Noninvasive assessment of liver fibrosis in patients with Fontan circulation using transient elastography and biochemical fibrosis markers

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Objective: Studies have shown that patients with failing Fontan circulation may develop liver cirrhosis with its sequelae. Therefore, early detection of fibrosis is essential to identify patients at risk. Transient elastography has been evaluated for noninvasive staging of liver fibrosis in a multitude of studies.

Methods: A total of 39 patients who underwent the Fontan procedure were included in the study. All patients underwent an abdominal ultrasound, transient elastography, and detailed laboratory testing. Conventional echocardiography assessment of inflow and outflow and ventricular function were performed.

Results: Significant signs of liver fibrosis were found in 36 of 39 children according to the elastography method and in 28 of 39 children according to the measured biochemical fibrosis markers. The Spearman correlation coefficient between the liver stiffness measured with transient elastography and the time interval since the Fontan operation was highly significant (0.514, $P = 0.001$).

Conclusion: The present study shows that patients who undergo the Fontan procedure are at increased risk of developing liver fibrosis and liver cirrhosis. The risk increases with the age of the patient and the time interval since the Fontan procedure. The non-invasive measurement of liver fibrosis using transient elastography and fibrosis marker scores can be a useful tool to identify patients at risk and for noninvasive surveillance.

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The Fontan operation was first reported in 1968 by Fontan and has undergone several surgical modifications to bypass the inferior venous blood return to the lung using intracardial and extracardial conduits to the common atrium. It has been shown to be an effective palliative operation for children with a morphologically and functionally univentricular heart. However, during long-term follow-up, altered univentricular diastolic function^{1,2} and absence of pulsation in the pulmonary circulation¹ may lead to increased pulmonary vascular resistance associated with hemodynamic changes, including central venous and liver congestion.³⁻⁶ Long-lasting liver congestion and altered venous flow may lead to structural and functional alteration of the liver. The precise pathophysiologic mechanism is not clear. Studies have shown a correlation of the systemic venous pressure and the duration of the Fontan circulation with liver fibrosis and cirrhosis.^{4,6} Thus, staging of liver fibrosis is important in patients after the Fontan operation to identify patients at risk.

At present, liver biopsy is still the gold standard for the assessment of liver fibrosis. However, it is an invasive method associated with discomfort for the patient, and in rare cases serious complications have been reported.⁷ In addition, the accuracy of liver biopsy is limited because of significant intra- and interobserver variability and sampling errors.⁸ Therefore, research has been focused on the evaluation of noninvasive methods for the assessment of liver fibrosis. The different approaches include

Abbreviation and Acronym

ALT = alanine aminotransferase

routine hematologic and biochemical tests, surrogate fibrosis markers in the blood and respective algorithms, and recently transient elastography (FibroScan, Echosens, Paris, France). A combination of different blood markers and the assessment of tissue elasticity based on transient elastography have shown the best results in accurate determination of the degree of liver fibrosis in adults with different liver diseases.⁹ Studies have shown that transient elastography and serum fibrosis markers can be used in children just as well as in adults.^{10,11} In the present study we assessed possible fibrotic changes in the liver in patients with Fontan circulation using the combination of transient elastography and biochemical serum fibrosis markers.

Materials and Methods**Patients**

Thirty-nine patients who underwent the Fontan procedure and attended the Department of Pediatric Cardiology at the Saarland University Hospital for follow-up in 2006 were consecutively enrolled in the study. The baseline characteristics of the patients are shown in Table 1.

Other causes of liver disease (chronic hepatitis B and C, hemochromatosis, Wilson's disease, deficiency in alpha-1 antitrypsin, and autoimmune liver disease) were excluded serologically in all patients.

All patients received a B-mode sonography of the liver and spleen including duplex Doppler sonography of the liver and transient elastography. Blood parameters were obtained on the same day as the performance of ultrasound and transient elastography (for further description see "Blood Markers" in this section). All patients underwent standard echocardiography examination, including Doppler assessment of inflow and outflow in the univentricular heart.

The present study was performed in accordance with the ethical guidelines of the Helsinki Declaration. Informed consent from the parents was obtained for the examinations and blood sampling.

Sonographic Examination

All patients were examined by B-mode and duplex Doppler sonography using a 3.5-MHz transducer (Hitachi EUB-8500, Tokyo, Japan) in a supine position. All patients had fasted overnight. Focal lesions and vascular malformations in the liver were excluded by careful sonographic scanning of the liver. The liver parenchyma was examined for echogenicity, homogeneity, liver surface nodularity, hypertrophy of segment I, and signs of portal hypertension. The spleen length was measured and documented as absolute length and as relative length in relation to the age-adjusted medium length of a healthy subject of the same age.¹²

For duplex scanning the sample gate was adjusted to 2 to 6 mm depending on the diameter of the examined blood vessel. Spectral analysis was recorded and repeated twice to confirm the reproduc-

ibility of the Doppler sonography spectrum. Doppler sonography of the right hepatic vein was performed via the intercostal approach in a 30-degree left lateral position of the patient with the right arm raised above the head. The diameter of the right hepatic vein was measured in this position 2 to 3 cm distal to the confluence of the hepatic veins. The Doppler sonography spectrum was recorded if possible in short breathing pauses of 5 seconds. The flow pattern was measured 2 to 3 cm distal to the confluence of the hepatic veins. The maximum and minimum (reversed flow) velocity (centimeters/second) were recorded in each patient.

The portal vein diameter and flow pattern were measured at the corresponding site next to the right hepatic vein. The Doppler sonographic spectrum of the portal vein was documented if possible during a 5-second breathing pause to avoid influences of respiration and intra-abdominal pressure. The maximum (Vmax) and medium (Vmed) velocities (centimeters/second) were recorded in each patient.

Transient Elastography

The FibroScan is a medical device based on transient elastography. It is equipped with a probe including an ultrasonic transducer mounted on the axis of a vibrator. A vibration transmitted from the vibrator toward the tissue induces an elastic shear wave that propagates through the tissue. These propagations are followed by pulse-echo ultrasound acquisitions, and their velocity is measured (which is directly related to tissue stiffness). The harder the tissue, the faster the shear wave propagates. The results are expressed in kilopascals.^{9,13-15} Transient elastography has been evaluated in a multitude of liver diseases, and different studies evaluating the reproducibility of transient elastography have reported high intraobserver (96%–98%) and interobserver (89%–98%) agreement.^{16,17}

Recently, this method was validated in children with liver fibrosis receiving liver biopsy with similarly good diagnostic accuracy as in adults.¹⁰

The patients were examined in a supine position with the right arm elevated above the head. Patients were instructed to continue breathing as usual. The examination was performed on the right lobe of the liver through the intercostal space in all patients. An area was chosen where the liver tissue was free of large blood vessels. After the area of measurement was located, the examiner pressed the button of the probe to start the acquisition. The measurement depth was between 25 and 65 mm. Ten successful acquisitions were performed on each patient. The success rate was automatically calculated by the machine as the ratio of the number of successful acquisitions over the total number of acquisitions. Only transient elastography results obtained with 10 valid measurements and with a success rate of at least 60% were considered. These are quality criteria evaluated by previous studies. The median of all successful measurement was automatically calculated by the machine as representative of the liver elasticity module. The entire examination lasted approximately 5 minutes per patient.

Because the probe of the FibroScan was developed for adults and the liver stiffness measurement was standardized between 25 and 65 mm below the skin surface, overestimation of the transient elastography values in young children was expected. Therefore, the raw data of each measurement were analyzed and the liver stiffness was recalculated between 25 and 55 mm below the skin surface in children less than 7 years of age. This was performed in accordance

TABLE 1. Patients' baseline characteristics

Characteristics	Mean $\pm$ SD	Median	Range
Demographics (n = 39 patients)			
Male	20 patients		
Female	19 patients		
Age (y)	11.6 $\pm$ 5.5	11.0	3–23
Time interval since Fontan operation (mo)	67.9 $\pm$ 41.3	69.0	2–180
BMI (kg/m ²)	17.4 $\pm$ 3.2	16.5	13–26
Echocardiographic parameters			
Functionally univentricular right heart	13 patients		
Functionally univentricular left heart	22 patients		
Intracardiac tunnel (extracardiac tunnel)	37 (2) patients		
Fenestration open	35 patients		
Biochemistry			
ALT (U/L)	28.9 $\pm$ 12.7	27.0	9–69
ALT ($\times$ ULN)	0.90 $\pm$ 0.46	0.88	0.3–2.16
Aspartate aminotransferase (U/L)	46.9 $\pm$ 14.0	44.0	21–76
Aspartate aminotransferase ($\times$ ULN)	1.09 $\pm$ 0.45	1.10	0.3–1.8
Gamma-glutamyl transpeptidase (U/L)	57.8 $\pm$ 34.3	47.5	16–144
Gamma-glutamyl transpeptidase ($\times$ ULN)	1.97 $\pm$ 1.55	1.64	0.2–7.11
Bilirubin total (mg/dL)	0.98 $\pm$ 0.64	0.90	0.1–2.5
Lactate dehydrogenase ($\times$ ULN)	1.06 $\pm$ 0.43	1.06	0.3–2.0
Alkaline phosphatase ($\times$ ULN)	0.74 $\pm$ 0.38	0.75	0.3–2.2
Albumin (g/L)	46.1 $\pm$ 4.5	47.0	34–52
Hemoglobin (g/dL)	15.4 $\pm$ 1.6	15.7	12.4–19.6
Leucocyte count (10 ³ /mm ³)	7.7 $\pm$ 2.9	6.65	3.3–14.7
Platelet count (10 ³ /mm ³)	260.3 $\pm$ 100.0	248.5	120–514
C-reactive protein (mg/L)	4.5 $\pm$ 6.3	1.5	0.6–30.8
INR	1.97 $\pm$ 0.6	1.91	1.11–4.6
PTT (sec)	36.0 $\pm$ 4.9	36.0	28–49
Factor V (%)	69.6 $\pm$ 14.8	72.0	45–98
Antithrombin III (%)	103.8 $\pm$ 10.6	103.0	84–124
Immunoglobulin G (mg/dL)	1024 $\pm$ 217	1035	543–1580
Immunoglobulin M (mg/dL)	94.1 $\pm$ 33.6	89.5	41–168
Glucose (mmol/L)	3.5 $\pm$ 1.4	3.8	1.0–5.72
Triglycerides (mmol/L)	0.74 $\pm$ 0.47	0.62	0.2–2.75
Cholesterol (mmol/L)	3.25 $\pm$ 0.99	3.33	2.0–6.08

SD, Standard deviation; BMI, body mass index; ULN, upper limit of normal; ALT, alanine aminotransferase; INR, international normalized ratio; PTT, prothrombin time.

with a previous study of children who received a liver biopsy and a transient elastography.¹⁰

The staging of liver fibrosis was performed according to the study of Foucher and colleagues.¹⁵ In this study, transient elastography values were analyzed in comparison with histology using the semiquantitative histologic staging system of METAVIR:¹⁸ F0 = no fibrosis, F1 = portal fibrosis without septa, F2 = portal fibrosis with few septa, F3 = numerous septa without cirrhosis, F4 = cirrhosis. The most discriminant cutoff values for transient elastography in the study of Foucher and colleagues were 7.2 kPa for F $\geq$ 2, 12.5 kPa for F $\geq$ 3, and 17.6 for F = 4.⁹ These cutoff values were used for staging of liver fibrosis in the present study.

Echocardiography

All patients were examined using a 3.5 or 2.5-MHz transducer interfaced with a Vingmed ultrasound system (GE-Vivid 7, Horten, Nor-

way). Standard transthoracic imaging from the parasternal, apical, and subcostal views was performed in the left lateral decubitus position. Initial routine diagnostic imaging included color flow mapping and pulsed-wave Doppler of the inflow of the lung veins in the common atrium, inflow at the atrioventricular valve, and outflow at the aortic valve for possible regurgitation. In an apical “4-chamber” view, the atrioventricular valve inflow velocity profiles were recorded with the Doppler sample placed at the tip of the atrioventricular valves with simultaneous electrocardiogram recordings in each patient. In case of double-inlet morphology the inflow from the left atrium was considered for the measurement of inflow patterns. Atrioventricular inflow was analyzed in 5 heart cycles for the maximal inflow velocity (E wave), late atrial wave (A wave), and E/A ratio. The usual size of the pulsed Doppler gate was 1.5 mm, and the filter was set at 100 Hz. For optimal acquisition, care was taken to direct the transducer beam as closely as possible to

the Doppler beam, at less than 20 degrees in selected planes. All measurements were recorded digitally in the EchoPac program for subsequent off-line data analysis. The mean value of each parameter was calculated in 5 consecutive cycles.

Blood Markers

The following blood parameters were determined after overnight fasting in the same laboratory: aspartate aminotransferase, alanine aminotransferase (ALT), γ -glutamyl transpeptidase, total bilirubin, lactate dehydrogenase, alkaline phosphatase, albumin, hemoglobin, platelet count, leucocyte count, C-reactive protein, international normalized ratio, prothrombin time, factor V, antithrombin III, immunoglobulin G and M, cholesterol, triglycerides, fasting glucose, α 2-macroglobulin, apolipoprotein A1, and haptoglobin.

Enzymatic activity was measured at 37°C according to International Federation of Clinical Chemistry standards.

The laboratory followed the preanalytic and analytic recommendations required to obtain the fibrosis marker score results.¹⁹ The FibroTest and ActiTest (Biopredictive, Paris, France), the serum necroinflammatory marker score, were computed on the Biopredictive website (www.biopredictive.com). The security algorithms on the industrial website permitting the exclusion of patients with a high-risk profile of false-positive and negative results were respected. This formula has been evaluated in various liver diseases for the diagnosis of liver fibrosis and is currently the best evaluated noninvasive serum fibrosis marker algorithm and is commercially available throughout the world.^{19,20} In studies comparing these noninvasive serum markers with the reference method liver biopsy, the FibroTest showed good diagnostic accuracy for the assessment of liver fibrosis, whereas the ActiTest was found to be a useful tool for the assessment of necroinflammatory activity in the liver.¹⁹⁻²¹

Statistical Analysis

Statistical analysis was performed using the Statistical Package for the Social Sciences (version 12.0, SPSS Inc, Chicago, Ill). Clinical and laboratory characteristics of patients were expressed as mean $\pm$ standard deviation. All tests were 2-sided. Relationships between variables were examined using the Spearman's rank correlation coefficient. The relation of the time interval since the Fontan operation and the fibrosis staging using the transient elastography and the necroinflammatory grading using the ActiTest are illustrated as the median and 25th to 75th percentile values (box-plot). The relation of the time interval since the Fontan operation and the fibrosis staging using the fibrosis marker score are demonstrated in a range diagram.

TABLE 2. Liver fibrosis stages with FibroScan and FibroTest

Method	F 0/1	F 1-2	F 2	F 3	F 3-4	F 4	Mean $\pm$ SD	Median
FibroScan (Echosens, Paris, France)	13%	—	26%	20%	—	41%	2.9 $\pm$ 1.0	3.0
FibroTest (Biopredictive, Paris, France)	32%	35%	6%	21%	3%	3%	1.7 $\pm$ 1.0	1.5

SD, Standard deviation. *P* value < .001 for all values.

Results

Altogether, 39 patients who underwent the Fontan procedure and attended the Department of Pediatric Cardiology at the Saarland University Hospital for follow-up in 2006 were consecutively enrolled in the study. Thirteen patients had a functionally univentricular right heart, 22 patients had a functionally univentricular left heart, and 4 patients had a functionally univentricular right heart. The baseline characteristics of the patients are shown in Table 1.

Successful measurement of liver stiffness using transient elastography was performed in all examined children. Significant signs of liver fibrosis were found in 36 of 39 children according to the elastography method and in 28 of 39 children according to the measured biochemical fibrosis markers in the studied patients after the Fontan operation. The number of patients assigned to each fibrosis stage according to Meta-vir using transient elastography and the fibrosis marker score is shown in Table 2.

Liver stiffness measured with transient elastography correlated significantly with the biochemical fibrosis markers (0.344, *P* < .05) and the necroinflammatory serum marker (0.480, *P* < .05).

Factors Influencing Liver Stiffness, Fibrosis, and Necroinflammatory Activity in Patients After the Fontan Operation

Liver stiffness measured with transient elastography correlated significantly with the patient's age (0.439, *P* < .05) and the time interval since the Fontan operation (0.544, *P* = .002) (Figure 1). In addition, significant positive correlation was found between the liver stiffness measured with transient elastography and the laboratory values ALT (0.411, *P* < .05) and factor V (-0.429, *P* < .05).

A relationship between the liver fibrosis measured with transient elastography and echocardiographic parameters was found for the inspiration/expiration diameter ratio of the vena cava (0.456, *P* = .05) and for the E/A value across the atrioventricular systemic valve (0.372, *P* < .05). No correlation was found between the liver stiffness and the ejection fraction of the univentricular heart or other echocardiographic markers of global systolic and diastolic function, the gradient at the Fontan fenestration, the flow velocities in the liver veins, and the estimated hepatic venous blood flow.

The serum fibrosis marker correlated significantly with the age of the patient (0.453, *P* < .01) at the time of examination, the age of the patient at the time of the Fontan procedure (0.374, *P* < .05), the serum necroinflammatory marker (0.547, *P* = .001), and the transient elastography (0.344, *P* < .05). No significant correlation could be found between the serum fibrosis marker and the time interval since the Fontan operation (0.192, *P* = .276) (Figure 2).

The necroinflammatory marker correlated significantly with the time interval since the Fontan procedure (0.374, *P* < .05) (Figure 3) and the age of the patient at the time

Figure 1. Box-plots of the time interval since the Fontan operation for each fibrosis stage according to Metavir using transient elastography. The top and bottom of the boxes represent the first and third quartiles, respectively. The length of the box represents the interquartile range within which 50% of values are located. The thick line through the middle of each box represents the median. The error bars mark the minimum and maximum values (range). *OP*, Operation.

Figure 2. Range diagram of the time interval since the Fontan operation for each fibrosis stage according to Metavir using the serum fibrosis marker. No significant correlation could be found between the biochemical fibrosis staging using the serum fibrosis marker and the time interval since the Fontan operation. However, a trend can be seen in the present figure showing increasing FibroTest (Biopredictive, Paris, France) values with increasing time interval since the Fontan operation. *OP*, Operation.

of the present examination (0.359, $P < .05$). In addition, the serum marker of necroinflammatory activity in the liver correlated significantly with transient elastography values (0.480, $P < .05$), the retrograde flow (positive flow) in the right hepatic vein (0.347, $P < .05$), and the E/A values (0.407, $P < .05$).

Time Interval Since the Fontan Operation

The time interval since the Fontan operation seems to play an important role in disease progression. In addition to the significant correlation between the time interval since the Fontan operation and transient elastography (0.544, $P < .005$), as well as the serum necroinflammatory marker (0.374, $P < .05$), the time interval since the Fontan operation correlated significantly with the inspiration/expiration diameter ratio of the vena cava (0.477, $P < .05$) (as a sign of congestion of the liver) and the echocardiographic parameters indicating altered diastolic function, such as the E/A value across the atrioventricular systemic valve (0.431, $P < .01$).

Conventional Sonographic Examination of the Liver

Adequate visualization of the hepatic and portal veins by B-mode sonography and sufficient analysis of respective flow patterns were achieved in all examined patients. How-

ever, the very young patients ($n = 7$) were not able to hold their breath. No correlation was found between the size of the spleen and the fibrosis stage or blood parameters. All patients had a triphasic flow pattern in the right hepatic vein, and no focal lesions were detected in the liver in any patient.

No significant correlation could be found between the liver fibrosis stages measured with the transient elastography or serum fibrosis marker and the blood velocity (maximum and minimum) in the portal and right hepatic veins.

However, significant correlation was found between the magnitude retrograde flow in the right hepatic vein and the serum necroinflammatory marker. The laboratory values aspartate aminotransferase (0.347, $P < .05$), ALT (0.495, $P < .005$), and $\alpha 2$ -macroglobulin (0.391, $P < .05$) were also significant.

Discussion

Several studies have shown that the Fontan circulation may lead to venous liver congestion and possible structural and functional liver alteration. This can finally lead to liver cirrhosis with its sequelae.^{4,6} Hepatocellular carcinoma in patients with liver cirrhosis after the Fontan procedure was recently reported.^{4,6}

Figure 3. Box-plots of the time interval since the Fontan operation according to the grading of necroinflammatory activity using the ActiTest (Biopredictive). Twenty-three patients had ActiTest A0 (no necroinflammatory activity), 7 patients had A0-1, 5 patients had A1, 4 patients had A1-2, and no patient had A2, A2-3, A3, A3-4 or A4 (severe necroinflammatory activity). A significant correlation was found between the ActiTest and the time interval since the Fontan procedure. Increasing inflammatory activity is shown with increasing time interval since the Fontan operation. The decrease at the highest measured grade of activity might be due to the small number of patients in this group. OP, Operation.

The liver has not been routinely examined in patients after the Fontan operation, because it did not seem to play a crucial role in the morbidity and mortality in the mid- and long-term follow-ups. In the meantime, however, life expectancy in these patients has increased, and morbidity and mortality caused by failing of the Fontan circulation have decreased. The standard procedure for the staging of liver fibrosis is the liver biopsy. However, it is ethically not justified to perform a liver biopsy in all pediatric patients with Fontan circulation.

Therefore, the aim of this study was to draw attention to this important complication in the liver of these patients with Fontan circulation using the novel noninvasive method of transient elastography in combination with biochemical serum fibrosis markers. By using the combination of 2 different methods, it was possible to detect structural and functional alterations that are of prognostic value in the staging of liver fibrosis and in the identification of those patients at risk of developing liver cirrhosis. The inverse significant correlation of transient elastography values with factor V, which

is known to decrease with the progression of liver disease, substantiates the use of transient elastography as a marker of liver disease progression. Only 1 patient had clinic and sonographic signs of liver cirrhosis (low platelet count, low albumin, elevated bilirubin, liver surface nodularity, hypertrophy of segment I, and signs of portal hypertension).

In the present study, a significant correlation between the fibrosis staging using transient elastography and the time interval since the Fontan operation was found. The number of patients with significant liver fibrosis after the Fontan procedure increased dramatically especially at 5 years (Figure 1).

The morphologic and functional changes in the liver with increasing time after the operation and increasing age are simultaneous to alteration in systolic and diastolic function of the univentricular heart.²² However, the precise pathophysiologic mechanism leading to liver injury in association with Fontan circulation is not clearly elucidated. The findings of a significant correlation of the serum necroinflammatory marker, as an indirect marker of necroinflammatory activity (necrosis) with other studied parameters, is suggestive for hemodynamically induced liver congestion and altered blood flow to the pulmonary circulation. This may result in inflammatory activity in the liver, followed by increased fibrosis. This assumption is supported by the significant correlation between transient elastography values and ALT levels as a marker of inflammation. These correlations may explain the abnormal sonographic findings of liver morphology in our study, which may be in part induced by long-lasting venous liver congestion. The findings are in accordance with previous studies reporting a correlation of the systemic venous pressure and the duration of the Fontan circulation with liver fibrosis and cirrhosis.^{4,6} Abnormal liver histology findings have been reported in patients with failing Fontan circulation and adults with right-sided heart failure.^{23,24} The pathologic substrate seems to be similar. The congestive hemodynamic process usually starts in the form of sinusoidal dilatation, parenchymal atrophy, and progressive sinusoidal collagen deposition in the perivenular distribution. With long-lasting congestive effect, these morphologic changes advance leading to profound alterations in liver architecture and diffuse sinusoidal fibrosis and cirrhosis.^{23,24} The abnormal hemodynamic conditions of passive low flow associated with liver venous congestion in the patients with Fontan circulation may result in the same pathophysiologic mechanisms of liver fibrosis and cirrhosis. Alterations of pulmonary resistance may be equally important; however, because of the noninvasive character of the present study, simultaneous invasive hemodynamic measurements of pulmonary arterial pressure and resistance were not performed.

Limitations

At present, liver biopsy is still the reference standard for the assessment of liver fibrosis. Therefore, a limitation of the

present study is the lack of comparison of the noninvasive methods with simultaneous histologic assessment of liver fibrosis by liver biopsy. However, the accuracy of liver biopsy is limited because of intra- and interobserver variability and sampling errors.⁸ A study of more than 10,000 virtual biopsies by Bedossa and colleagues⁸ showed that liver fibrosis stage is correctly diagnosed in only 65% of cases if the biopsy is at least 15 mm long and in 75% of cases if the biopsy is at least 25 mm long, and the optimal size should be 40 mm. However, most biopsies do not fulfill these optimal criteria even at large hepatology centers.²⁰ The ultimate validation of liver fibrosis as a marker of liver injury is its prognostic value in terms of morbidity and mortality. In a recently published study, the serum fibrosis marker was shown to have a 5-year prognostic value similar to that of liver biopsy.²⁵ Therefore, noninvasive methods of liver fibrosis may not be inferior to liver biopsy for the assessment of liver fibrosis. Thus, in the present study it did not seem to be ethically justified to perform a liver biopsy in these young children.

After the Fontan operation, patients have systolic and diastolic myocardial dysfunction, and abnormal venous and arterial hemodynamics that lead to congestion in the liver.²⁶ The increased systemic venous pressure leads to congestion in the liver, which is per se associated with an increased stiffness of the liver. Previously, no studies have evaluated transient elastography for the staging of liver fibrosis in patients with congestive liver disease. An overestimation of the liver fibrosis stage is expected in these patients because of the increased blood volume in the liver. This explains the differences in the assignment of fibrosis stages between the transient elastography and the fibrosis marker score in the present study. The mean fibrosis stage was 2.9 ± 1.1 using transient elastography and 1.7 ± 1.0 using the serum fibrosis marker. The biochemical serum fibrosis marker is independent from the congestion in the liver. Because a significant correlation was found between the serum fibrosis marker and the transient elastography values, an overestimation of transient elastography of approximately 1 fibrosis stage is assumed.

Transient elastography might be especially useful for individual follow-up, in which an increase of transient elastography value can be interpreted as a sign for the progression of liver fibrosis. However, longitudinal studies are necessary to support this assumption. The results need to be interpreted taking into account the results of hemodynamic examinations and the quantitative assessment of systolic and diastolic univentricular function. The detectable liver changes in the present study measured with transient elastography and the fibrosis marker score do not seem to be directly related to altered univentricular function. Because of the noninvasive characteristic of this study, simultaneous invasive hemodynamic measurements of pulmonary arterial pressure and resistance were not performed. We speculate that the passive

slow flow in the Fontan circulation without the physiologic antegrade pump function may be responsible for the fibrotic changes in these patients.

Conclusions

This is at present the largest prospective cross-sectional epidemiologic study showing that patients who have undergone the Fontan procedure are at risk of developing liver fibrosis and cirrhosis with its sequelae. This study shows that this risk increases with the time interval since the Fontan operation and the age of the patient. Noninvasive methods, such as transient elastography and fibrosis marker scores, seem to be useful tools to assess liver fibrosis in these patients and may be helpful to recognize a progression of the liver disease during routine follow-up. Therefore, assessment of liver structure and function using transient elastography and fibrosis marker scores should be included in the long-term follow-up of patients with Fontan circulation. Such screening programs may have significant consequences when patients with failing Fontan circulation are candidates for orthotopic heart and/or liver transplantation.

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