HEPATIC VEINS CIRCULATION IMPACTING CONDITIONS:
DOPPLEROGRAPHIC AND MORPHOLOGICAL CORRELATION IN CHRONIC VIRUS HEPATITIS C PATIENTS

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ABSTRACT

Key words: Hepatic veins, Dopplerography, cirrhosis, fibrosis, steatosis, HAI.

Objective. The aim of study is to assess the influence of liver fibrosis, steatosis, patient weight and activity of hepatitis inflammation to hepatic vein hemodynamic.

Patients and methods. 176 consecutive patients with chronic virus hepatitis C underwent both liver biopsy and ultrasound with duplex-Dopplerography of right hepatic vein. Hepatic venous Doppler curve and patients body mass index was evaluated and compared with histological findings: Knodell liver fibrosis score, hepatitis activity index and level of steatosis.

Results. The incidence of flattened waveforms (biphasic and monophasic) in patients with mild, moderate liver fibrosis and without them was similar, 25% to 28.6%. In patients with severe fibrosis /cirrhosis it was 83.3%. The pathological flow pattern in patients with no steatosis, with mild, moderate and severe steatosis was found in 16 cases of 67 (23.9%), in 17 cases of 69 (24.6%), in 11 cases of 30 (35.5%) and in 4 cases of 9 (44%) respectively. No correlation was found between flattened Doppler curve and hepatitis activity or body mass index.

Conclusions. Severe liver fibrosis or severe steatosis has strong influence to hemodynamic of hepatic veins, what appears as flattened, biphasic or monophasic Doppler curve. Doppler ultrasound is not useful for diagnosis or differentiation of mild or moderate liver fibrosis. The incidence of mono-and biphasic hepatic venous flow has tendency to increase with increase of liver steatosis level, although this increase is not statistically significant. Activity of hepatitis inflammation and body mass of patient has no influence to blood flow pattern in hepatic veins.

INTRODUCTION

The main diagnostic tool used for assessment of liver damage in the cases of various diffuse liver diseases is punction biopsy. This method has several limitations including morbidity and mortality (“large complications” 0.4–2.8%, lethality 0–0.2% [1]), diagnostic accuracy, sampling error [2–4], interobserver and intraobserver variability [5–7] and difficulties in follow up. For this reasons, non-invasive diagnostic methods, including various ultrasonography (US) techniques, have been proposed and tested as means for detection and evaluation liver damages.

US signs of liver cirrhosis are well known: increasing and later decreasing of liver size, hypertrophy of lobus caudatus, changes in ratio between size of right and caudate lobes, nodularity and coarse echo structure of liver [8]. B-mode US did not allow detect liver fibrosis in pre cirrhotic stages, but it is directly related to severity of process itself, regardless of the underlying aetiology. Liver blood flow undergoes complex changes with the progression of chronic liver disease, what should be detected with Dopplerographic examinations. Characteristic features of liver cirrhosis are decreasing of portal blood flow velocity, disappearing of pulsatility, changing of flow detection in portal vein from hepatopetal to hepatofugal in more severe cases [9–13], increasing of resistive index in hepatic artery [14–16]. The most characteristic Dopplerographic sign has been suggested the changes in hepatic vein Doppler curve waveform from triphasic to biphasic and monophasic. [11, 17, 18]. Blood flow in hepatic veins in healthy subjects is phasic, reflecting the variations in central venous pressure due to cardiac cycle. The Doppler spectral waveform obtained from normal hepatic veins, therefore is multiphasic: two large ante grade diastolic and systolic waves and small retrograde wave corresponding to
atrial systole [18]. Liver parenchyma disease can impair the compliance of the wall of the hepatic veins, decreasing and flattening phasic oscillations. The reasons of decreased compliance can be liver fibrosis, steatosis, and increased intra abdominal pressure due different causes like adiposity or gravidity [19]. Heart diseases, regurgitation of tricuspidal valves or constrictive pericarditis also has influence to hepatic vein Doppler curve [20, 21]. Ability of Dopplerography to detect the liver fibrosis in pre cirrhotic stages of disease or differentiate stages of severity of liver fibrosis is still under discussions and needs furthermore assessment [10, 12, 14].

Aim of our study was to evaluate hepatic vein Doppler curve waveform in patients with chronic hepatitis C with different stages of liver fibrosis, and assessed influence of steatosis, patient’s weight and activity of liver inflammation to hepatic vein blood flow.

PATIENTS AND METHODS

The study included a total of 216 patients – 117 males (54.2%) and 99 females (48.5%). The study group included 176 viral hepatitis C patients from Infectology Centre of Latvia who underwent liver ultrasound examination and liver puncture biopsy. The number of males and females was 97 (55.1%) and 79 (44.9%) accordingly. The diagnosis of chronic hepatitis C was confirmed by the elevated ALT levels and positive anti HCV and HCV-RNA in serum.

The control group consisted of 40 patients from Pauls Stradins Clinical University Hospital who, for various reasons, underwent abdominal US imaging. The number of males and females was equal – 20. The average age of patients was 54.3±17.8 years. The study included patients who had no history or laboratory data on liver or heart disease and who had a normal ECG and normal levels AST, ALT and bilirubin. US imaging did not show any evidence on liver disease. Patients with chronic pancreatitis were not included in the study.

All patients included in the study had their body weight and height registered and body mass index (BMI) was calculated according to the equation BMI = body weight (kg)/height² (m²). BMI less than 20 was rated as low, 20–25 as normal, 26–30 as elevated, >30 as obese.

US examinations were performed on Philips ultrasound systems HDI 15, iU22 (Philips Health Care, Bothell, Wa) or Toshiba Aplio XG(SSA 790A Toshiba Medical Systems Corporation, Japan) using a convex multi frequency array probe (2.5–5 MHz).

In all cases the procedure started with a routine abdominal organ ultrasound examination according to the generally accepted standard protocol. Hepatic vein Doppler ultrasound imaging was performed in patient lying on the left side with raised right hand over the head. To minimize the impact of cardiac motion artefacts the right main hepatic vein was chosen for the examinations. In order to standardize examination conditions the waveforms were recorded during a light expiration after non-forced inspiration.

Doppler waveforms were classified into three groups: a normal waveform was presumed to be a triphasic curve, biphasic or monophasic were classified as pathologic. Liver puncture biopsy was made during the same examination under local anaesthesia using for the anaesthesia Sol. Marcati 0.5% 5–10 ml. Tissue samples were taken from various depths of the right liver lobe guiding the needle through the inter costal space along lin. axillaris ant.or media. For each patient two visually qualitative specimens were yielded and fixed in formalin solution. The histological assessment of the specimens was made by one experienced pathologist, who knew the clinical diagnosis of the patient, but was neither familiar with the data from US imaging nor knew whether the patient was enrolled in the study. Tissue samples were processed according to a standardized liver biopsy histochemical visualization protocol which includes colouring with haematoxylin and eosin, with Masson’s trichrome method, Perls method and PAS method. Morphological changes were assessed using hepatitis activity detection score recommended by Knodell [22]. For comparison in the study the common Knodell hepatitis activity index (HAI) and degree of hepatic fibrosis were used in following gradation: 0 – no fibrosis, 1 – mild fibrosis (spread of portal fibrosis), 3 – moderate fibrosis (porto-portal or porto-central bridging fibrosis), 4 – cirrhosis. Severeness of liver steatosis richness was expressed on a scale of four grades: no steatosis, mild steatosis (affects less than 25% of hepatocytes), moderate steatosis (26–50% of hepatocytes) and severe steatosis (more than 50% of hepatocytes). Liver puncture biopsy was well tolerated. There were no major complications that require intensive therapy, blood transfusion or surgical intervention. In some cases patients experienced soreness at the puncture site, irradiation of the pain towards the shoulder, soreness during inspiration, which resolved on their own or after a single administration of analgesic medication.

Abdominal US imaging for the patients from the control group was performed according to the conventional standard protocol requiring fasting at a minimum of 8 hours. Registration and evaluation of Doppler waveform of liver veins were similar to the study group. In the control group, for obvious reasons, liver puncture biopsy was not done.

DATA STATISTICAL PROCESSING

The study data statistical analysis was performed using generally established statistical parameters in medical studies: 1) central trends (mean, median, etc.), 2) parameters
describing data distribution (standard deviation, standard error of the mean, etc.). To test the hypotheses there were used data appropriate tests - an independent - sample t test of two normally distributed data to compare the mean values, analysis of variance (ANOVA) to compare three or more independent samples of mean value of normally distributed data, Pearson chi-squared test for to establish cases of equality of distribution. If the data distribution defied normal probability distribution, nonparametric statistical methods were applied.

To assess closeness of the relationship between variables there were applied methods of correlation analysis. In all cases the criterion of the significance level, which rejected the null hypothesis was $p < 0.05$. In some cases there were assessed confidence interval limits of 95%, too.

RESULTS

The study population included 176 patients, of whom 97 were males (55.1%), and 79 (44.9%) – females. The average age was 38.6±12.5 years. The control group included 40 patients, 20 females and 20 males aged 20 to 78, the average of 53.3±17.18 years. Triphasic shape of VHDx Doppler waveform in study group was established in 128 cases (73%), biphasic – in 26 cases (15%), monophasic – in 22 cases (13%). In the control group of 40 patients triphasic waveform was recorded in 29 cases (72.5%), biphasic in 7 cases (17.5%), monophasic in 4 cases (10%).

In the study group liver fibrosis was not found, i.e., Knodell fibrosis score was 0 in 42 patients (23.9%). Of these, 19 were females (46%), 23 were males (54%). Mild fibrosis, i.e. Knodell score 1, was found in 96 patients (54.5%). Of these, 42 were females (44%), 54 were males (56%). Severe fibrosis, i.e. Knodell score 3, was found in 32 patients (18.2%). Of these, 15 were females (47%) and 17 were males (53%). Cirrhosis, i.e. Knodell score 4, was found in 6 patients (3.4%). The numbers of females and males were equal – 3.

Triphasic VHDx Doppler curve in study group was registered in 128 cases (73%), biphasic in 26 cases (15%), monophasic in 22 cases (13%). In 40 patients of control group triphasic waveform was registered in 29 cases (72.5%), biphasic in 7 cases (17.5%), monophasic in 4 cases (10%).

Frequency of abnormal VHDx Doppler waveform curves in study group patients without liver fibrosis, with mild to moderate fibrosis showed no significant differences (28.6%, 23.9% and 25%, respectively). A relevant difference among frequencies of pathological forms of Doppler waveforms was found in patients without fibrosis or with mild to moderate fibrosis, and in patients with severe fibrosis (83.3%), however, due to the small number of patients with severe fibrosis, this difference was not statistically significant. Frequency of abnormal VHDx Doppler waveform curves did not differ significantly in the control group (27.5%) and in study groups patients without fibrosis and with mild to moderate fibrosis (28.6%, 23.9% and 25%, respectively).

Frequency of abnormal curves VHDx curves in fibrosis groups is shown in Table No. 1. Figure No. 1

<table>
<thead>
<tr>
<th>Knodell fibrosis score</th>
<th>Number of patients</th>
<th>Patological VHDx waveform n</th>
<th>%</th>
<th>95% CI limits Lower</th>
<th>Upper</th>
</tr>
</thead>
<tbody>
<tr>
<td>F-0</td>
<td>42</td>
<td>12</td>
<td>28.6</td>
<td>17.2</td>
<td>43.6</td>
</tr>
<tr>
<td>F-1</td>
<td>96</td>
<td>23</td>
<td>23.9</td>
<td>16.5</td>
<td>33.4</td>
</tr>
<tr>
<td>F-3</td>
<td>32</td>
<td>8</td>
<td>25.0</td>
<td>13.3</td>
<td>42.1</td>
</tr>
<tr>
<td>F-4</td>
<td>6</td>
<td>5</td>
<td>83.3</td>
<td>43.6</td>
<td>97.0</td>
</tr>
</tbody>
</table>

Figure 1. Pathological VHDx frequency curves depending on the degree of liver fibrosis
shows increment of frequency of pathological waveforms associated with the degree of fibrosis.

When comparing patients without liver fibrosis with patients with severe fibrosis and cirrhosis, irrespectively of the degree of liver steatosis, sensitivity of hepatic vein Dopplerography is 83.3%, specificity 71.4%, the positive predictable value (PPV) is 29.4%, and the negative predictable value (NPV) is 96.8%.

Pathologic waveforms in patients with severe steatosis were found in almost half of cases (44%), in group of moderate steatosis in 35.5% of cases. In groups with mild steatosis and without them incidence of flattened waveform was similar – 24.6% and 23.9% (tab. No. 2).

To determine the impact of steatosis on blood flow in liver veins, regardless of the degree of fibrosis, the frequency of pathological form of waveforms was evaluated separately in groups of patients without fibrosis and its mild form, as well as in patients with moderate fibrosis. In the both groups incidence of pathologic waveforms has tendency to increase, although this rise is not statistically significant (Figure Nr. 2).

Sensitivity of Doppler ultrasound imaging of hepatic veins in detection of severe steatosis is 44.4%, specificity – 76.1%, PPV – 20%, and NPV – 91.1%.

All patients were assessed for hepatitis activity index (HAI) according to Knodell score. Its value ranged from 1 to 15. Compared to hepatitis activity index and frequency of VHDx waveform types, the correlation was not found, there is no statistically significant increase of abnormal Doppler waveform with the acceleration of activity of hepatitis. P value for Pearson $\chi^2$ test is 0.066.

Since the distribution of HAI does not conform to the normal distribution of probabilities to find HAI in VHDx waveform groups Kruskal-Wallis test of nonparametric statistics was used. It was established that median ranks (average spot arranged in row) in VHDx groups did not show statistically significant difference ($2=0.394$; $df=2$; $p=0.821$).

Body weight assessed according to body mass index in almost half of patients enrolled in the study was normal (107 cases – 49.5%) or high (64 cases – 29.6%). Reduced body weight was found in 4 patients (1.9%), markedly increased weight or obesity was found in 41 patients (18.9%). To assess the impact of weight on the blood flow type in liver veins, a comparison of VHDx waveforms and body weight was made. For patients with triphasic VHDx waveform, it was $25.77\pm4.767$, with biphasic and monophasic waveforms – $26.73\pm4.96$ and $27.00\pm5.005$, accordingly. Analyzing frequency correlations between patients’ body weight and types of VHDx waveforms it was
found that frequency of abnormal Doppler waveform does not correlate with body mass. In patients with normal and abnormal waveforms BMI showed no statistically significant difference (p=0.364).

DISCUSSION

Doppler spectral waveform of liver veins is recognized to be the most useful parameter of Doppler ultrasound imaging for assessment of liver cirrhosis and fibrosis [10–12, 23–26], which is the focus also in our study. Triphasic waveform shape was characteristic to patients without fibrosis, as well as for patients with mild to moderate form of fibrosis. Their frequency within these groups did not differ. In the fibrosis group of F0, F1 and F3 it was 71.4%, 76.9% and, 75%, accordingly.

Certain amount of abnormal waveforms was found in all groups of patients with fibrosis. In the F0 group it was in 28.6% of cases, in the group of mild fibrosis – 23.9% of cases and in the group with moderate fibrosis – in 25% of cases. A similar frequency of abnormal waveforms was also found in the control group – 27.5%. Thus, the study shows that the Doppler ultrasound imaging of liver veins does not allow revealing mild forms of liver fibrosis or differentiating between mild and moderate forms from each other. In our study Knodell fibrosis severity grading system was used in which mild fibrosis was concerned in case of portal expansion of the fibrosis, and as fibrosis of moderate severity bridging fibrosis (porto-portal or porto-central) were concerned. In our opinion, the magnitude of the changes is too small to significantly increase the overall hardness of the liver tissue and to significantly reduce their ability to resign to wall pulsations of liver veins caused by the heart.

Other studies on this issue are controversial. Similarly to our study, differentiation between degrees of fibrosis was unsuccessful also for Bernatik with colleagues [12]. Significantly more abnormal waveforms in case of moderate fibrosis were found by Schneider [10] – in 38.2% of cases, while in patients with cirrhosis they were found by authors only in 52.9% of cases. Increase of abnormal waveform with an increase of severity of hepatitis is shown in Lim study [14] in 29%, 55% and 60%, respectively. In this study, some degree of fibrosis is not isolated, but the hepatitis activity index is estimated as a whole. Most studies on the subject for morphological assessment of liver use other scoring systems: usually METAVIR or its modification, where the degree of fibrosis is divided more subtly than in the Knodell system used by us. This could lead to different types of abnormal waveforms in groups with moderately severe fibrosis. However, in all the analysed studies, including our study, Dopplerography of liver veins is not enough efficient in diagnosis and differentiation of early forms of fibrosis, as well as in the differentiation between mild fibrosis and healthy liver.

Within the group of severe fibrosis/cirrhosis abnormal waveform was found significantly more frequently – in 83.3%. These results suggest that revealing of mild fibrosis and differentiation between them according to liver vein Doppler ultrasound data is not possible while cirrhosis is characterized by such form. With regard to the detection of cirrhosis such results correlate which the data from other authors. Frequency of abnormal waveform from cirrhotic patients according to data from some other authors’ data ranges from 50% [17] to 85% [9]. In our study in patients with mild steatosis or no steatosis at all abnormal waveforms were found in the same frequency, at around 24% of cases. Thus, accumulation of fat in less than 25% of liver cells do not significantly affect tissue hardness and elasticity, they retain the ability to respond to pulsations of liver veins. Increase of steatosis severity indicates a tendency for increase of frequency of pathological forms, although the level of statistical significance is not achieved (p=0.08). In the group with moderate steatosis this rate is 35.5%, in the group of patients with severe steatosis - 44%.

Our results are in line with research data from other investigators described in literature. A very strong impact of steatosis on the flow in liver veins is found by Dieterich [11]. Patients with severe liver steatosis (>50% of hepatocytes were affected by fat), monophasic Doppler waveform was found in 44 cases out of 49 (90%), and only in 3 cases out of 57 (5%) – in patients with mild steatosis (fat affected <25% hepatocytes), or no steatosis at all. Types of the waveform did not correlate with histological form of steatosis: micronodular, macronodular or mixed.

Impact of steatosis on the flow of liver veins was evaluated also by Schneider [10]. The authors analyse VHC patients with various degrees of liver fibrosis and steatosis, but they were not analysed separately. Frequency of abnormal VHDx curves had a better correlation with the degree of steatosis than with liver fibrosis. In patients with mild steatosis abnormal shapes of waveforms were found in 25.5%, in patients with severe steatosis 90.2%. Sensitivity and specificity of Doppler ultrasound in evaluation of severe steatosis was 88.2% and 74.5%, accordingly. The authors concluded that the triphasic shape of the curve reliably excludes opportunity of presence of severe steatosis, while the monophasic or biphasic shape are high sensitive to reveal such expressed steatosis. The drawback of the method is the low PPV – 36.6%. In our study, in patients with severe steatosis the frequency of abnormal shape of waveforms is lower than in a number of listed trials. This could be explained by different grading system of steatosis severity, which considered a severe steatosis to be of >66% involvement of hepatocytes. In another study the threshold...
of involvement of hepatocytes in expressed steatosis is considered to be 33% [27].

Traditionally it is believed that the flow in hepatic veins is affected by intra-abdominal pressure. One of the most important determinants of this pressure is the body mass. However, its effects on blood flow in the liver vasculature are relatively little analysed. O’Donohue [23] in his study analyzed flow parameters in the liver artery and in the portal veins of healthy individuals. Body mass index did not correlate neither with RI in the hepatic artery, nor with the flow rate in the portal vein, nor with its diameter. Hepatic venous flow types in relation to body obesity were analyzed by Karabulut [28]. The authors compared VHDx curves for patients with normal weight (BMI <25) and obesity (BMI >30). Abnormal wave form frequency was 1.1% (one in 84) and 45.1% (46 out of 102), respectively. Authors explain this difference with liver steatosis which in the study is evaluated according to US parameters; however, they are not sufficiently accurate indicators. Similar results have been gained also in our study. Body mass index does not show significant correlation with the type of VHDx waveform (Spearman’s rank correlation coefficient 0.132, p=0.103). For patients with triphasic VHDx waveform the average BMI is 25.77±4.7678, with a biphasic waveform – 26.73±4.96, with monophasic – 27.0±5.005. Body mass does not show significant impact on the flow type in liver veins.

A separate, specific factor which might affect liver hemodynamic is inflammatory activity of hepatitis. It has been analyzed in several studies. In our study to clarify the role of inflammatory activity frequency of abnormal VHDx waveforms were compared to Knodell HAI. No correlation was found, Spearman’s rank correlation coefficient was 0.047, p=0.533. Detailed analysis of relationship between inflammatory activity and Doppler waveform was analyzed by Dieterich [11]. In total, the flow of hepatic veins did not correlate with hepatitis activity index according to Knodell score, but splitting it into components the authors found a correlation with the degree of perportal changes (with or without the occurrence of hepatocellular bridge-like necroses). Other inflammatory factors in relation to the flow of liver veins were not observed. The authors found no explanation to this phenomenon. No correlation between inflammatory activity and type of VHDx waveforms was suggested also by Haktanir [29].

CONCLUSIONS

A high degree hepatic fibrosis affects blood flow in the veins in the form of a flattened (biphasic or monophasic) Doppler waveform. Dopplerography of liver veins does not make possible to distinguish mild and moderate fibrosis and healthy liver, neither to differentiate among early forms of fibrosis. Moderate and severe liver steatosis increases frequency of abnormal levelled waveforms; for mild steatosis levelled Doppler waveform is not typical. Patients body weight and hepatitis inflammatory activity in patients with viral hepatitis C does not affect blood flow in liver veins and raises no alterations in VHDx Doppler waveform.

REFERENCES

18. Coll A, Coccioletto M, Riva C, Martinez E et al. Abnormalities of

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