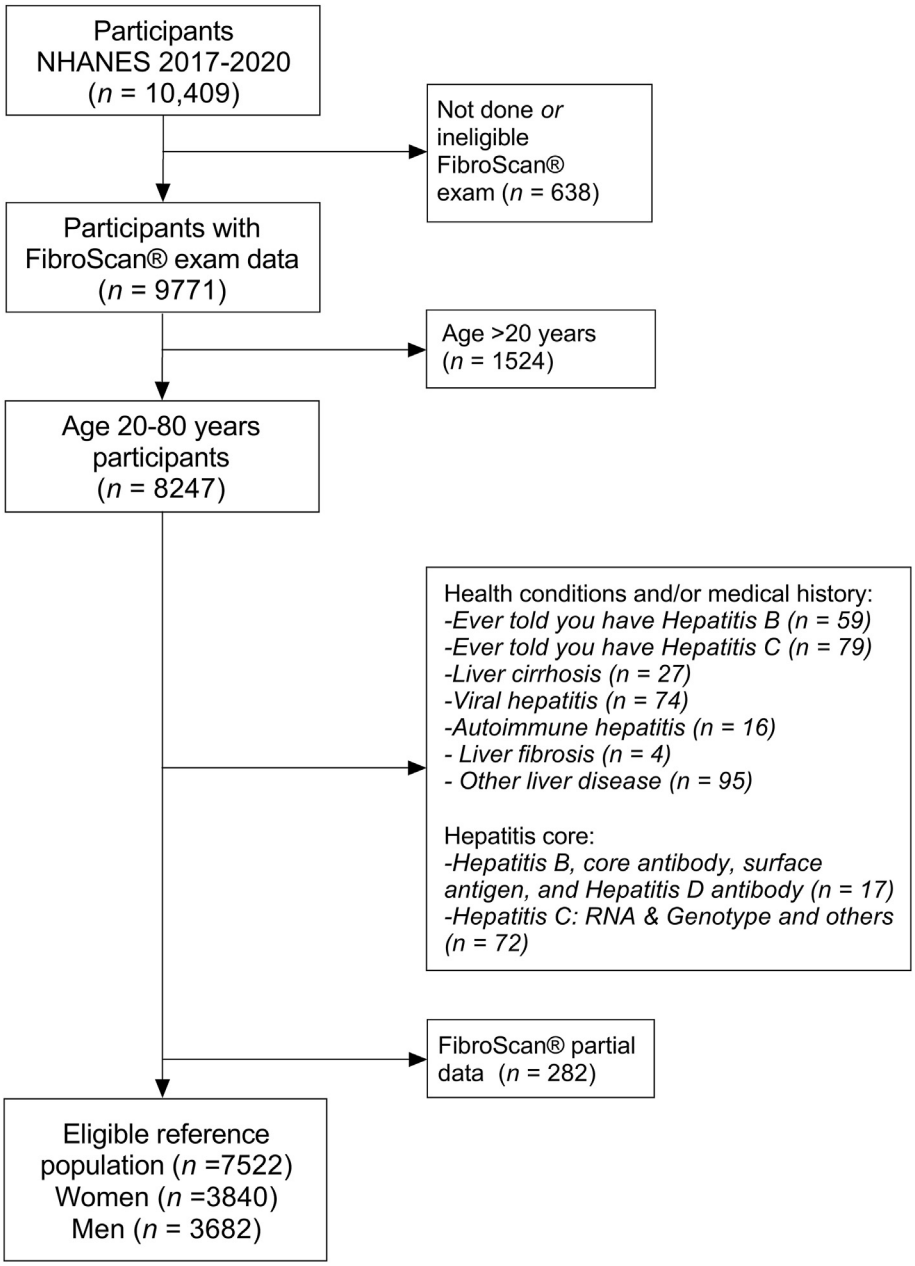

Figure 1 Flow diagram of participants aged 20–80 years from NHANES (2017–2020).

for CAP (mean difference: 4.5 ± 19.8 dB/m). For an exhaustive account of the quality assurance and control measures for this component, refer to the Procedures Manual (https://wwwn.cdc.gov/nchs/data/nhanes/2017-2018/manuals/2018.pdf).

Anthropometric measurements including body mass, height, and waist circumference were obtained by NHANES' cadre of skilled health technicians. BMI was determined using the formula: weight in kilograms divided by the square of height in meters. Participants were tested on routine cardiometabolic parameters. Resting systolic and diastolic blood pressures were measured 3–4 times with a mercury sphygmomanometer by trained staff. Triglycerides, total cholesterol, high-density lipoprotein (HDL), glucose, albumin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), γ-glutamyl transferase (GGT), and high-sensitivity C-reactive protein (hs-CRP) concentrations were measured on the Roche Cobas 6000 (c501 module) analyzer using a standard protocol by highly trained medical personnel in the MEC. The TyG index was calculated as the natural logarithm (Ln) of the product of plasma glucose and TG using the formula: Ln (TG [mg/dL] × glucose [mg/dL]/2) FAST score was calculated for each patient by inserting LSM, CAP, and AST levels in a formula provided by FibroScan®. FAST score varied on a scale from 0 to 1, with a score of 1 being associated with more advanced fibrosis.

In addition, number of hours of usual sleep on weekdays or workdays and weekends were recorded (range 3–13.5 h). The use of tobacco products (i.e., cigarettes, pipes, cigars, little cigars or cigarillos, water pipes, hookahs, or e-cigarettes) in the past 5 days and drink alcoholic beverage (in the past 12 month) were self-reported per design of NHANES survey.

Sociodemographic characteristics were all assessed by self-report during an in-home interview, such as age, sex, race/ethnicity (non-Hispanic white; non-Hispanic black; Mexican American or other Hispanic; and other, including multiracial), education (less than 9th grade 9–11th grade, high school graduate/GED or equivalent, some college or AA degree, college graduate or above, refused/don't know), and marital status (married/living with partner, widowed/divorced/separated, never married, or refused/don't know). Clinical condition data including arthritis, thyroid disease, congestive heart failure, coronary heart disease, stroke, emphysema, asthma, gallstones, and cancer (all types), were all self-reported by participants.

2.4. Statistical analyses

The statistical analysis comprised both descriptive statistics elements (median, standard deviation [SD], and 95% confidence interval [CI]) and inferential statistics. The normality for selected variables was verified using histograms and Q-Q plots. Student's *t*-test was used to compare the distribution of continuous variables, such as the anthropometric/clinic measurements and blood biochemistry by sex group. Chi-square tests were performed to compare the distribution of categorical variables, such as race/ethnicity, marital status, education level, and comorbidities by sex group.

CAP and LSM median values were then used to generate sex- and age-specific normative centiles in LMSchartmaker Pro (V.2.43, The Institute of Child Health, London, UK), which analyzes data using the Lambda Mu Sigma (LMS) method [23]. Under the LMSchartmaker Pro protocol, curve modelling involves selecting the appropriate age scale and choosing the proper equivalent degrees of freedom values to optimize the L, M and S curves. Deviance measure is the benchmark for model fitting and curves optimization: the smaller the deviance measure, the better the optimization of the L, M and S curves. As a deviance measure, this study followed the authors' recommendations, and therefore Schwarz Bayesian Criterion (SBC) was used as a deviance measure for model fitting [24]. Following software protocol [24], percentile curves for the 3rd (P3), 10th (P10), 50th (P50), 90th (P90), and 97th (P97) were chosen as age- and sex-specific reference values that can be expressed in terms of smoothing parameters or equivalent *df* [24]. The 90th percentile was defined as upper limit of the reference interval.

To determine the strength of the relationship of the different parameters, a hierarchical linear regression analysis was performed between the sociodemographic data, anthropometric, biochemical and CAP or LSM parameters. Finally, an ANCOVA was used to investigate whether clinical characteristics differed by CAP and LMS median values group by applying the 90th percentile cut point in both gender and age groups, controlling for age, ethnicity/race origin, education level, BMI, and ratio of family income to poverty. A p-value <0.05 was considered significant. All statistical analyses were two-tailed, with a significance level of 0.05. We performed all analyses using SPSS statistical package version 26 (IBM Inc., Chicago, IL). The data were analyzed anonymously.

3. Results

Table 1 delineates the sex-specific descriptive statistics. Notably, men exhibited significantly higher measurements for body mass, height, BMI, waist circumference, and a

Table 1 Clinical, cardiometabolic, liver ultrasound parameters and demographic characteristics of the study population by sex.

Characteristics	Women (n = 3840)					Men (n = 3682)					*P* value
	N	Mean	SD	95% CI		N	Mean	SD	95% CI		
Anthropometric											
Age (years)	3840	50.23	17.19	49.69	50.78	3682	50.50	17.70	49.93	51.07	0.511
Body mass (kg)	3817	78.42	22.66	77.70	79.14	3649	88.97	22.00	88.26	89.69	<0.001
Height (cm)	3814	1.60	0.07	1.60	1.60	3648	1.74	0.08	1.73	1.74	<0.001
Body mass index (kg/m^2)	3812	30.53	8.20	30.27	30.79	3640	29.38	6.47	29.17	29.59	<0.001
Waist circumference (cm)	3694	99.24	17.62	98.67	99.81	3551	102.30	16.32	101.76	102.84	<0.001
Cardiometabolic parameters											
Total cholesterol (mg/dL)	3603	189.66	39.49	188.37	190.95	3413	183.38	41.11	182.00	184.75	<0.001
Triglycerides (mg/dL)	3583	123.13	66.64	120.95	125.31	3352	142.95	85.19	140.07	145.84	<0.001
HDL cholesterol (mg/dL)	3618	57.89	16.35	57.36	58.42	3435	48.67	13.56	48.21	49.12	<0.001
LDL cholesterol (mg/dL)	1784	109.59	34.87	107.97	111.21	1678	108.65	35.76	106.94	110.37	0.435
Glucose (mg/dL)	1826	110.60	35.16	108.98	112.21	1726	116.36	40.59	114.45	118.28	<0.001
Insulin (IU/mL)	1787	14.45	21.39	13.46	15.45	1689	14.61	21.44	13.59	15.63	0.828
HOMA-IR	1753	3.55	3.04	3.41	3.70	1653	3.64	3.23	3.48	3.79	0.427
Triglyceride-glucose (TyG) index	3601	4.63	0.30	4.62	4.64	3413	4.73	0.35	4.71	4.74	<0.001
Alanine aminotransferase (IU/L)	3602	17.98	12.46	17.57	18.38	3412	25.77	17.75	25.18	26.37	<0.001
Aspartate aminotransferase (IU/L)	3592	19.60	10.91	19.24	19.96	3395	23.17	12.44	22.75	23.58	<0.001
γ-Glutamyl transferase (IU/L)	3602	25.43	33.18	24.34	26.51	3412	37.15	63.22	35.03	39.28	<0.001
AST-ATL ratio	3592	1.22	0.40	1.21	1.24	3395	1.04	0.40	1.02	1.05	<0.001

Table 1 (*continued*)

Characteristics	Women (n = 3840)					Men (n = 3682)					*P* value
	N	Mean	SD	95% CI		N	Mean	SD	95% CI		
Ferritin (ng/mL)	3644	102.99	130.34	98.76	107.22	3455	214.31	113.26	207.20	221.43	<0.001
hsC-reactive protein (mg/L)	3592	4.48	7.03	4.25	4.71	3403	3.34	6.41	3.12	3.55	<0.001
Fatty liver index (FLI) score	3493	65.02	38.47	63.74	66.29	3354	73.20	35.91	71.98	74.41	<0.001
FibroScan-AST (FAST) score	3486	0.09	0.12	0.08	0.09	3371	0.14	0.16	0.14	0.15	<0.001
Systolic blood pressure (mmHg)	3433	122.19	20.51	121.50	122.88	3383	126.89	16.89	126.32	127.46	<0.001
Diastolic blood pressure (mmHg)	3433	73.94	11.37	73.56	74.32	3383	75.60	11.42	75.22	75.99	<0.001
Liver ultrasound transient elastography											
Median LSM, (kPa)	3840	5.50	4.12	5.37	5.63	3682	6.36	5.63	6.18	6.54	<0.001
Stiffness E interquartile range (IQR)	3840	0.93	1.88	0.87	0.99	3682	1.10	2.75	1.02	1.19	0.001
Median CAP (dB/m)	3840	258.27	61.02	256.34	260.20	3682	273.43	63.56	271.38	275.49	<0.001
CAP interquartile range (IQR)	3840	36.79	19.13	36.18	37.39	3682	37.55	21.15	36.86	38.23	0.102
Ratio: LSM IQR/median LSM	3840	15.76	24.63	14.98	16.54	3682	15.07	14.90	14.59	15.55	0.146
Lifestyles											
Sleep hours - weekdays or workdays	3811	7.67	1.65	7.62	7.72	3649	7.42	1.63	7.37	7.47	<0.001
Sleep hours - weekends	3810	8.39	1.77	8.33	8.45	3644	8.10	1.79	8.04	8.15	<0.001
Past 12 m how often drink alcoholic beverage	3147	5.32	3.94	5.19	5.46	3318	4.54	4.06	4.40	4.68	<0.001
Smoked tobacco last 5 days? (Yes), N (%)	609	(16.9)				929	(26.3)				0.001
Education level, N (%)											
Less than 9th grade	265	(6.9)				301	(8.2)				0.001
9-11th grade (Includes 12th grade with no diploma)	379	(9.9)				430	(11.7)				
High school graduate/GED or equivalent	885	(23.0)				917	(24.9)				
Some college or AA degree	1329	(34.6)				1109	(30.1)				
College graduate or above	977	(25.4)				920	(25.0)				
Refused/Don't Know	5	(0.2)				5	(0.1)				
Marital status, N (%)											
Married/Living with Partner	2034	(53.0)				2349	(63.8)				0.002
Widowed/Divorced/Separated	1046	(27.2)				598	(16.2)				
Never married	754	(19.6)				733	(19.9)				
Refused/Don't Know	6	(0.1)				2	(0.1)				
Race/ethnicities, N (%)											
Mexican American	450	(11.7)				445	(12.1)				0.672
Other Hispanic	412	(10.7)				369	(10.0)				
Non-Hispanic white	1285	(33.5)				1287	(35.0)				
Non-Hispanic black	1039	(27.1)				954	(25.9)				
Other Race - Including multi-racial	654	(17.0)				627	(17.0)				
Clinical conditions (self-reported, yes), N (%)											
Arthritis	1282	(33.4)				909	(24.7)				0.001
Thyroid	649	(16.9)				203	(5.5)				
Congestive heart failure	82	(2.1)				122	(3.3)				
Coronary heart disease	74	(1.9)				218	(5.9)				
Stroke	163	(4.2)				174	(4.7)				
Emphysema	332	(8.6)				288	(7.8)				
Asthma	676	(17.6)				490	(13.3)				
Gallstones	558	(14.5)				210	(5.7)				
Cancer (all types)	401	(10.4)				349	(9.5)				

Values are expressed as mean, standard deviation (SD) and 95% confidence interval (95% CI); or numbers (%) when appropriate, HOMA-IR (Homeostatic Model Assessment for Insulin Resistance.

majority of cardiometabolic risk factors—including triglycerides, glucose, TyG index, liver enzymes, and blood pressure—compared to women ($p < 0.01$). Conversely, women recorded lower levels of total cholesterol, HDL cholesterol, and incidences of recent tobacco use ($p < 0.001$). Additionally, women showed a higher prevalence of arthritis, thyroid disorders, and gallstones, whereas men were more affected by congestive heart failure and coronary artery disease.

Age- and sex-specific reference values for CAP (dB/m) and LSM (kPa) are illustrated in Fig. 2's percentile curves (P10, P50, P90) and further detailed in Supplemental Fig. S1's boxplots. These reference values, along with the coefficients of variation (λ, μ, σ), are systematically tabulated in Tables 2 and 3 for distinct age brackets and sexes.

The percentile distributions for CAP reveal a consistent pattern across sexes, with a peak in the 40–49 and 50–59 age groups, then diminishing in the median values for women aged 60–69 and men aged 50–59. Median CAP ranges from 243.18 to 272.35 dB/m in women and 264.55–282.83 dB/m in men, with the 90th percentile extending from 312.17 to 354.09 dB/m in women and

334.37–367.07 dB/m in men (Fig. 2[a]). LSM trends disclose gender-specific variances; women's P50 steadily climbs across all age groups, while men's LSM surges to a peak at ages 60–69, then stabilizes (Fig. 2b).

The univariate regression analysis, encompassing ages 20 to 80 with comprehensive data, assessed both sexes for independent correlates of CAP and LSM. Notably, CAP and LSM did not correlate significantly with age but did with each other ($p < 0.001$), as depicted in Supplemental Fig. S2.

Note that we used the entire population with a TE measurement ($n = 7522$). In both groups, these thresholds were used to categorize individuals into two risk categories (i.e., low and high risk) based on the combined criteria of sex group and combined LSM and CAP median values. In both sexes, we found that increasing liver CAP values (>90th percentile) were associated with higher levels of ALT, AST, γ-GGT, ALT/AST ratio, HOMA-IR, ferritin, triglycerides, TyG index, FLI and FAST score compared to participants with lower values (<90th percentile), $p < 0.001$ (Fig. 3). As shown in Fig. 4, after adjusting for age, ethnicity/race origin, education level, BMI, and ratio of family income to poverty only, triglycerides levels were significantly associated with higher LMS values (>90th percentile) in women ($p < 0.01$).

4. Discussion

While CAP is an established tool for gauging liver fat content, normative references for CAP and LSM within healthy, multi-ethnic adult populations are scarce. Our data is particularly valuable for clinical application, potentially enhancing the detection of at-risk individuals for liver steatosis through population-wide TE-based preventative programs. Our findings are multifaceted. Primarily, we observed that CAP and LSM metrics were notably higher in men than in women. Additionally, median CAP and LSM values exhibited an upward trend with age, yet without statistically significant differences. A crucial observation is that higher CAP values (exceeding

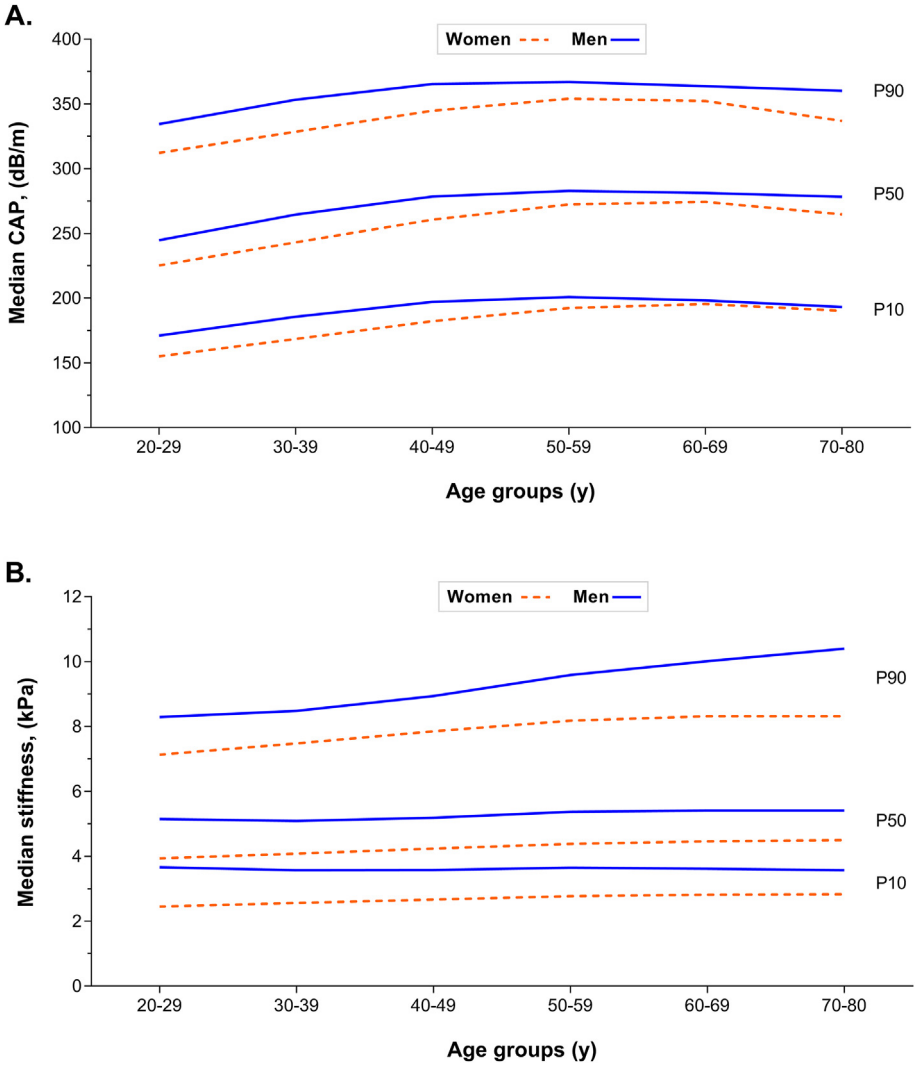

Figure 2 Smoothed centile curves (P10, P50 and P90) for median CAP and median LSM of participants aged 20–80 years from NHANES (2017–2020). Note: The line indicates the average values. The error band indicates 10 to 90 percentile for each age group.

Table 2 Centiles by age and sex based on median CAP (dB/m) of adults aged 20–80 years from NHANES (2017–2020).

Sex/age group	n	L	S	P3	P10	P25	P50 (M)	P75	P90	P97
Women (3840)										
20–29	581	0.38	0.26	125.93	155.11	188.14	225.21	266.50	312.17	362.41
30–39	628	0.59	0.25	135.56	168.57	204.48	243.18	284.58	328.62	375.22
40–49	623	0.78	0.23	145.61	182.32	220.74	260.69	302.05	344.70	388.54
50–59	710	0.93	0.22	153.29	192.40	232.12	272.35	313.02	354.09	395.52
60–69	701	1.03	0.21	155.73	195.50	235.01	274.31	313.42	352.37	391.18
70–80	597	1.11	0.21	151.69	190.20	227.83	264.76	301.11	336.97	372.39
Men (3682)										
20–29	587	0.41	0.25	139.95	171.11	206.02	244.78	287.53	334.37	385.41
30–39	560	0.63	0.24	150.28	185.70	223.83	264.55	307.74	353.31	389.19
40–49	604	0.79	0.23	158.74	197.06	237.04	278.51	321.33	365.40	391.63
50–59	591	0.91	0.22	160.96	200.87	241.53	282.83	324.70	367.07	393.89
60–69	699	1.02	0.22	156.60	198.30	239.85	281.26	322.57	363.78	386.90
70–80	641	1.13	0.22	148.88	193.19	236.25	278.32	319.60	360.21	382.24

Considering the purposes of the current study, seven percentiles (P) were constructed through the LMS method, described by Cole (3rd, 10th, 25th, 50th, 75th, 90th, and 97th) were generated. LMS method summarizes the distribution of the variable of interest according to age-groups, based on three parameters or curves: L (λ), M (μ), and S (σ). These three parameters indicate the power in the Box-Cox transformation for the skewness adjustment (L), the median (M), and the generalized coefficient of variation (S) for each median CAP measurement.

the 90th percentile) correspond with substantial differences in clinical and biochemical profiles, a pattern not mirrored in LSM values.

The cutoff of CAP is also a matter of debate. In the two meta-analyses, the optimal cutoffs to detect fatty liver were 248 dB/m and 297 dB/m using the M and XL probes, respectively [18,25]. In the same line, two recent prospective studies, the suggested cutoffs to detect state 1 steatosis were 244 and 295 dB/m separately [26,27]. This research innovates in threshold determination, incorporating liver fat content, fibrosis, and clinical priorities within percentile curves. Although this study adopted the 90th percentile cutoffs we acknowledge the selection of cut-offs is arbitrary and represents a compromise between sensitivity and specificity. For instance, the upper limit (90th percentile are 312.17–354.09 dB/m in women and 334.37–367.07 dB/m in men) for CAP values exceeds the steatosis indication threshold cited in recent literature and guidelines [28,29]. Likewise, studies from the USA consistently yielded higher optimal cutoffs of around 300 dB/m [26]. Furthermore, the median LSM ranges align with established thresholds indicative of significant fibrosis, emphasizing the utility of LSM in evaluating fibrosis severity.

In our investigation, median CAP and LSM values displayed an age-related increase, though these differences were not statistically significant. This trend contrasts with the findings of Tokuhara et al. [30], who reported no age-dependent variations in CAP values among the youth. Supporting our observations, other research has identified a rise in NAFLD prevalence with advancing age [31,32], and age as a predictor of severe fibrosis [33]. Age-specific hepatic changes, including hepatocyte enlargement, binucleation, and mitochondrial reductions, may precipitate steatosis [34]. Metabolic alterations, like increased lipid accumulation and altered lipogenesis and β-oxidation,

Table 3 Centiles by age and sex based on LSM (kPa) of adults aged 20–80 years from NHANES (2017–2020).

Sex/age group	n	L	S	P3	P10	P25	P50 (M)	P75	P90	P97
Women (3840)										
20–29	581	−0.43	0.39	2.00	2.45	3.07	3.94	5.20	7.13	10.27
30–39	628	−0.48	0.39	2.09	2.56	3.19	4.08	5.40	7.48	11.01
40–49	623	−0.53	0.39	2.20	2.67	3.32	4.24	5.62	7.85	11.78
50–59	710	−0.57	0.39	2.29	2.77	3.43	4.38	5.82	8.18	12.48
60–69	701	−0.57	0.39	2.33	2.82	3.49	4.46	5.93	8.32	12.65
70–80	597	−0.52	0.39	2.33	2.83	3.52	4.50	5.97	8.32	12.40
Men (3682)										
20–29	587	−0.83	0.30	3.18	3.66	4.29	5.15	6.38	8.29	11.59
30–39	560	−0.84	0.31	3.09	3.57	4.21	5.09	6.40	8.48	12.27
40–49	604	−0.84	0.33	3.08	3.58	4.25	5.19	6.61	8.94	13.44
50–59	591	−0.85	0.34	3.13	3.65	4.36	5.37	6.93	9.59	15.03
60–69	699	−0.86	0.36	3.09	3.62	4.35	5.41	7.07	10.01	16.47
70–80	641	−0.86	0.37	3.04	3.57	4.32	5.41	7.17	10.40	18.08

Considering the purposes of the current study, seven percentiles (P) were constructed through the LMS method, described by Cole (3rd, 10th, 25th, 50th, 75th, 90th, and 97th) were generated. LMS method summarizes the distribution of the variable of interest according to age-groups, based on three parameters or curves: L (λ), M (μ), and S (σ). These three parameters indicate the power in the Box-Cox transformation for the skewness adjustment (L), the median (M), and the generalized coefficient of variation (S) for each median LMS measurement.

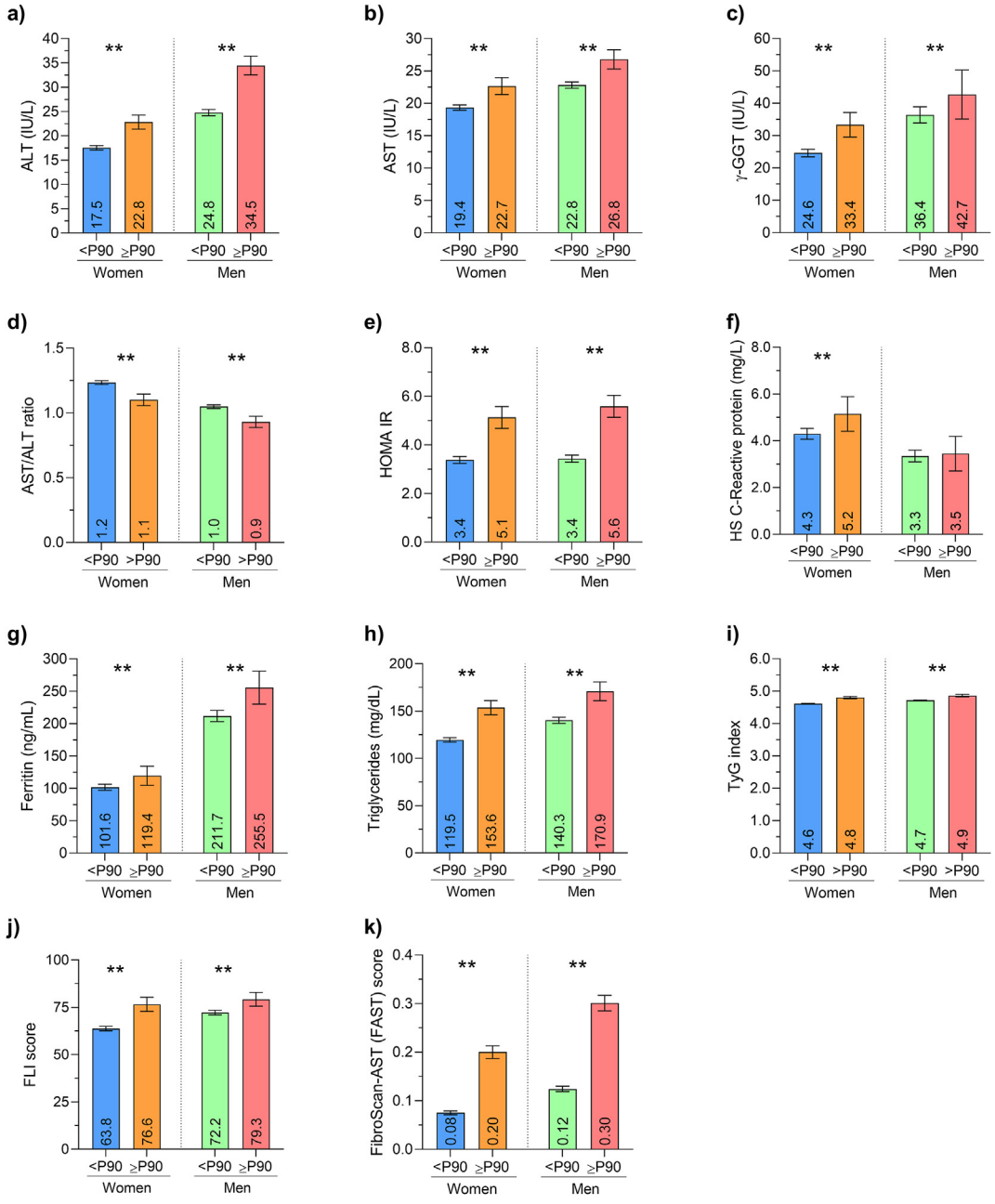

Figure 3 Adjusted age and sex thresholds for high/low (90th percentile) CAP median values for clinical and biochemical profile differences of participants aged 20–80 years from NHANES (2017–2020). Note: Controlling for age, ethnicity/race origin, education level, BMI, and ratio of family income to poverty. ** $p < 0.001$.

have been implicated in steatosis development [35]. Furthermore, sarcopenia's association with NAFLD progression is noted, marked by declining muscle mass and increased adipose and fibrous tissues [36,37].

Our data also highlight gender as a significant determinant of CAP and LSM echoing the findings from a small-scale study [38] and confirming the need for gender-specific interpretation of TE parameters. The female gender is associated with lower LSM values in various populations [39], and men have a reported higher NAFLD prevalence [3,31,32]. The mechanisms by which gender influences hepatic steatosis include links between male gender, elevated ALT levels, and abdominal obesity, all of which elevate liver disease risk [40]. This study reinforces the importance of considering both age and gender in the clinical evaluation of liver health.

Our study enriches the existing literature by offering valuable insights into the potential markers of liver fibrosis in adults presumed to be healthy, highlighting that elevated CAP values—those above the 90th percentile—are associated with increased ALT, AST, γ-GGT, ALT/AST ratio, HOMA-IR, ferritin, triglycerides, TyG index, FLI, and FAST score, in contrast to LSM values. Participants with >90th percentile CAP also higher levels of BMI, waist

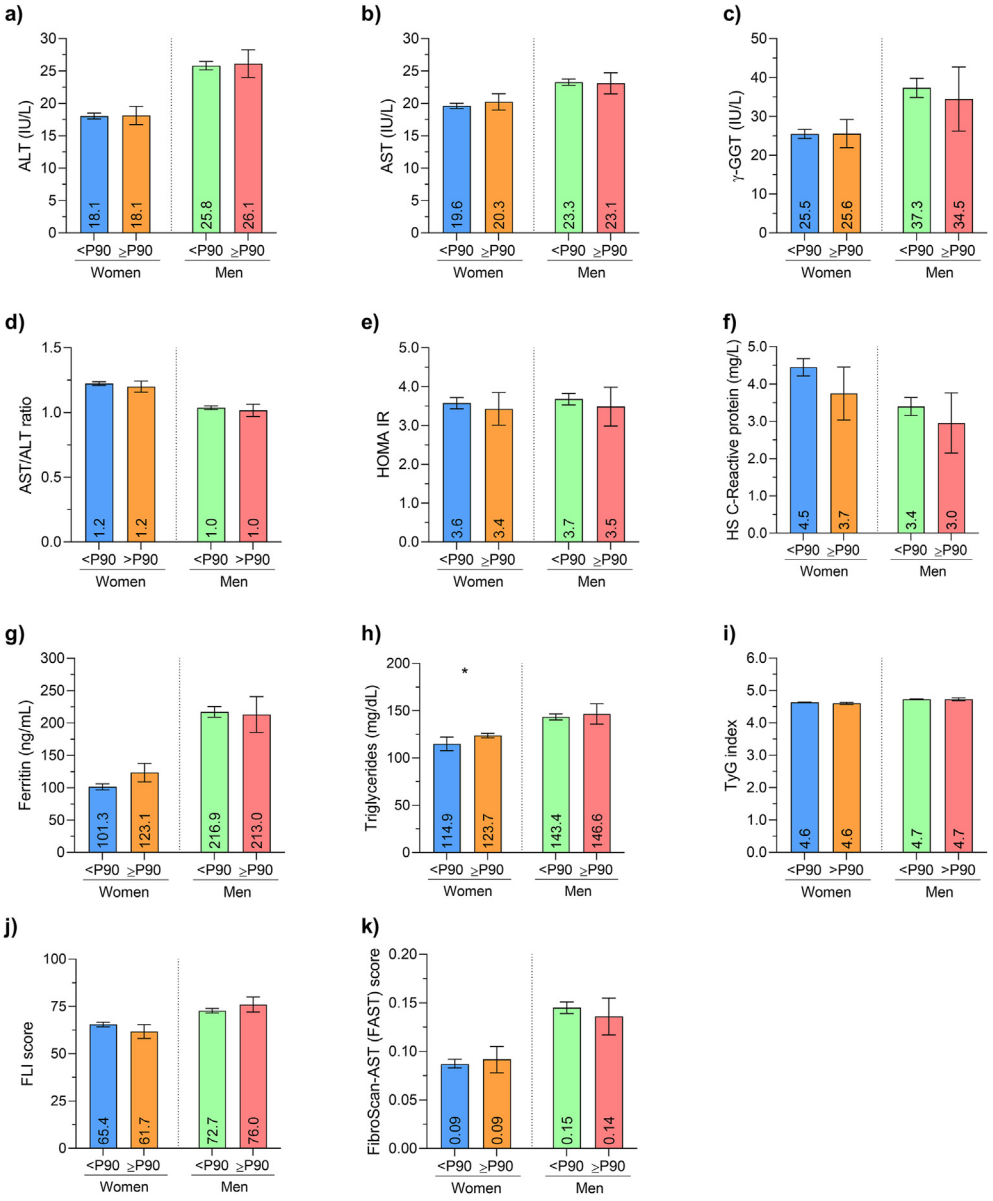

Figure 4 Adjusted age and sex thresholds for high/low (90th percentile) LMS median values for clinical and biochemical profile differences of participants aged 20–80 years from NHANES (2017–2020). Note: Controlling for age, ethnicity/race origin, education level, BMI, and ratio of family income to poverty. $* p < 0.01$.

circumference, waist-to-height ratio and blood pressure compared to the rest of the cohort, in both sexes, $p < 0.001$ (Supplemental Fig. S3). This association persists independently of traditional risk factors and supports the link between NAFLD and heightened cardiovascular event risk [41]. No significant association were observed for the LSM values and any pre-risk factors for liver diseases (data no shown).

Despite the reported findings, the main limitation of the study is the failure to consider NAFLD risk factors in the generation of the percentile curves. Applying the effect of risk factors could result in improved accuracy. However, given the cross-sectional nature of this study, causality cannot be inferred, and the absence of biopsy-verified fibrosis in our cohort remains a limitation. Another limitation is the extrapolation of results to other populations. The development and application of percentile curves should consider the geographic area and ethnicity of the study population. Percentile curves for different geographic regions are needed to define global strategies for NAFLD prevention. Lastly, the lack of detailed lifestyle data could influence the outcomes. Despite these limitations, our study's strengths lie in providing age- and sex-specific reference values for CAP and LSM in a multi-ethnic population from the USA, which stands to greatly benefit clinical liver function assessment and allow for its standardized procedures, lends further weight to our findings [42].

In summary, our research offers normative values for CAP and LSM that are stratified by age and sex, using a

large and nationally representative sample of healthy adults. With the increasing global prevalence of obesity, these TE component reference values could prove crucial for identifying individuals at an elevated risk of NAFLD and its progression and its cost-effectiveness compared with other modalities to develop optimal strategies for the screening of NAFLD are need. Notably, our results underscore the sex-dependent nature of TE test responses, a finding that we anticipate will be elaborated upon in future investigations.

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Author disclosure statement

No competing financial interests exist.

Authors' contributions

All authors read and approved the final version of the manuscript. Robinson Ramírez-Vélez contributed to the conception and study design and reviewed/edited manuscript. María Correa-Rodriguez analyzed and interpreted the data and wrote the manuscript. Antonio García-Hermoso analyzed the data, performed statistical analyses and reviewed/edited manuscript. Mikel Izquierdo contributed to the conception, study design, data interpretation and reviewed/edited manuscript.

Ethical conduct of research

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. Written informed consent was obtained for all participants.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.numecd.2024.04.004.

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