Transient Elastographic Evaluation of Subjects Without Known Hepatic Pathology: Does Age Change the Liver Stiffness?

Roxana Şirli, Ioan Sporea, Adriana Tudora, Alexandra Deleanu, Alina Popescu

Department of Gastroenterology and Hepatology, “Victor Babeş” University of Medicine and Pharmacy Timișoara, Romania

Abstract

Background and aim: Chronic liver diseases are investigated through invasive (liver biopsy) or non-invasive (FibroScan or FibroTest) methods. Recently the non-invasive methods have become more and more popular. The aim of this paper is to evaluate the liver stiffness (LS) measured by transient elastography in individuals without known hepatic pathology (“normal” subjects) and to see if it is influenced by age. Material and method: We examined a group of “normal” subjects by means of a FibroScan device (EchoSens, France). In each patient we performed 10 valid measurements and a median value was calculated by the device. The subjects were individuals without known hepatic pathology: healthy volunteers or patients from departments other than Gastroenterology in our hospital. Results: We evaluated 152 “normal” subjects (87 women and 65 men, mean age 45.3±17.6 years). The mean value of LS in “normal” subjects was 4.8±1.3 kPa, ranging from 2.3 to 8.8 kPa. The mean values of LS in age subgroups were: 18-29 years – 5±1.3 kPa; 30-39 years - 4.5±1.2kPa; 40-49 years – 5±1.1kPa; 50-59 years – 4.7±1.2kPa; 60-69 years – 5±1.3kPa; >70 years – 4.7±1.4kPa. There were no statistically significant differences between the mean values of LS in various age subgroups (p=0.5263). Conclusions: The mean value of LS measured by transient elastography in “normal” subjects was lower than 5 kPa. Age does not modify the LS.

Key words

Liver stiffness – transient elastography – normal subjects.

Introduction

Liver fibrosis is a very important factor associated with prognosis in chronic hepatitis (viral and non-viral). Hence, a precise evaluation of the severity of fibrosis is needed in order to perform a correct staging and to take decisions regarding the treatment.

Currently, the biopsy examination of the liver is considered to be the “gold standard” method for evaluating fibrosis [1]. However, the liver biopsy (LB) is not a perfect method due to: intra- and interobserver variability [2, 3]; sampling variability [4]; and, last, the fact that LB is an invasive method, with morbidity and mortality greater than 0.

Considering all these facts, non invasive methods for the evaluation of liver fibrosis have been developed in the last few years, in order to replace LB, among them liver stiffness (LS) evaluation by means of transient elastography (TE) using a FibroScan® device (EchoSens, Paris, France) [5, 6]. The method uses a probe with an ultrasonic transducer mounted on the axis of a vibrator. This vibrator induces a wave of mild amplitude and low frequency to the tissue. Thus, an elastic shear wave is created that propagates in the tissue and, in the meantime, a pulse-echo ultrasound is performed to follow the shear wave and measure its velocity. The propagation velocity is directly related to the tissue stiffness. The harder the tissue, the faster the shear waves propagates [7]. With this method, LS can be measured in normal individuals and in patients, the results being expressed in kiloPascals (values between 2.5 and 75 kPa).

This method was first developed in France [6], but lately, several studies from different parts of the world have been published [7-9], confirming its value.

In order to be able to use TE in clinical practice, valid measurements (VM) have to be obtained in the majority of evaluated cases. The method has to be reproducible (small inter- and intra-observer variability) and also able to establish in which domains it has the most reliable results. Last, but not least, the normal range of LS measurements must be established, in order to differentiate the normal liver from the fibrotic liver.
It has been demonstrated that age is an important factor associated with fibrosis progression in chronic hepatitis [10-12]. So, we asked ourselves if in patients without a known history of liver disease, fibrosis does not increase with age.

**Material and methods**

Our study included 152 subjects without known hepatic pathology ("normal" subjects), healthy volunteers or patients from other departments than Gastroenterology in our hospital (Nephrology, Surgery, Cardiology etc.), who agreed to participate in our study.

The healthy volunteers (109 individuals) were medical students, nurses and medical doctors (fellows and specialists) from our hospital. None of them had a history of liver disease (acute or chronic). We did not perform additional tests in this subgroup (such as biological tests, viral markers, abdominal ultrasound, etc). The hospitalized patients included in our study (43 individuals) had no history of liver disease (acute or chronic) and had normal values of aminotransferases. Also, none of them had a severe disease such as congestive heart failure, renal failure, or diabetes. Their main diagnoses included: acute pyelonephritis, appendicitis, chronic glomerulonephritis, arterial hypertension, chronic angina pectoris, etc.

In all the patients we performed LS measurement (LSM) by TE, using a FibroScan® device (EchoSens - Paris, France). Liver stiffness measurement was performed according to the classical methodology [13, 14] by three physicians who had previously performed at least 50 examinations each, considered to be sufficient for a proper training [15]. The measurements were made on patients lying in dorsal decubitus with the right arm in maximal abduction. The right lobe of the liver was aimed at, through the intercostal spaces. The tip of the probe transducer was covered with coupling gel and placed on the skin, between the ribs at the level of the right lobe of the liver. The operator, assisted by ultrasound time-motion and A-mode images provided by the system, located a portion of the liver free of large vascular structures that was at least 6 cm thick. Once the measurement area was located, the operator pressed the probe button to begin an acquisition. Ten successful acquisitions were performed in each patient.

The success rate was calculated as the ratio of the number of successful acquisitions over the total number of acquisitions. In each patient 10 VM were performed, after which a median value of the LS was obtained, measured in kPa. Only in patients in whom LSM had a success rate of at least 60%, with IQR<30%, the measurements were considered reliable (IQR = interquartile range, it is the difference between the 75th percentile and the 25th percentile, essentially the range of the middle 50% of the data). Failure was defined if 10 VMs could not be obtained with a success rate of at least 60%, with IQR<30%, as the result would not be reliable, according to the latest published data [16].

We calculated the mean value of LS for the whole group, as well as for different age groups. For a statistical study of quantitative variables, the mean and standard variations were calculated. One-way ANOVA test and t-tests were performed to compare mean values of LS in various age subgroups and in men vs. women and non-obese vs. obese patients, respectively. The statistical analysis was performed using Microsoft Excel and GraphPad Prism programs.

**Results**

We evaluated 152 “normal” subjects (87 women and 65 men, mean age 45.3±17.6 years). The age of our subjects ranged from 18 to 87 years: 28.3% (43 subjects) in the 18-29 years subgroup, 16.4% (25 subjects) in the 30-39 years subgroup, 11.2% (17 subjects) in the 40-49 years subgroup, 29.4% (31 subjects) in the 50-59 years subgroup, 14.5% (22 subjects) in the 60-69 years subgroup and 9.2% (14 subjects) in the >70 years subgroup.

In 8 cases (5.3%), we could not obtain VM. The rate of failure to obtain VM increased with the body mass index (BMI). From the 133 patients with BMI<30kg/m², failure was observed only in 1 case, but in the subgroup of 19 patients with BMI>30kg/m², failure to obtain VM occurred in 7 cases (36.8%), p<0.0001. Also, the failure rate was significantly higher in women - 7.87 (8.0%) as compared to men - 1.65 (1.5%) (p<0.0001).

In the 144 “normal” subjects, in whom VMs were obtained, the mean value of LS was 4.8±1.3 kPa, ranging from 2.3 to 8.8 kPa. The mean values of LS in each age group are presented in Table I and Fig.1.

**Table I. Mean values of liver stiffness in each age subgroup.**

<table>
<thead>
<tr>
<th>Age group (years)</th>
<th>Nr. of patients with VM</th>
<th>Mean value of LS ± SD (kPa)</th>
<th>Minimum (kPa)</th>
<th>Maximum (kPa)</th>
</tr>
</thead>
<tbody>
<tr>
<td>All patients</td>
<td>144</td>
<td>4.8±1.3</td>
<td>2.3</td>
<td>8.8</td>
</tr>
<tr>
<td>18-29</td>
<td>43</td>
<td>5±1.3</td>
<td>2.3</td>
<td>8.8</td>
</tr>
<tr>
<td>30-39</td>
<td>24</td>
<td>4.5±1.2</td>
<td>2.6</td>
<td>7.3</td>
</tr>
<tr>
<td>40-49</td>
<td>17</td>
<td>5±1.1</td>
<td>3.0</td>
<td>7.1</td>
</tr>
<tr>
<td>50-59</td>
<td>27</td>
<td>4.7±1.2</td>
<td>2.5</td>
<td>7.7</td>
</tr>
<tr>
<td>60-69</td>
<td>20</td>
<td>5±1.3</td>
<td>3.2</td>
<td>7.7</td>
</tr>
<tr>
<td>&gt;70</td>
<td>13</td>
<td>4.7±1.4</td>
<td>3.0</td>
<td>7.1</td>
</tr>
</tbody>
</table>
We performed a one-way ANOVA test in order to find out if there were statistically significant differences between the mean values of LS in various age subgroups, and we found out that the differences were not significant (p=0.5263).

The mean LS in women was 4.6±1.2 kPa, significantly lower than in men 5.1±1.2 kPa (p=0.0082) (Fig.2).

We also compared the mean value of LS in obese subjects (12 patients with BMI>30 kg/m$^2$) to that of the 132 subjects with BMI<30 kg/m$^2$, but we did not find significant differences: 5.3±1.6 kPa, vs. 4.8±1.2 kPa, p=0.3289 (Fig.3).

**Discussion**

Although TE is used more and more for staging chronic C hepatitis, its value being proven for predicting significant fibrosis (F≥2 Metavir) and cirrhosis [17, 18], as well as in chronic B hepatitis [8], cholestatic hepatitis [19] and NASH [20], few studies have been published regarding the normal range of LS in healthy patients [16].

The most comprehensive study of the normal values of LS in healthy individuals was recently published by Roulot et al [21]. It was performed on 429 consecutive apparently healthy subjects and the mean LS value was 5.49±1.59 kPa. Failure to obtain valid LSMs was observed in 4.6% of the cases, the failure rate increasing with BMI, being approximately 25% in obese patients (BMI>30 kg/m$^2$), reaching 88% for BMI values above 40 kg/m$^2$. In our study, the mean value of LS in healthy subjects was lower, 4.8±1.3 kPa, with a similar failure rate of 5.3%, also influenced by BMI, reaching 38.5% in obese patients. In a study performed by Corpechot et al [22], a similar mean value of LS (4.8 kPa) was obtained in a group of 71 healthy subjects.

It has been demonstrated that age is an important factor associated with fibrosis progression in chronic hepatitis [10-12]. So, we asked ourselves if in patients without known history of liver disease, fibrosis does not increase with age. We calculated the mean values of LS, as a marker of fibrosis, in various age groups. We found that the mean values of LS vary from 4.5±1.2 kPa in the 30-39 age group to 5±1.3 kPa in the 18-29 and 60-69 age groups, but with no statistical significance (p=0.5263). Also, in the study performed by Corpechot et al [22], age did not influence the mean value of LS in healthy individuals.

Regarding the influence of BMI, in the study performed by Roulot et al [21] the mean value of LS was significantly higher in obese subjects (BMI>30 kg/m$^2$): 6.26±1.89 vs. 5.37±1.51 kPa, p=0.0003. In our study, even if the mean LS value was higher in obese patients (5.3±1.6 kPa, vs. 4.8±1.2 kPa), the difference was not statistically significant. We must mention that in our group of patients only 12.5% (19/152) were obese; none of them with a BMI greater than 37 kg/m$^2$ and VMs were obtained only in 12 subjects. Maybe on a larger group of obese subjects the difference would become significant. In the study performed by Corpechot et al [19], BMI also did not influence the mean value of LS in healthy individuals.

Regarding the influence of gender, Roulot et al [21] found a mean value of LS higher in men than in women: 5.81±1.54 vs. 5.23±1.59 kPa (p=0.0002). In a smaller study, on 71 healthy subjects [19], the LS was also higher in men than in women (5.2±0.7 vs. 4.5±1.0, p<0.01). We also found that the mean LS in men was 5.1±1.2 kPa, significantly higher than in women 4.6±1.2 kPa (p=0.0082).

Another aspect is the large range of LS values obtained in healthy individuals: in the Roulot study (21) the proposed LS values for normal liver are 3.3–7.8 kPa in women and 3.8–8.0 kPa in men; in the Corpechot study [22], the LS values ranged from 2.5-6.9 kPa; and in our study, from 2.3 to 8.8 kPa.

This is an important issue, since in most of the studies the cut-off values for significant fibrosis in chronic C hepatitis (F≥2 Metavir) range from 7.1 kPa [6] to 8.8 kPa [17]. In a previous study on 199 patients with chronic C hepatitis, the cut-off value of LS for significant fibrosis (F≥2 Metavir) was 6.8 kPa, with PPV of 98%, NPV of 30.1%, a sensitivity of 59.6% and a specificity of 93.3% [23].

Various cut-off values for significant fibrosis (F≥2) were established in other chronic hepatopathies: 7 kPa in chronic B hepatitis [24]; 7.3 kPa in cholestatic hepatopathies [19]; 6.6 kPa [20] and 8.7 kPa in NASH [25]; 4.5 kPa in coinfection HCV+HIV patients [26].

In our group of 144 normal subjects in whom LS measurements were obtained, 11 (7.6%) had LS values ≥
6.8 kPa. We recommended for these patients to undergo further investigations, such as biological tests, viral markers, abdominal ultrasound, maybe even LB, in order to diagnose and stage an eventual underlying, unknown, chronic liver disease.

Also, an upper limit of the normal values of LS must be defined, but what should it be? Based on the published studies regarding LS values in healthy individuals [21, 22], on our own study, as well as on those regarding LS values significant for at least moderate fibrosis [6, 17, 19, 20, 24-26], we believe that an upper limit of 6-6.5 kPa for “normal LS” would be appropriate.

Conclusions

In our study, the mean values of liver stiffness measured by transient elastography in normal patients were lower than 5 kPa, were not influenced by the age of the subjects, or by their BMI, but were significantly higher in men than in women.

Conflicts of interest

None of the authors have any conflicts of interest

References

None of the authors have any conflicts of interest