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REVIEW OF PATIENTS WITH HEPATOSTEATOSIS AND CHRONIC HEPATITIS C

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We analysed a HCV RNA positive population with varied steatosis index admitted at Infectious Diseases and Hepatology Department, Medical University of Wroclaw in terms of existing abnormalities in biochemistry parameters, anthropometric differences as well as the antiviral therapy outcomes.

Słowa kluczowe: stłuszczenie wątroby, wirusowe zapalenie wątroby typu C, NASH
Key words: hepatosteatosis, chronic hepatitis C, anthropometric parameters, NASH

INTRODUCTION

The liver is a central lipid metabolism regulatory organ whose pathology may be induced by different factors leading either directly or indirectly to oxidative stress, organelle dysfunction, cell damage and steatohepatosis. "Two hits" hypothesis describes the overall non alcoholic steatohepatitis (NASH) pathology and steatosis evolution to inflammatory process (steatohepatitis). Modern models suggest underlying metabolic (overweight, glucose intolerance , lipid metabolism) and inflammatory agents as key factors for development of the lesions (1).

Hepatosteatosis is described in 30-70% of chronic hepatitis C patients which is a higher percentage than in chronic hepatitis B patients or other similar pathologies (1, 2). Some data suggest strong propensity of hepatitis C virus (HCV) itself to induce the changes since unless all major metabolic confounders are excluded still the steatosis prevalence ranges 30-40% ([3]). In this case, special particles of HCV (e.g. core protein) have been

proved to induce lipoperoxidation and finally cell damage and steatohepatosis as the final hit ([4]). Furthermore, it has been revealed that in chronic hepatitis C patients trigliceryde accumulation within hepatocytes is potentially attributed to inhibition of very low density lipoprotein particles (VLDL) secretion which is essential for HCV cell entry (5, 6). The fact is reflected by hypobetalipoproteinemia found in the sera of HCV infected patients ([7]) which may be regarded as a consequence of HCV cytopathic effects in cells ([8]). As the issue was specially interesting to us we reviewed it in detail in a former article which was a source data for the study ([9]).

In the analysis we retrospectively followed the history of chronically HCV infected patients with steatohepatosis within our center .We posed several questions concerning the population profile as histopathology lesions, clinical overview with special attention to liver enzyme activity and overall metabolic parameters and referred them to each other as well as some epidemiology source data.

MATERIALS AND METHODS

A random HCV-RNA positive population (HCV Ab-positive, HBcAb-negative) of Infectious Diseases and Hepatology Department, Medical University of Wroclaw was included into the analysis. All of the patients were hospitalized in 2003-2006 and underwent typical procedures essential for the therapy qualification (e.g. HCV genotype, viral load, autoantibodies, biochemical analysis, liver biopsy). A retrospective observation of the population was performed as given in the chronic hepatitis C database of our centre. Based on the steatosis index (according to Dixon`s scale) of the liver, assessed by one pathologist, we observed the population in terms of metabolic factors, liver enzyme activity and medical history underlying the HCV pathology.

The population characteristics. 119 Caucasian patients (76 men and 43 women) aged 44,4 yrs(19-68) Mē (median) 45,
Weight 74.3 kg (n=108, 40-145) Mē 74,
BMI 25.4 kg/m2 (n=108, 16.8-43.2) Mē 25.6,
serum cholesterol 164.9 mg% (n = 109, 61-287) Mē 160,
HDL 45.6 mg% (n=43, 26-90) Mē 42.9 ,
serum triglicerides 96.8 mg% (n=83, 19-271) Mē 84,
Alat 123.3 IU (n=117, 9-821) Mē 93,
GGTP 92.5 IU (n=116, 7-640) Mē 62,5,
bilirubine 1,18 mg% (n=54, 0.5-2.7) Mē 1,1,
HCV RNA viral load ranged 106 copies/ µl in all the population
Genotype profile (n =93): 1a/b=52; 3a=40; 4c/d=1
Liver pathology overview (METAVIR group):
Grading 1,76 (0-3.5);staging 2.01 (1-4) ,
Steatosis 1, 57 (0-3) (Dixon`s scale, Fig 1-3)

The population was stratified into 4 groups dependent on the steatosis grade: 1 (< 5%), 2 (5-25%), 3 (>25) - and 0 -no steatosis . No patients >60% steatosis were present in the study population.

The subpopulations were comparable for sex, age, cardiac and diabetic history.

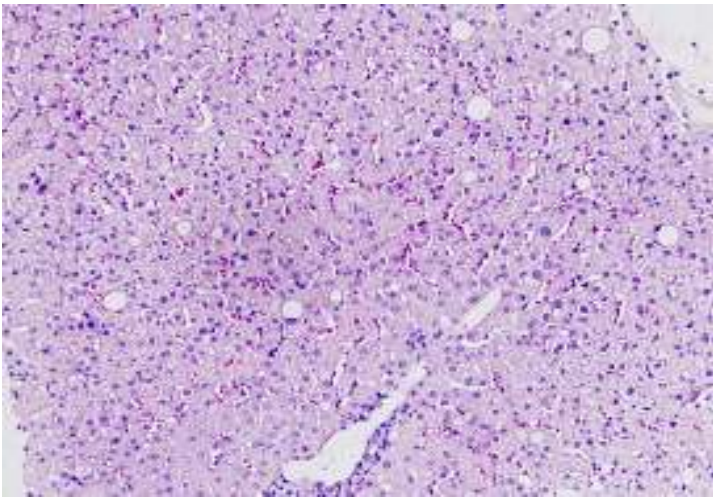
Because of low credibility data of alcohol consumption we could not differ the patients on the factor.

The overall observation in terms of chronic hepatitis therapy was 1 up to 4 years since the first liver biopsy. All patients to whom it was concerned were treated according to the Polish Panel Experts Guidelines with peg-interferon α 2 and ribavirine therapy. Effects of the therapy were described either by end-of treatment response (ETR) or sustained response (SVR). We had to use both of the parameters altogether in the analysis since not in all cases both of them were available. Three patients in case of negative ETR were not tracked with SVR as they did not come to the follow up visit.

Statistical analysis was performed using the STATISTICA PL 5.1 software. For variables with normal distribution, Student`s t-test was used when comparing the arithmetic averages of two independent samples. For variables with distribution other than normal non-parametric χ^2 test was used to verify hypothesis. The results with a significance level of $p \leq 0,05$ were considered statistically significant.

RESULTS

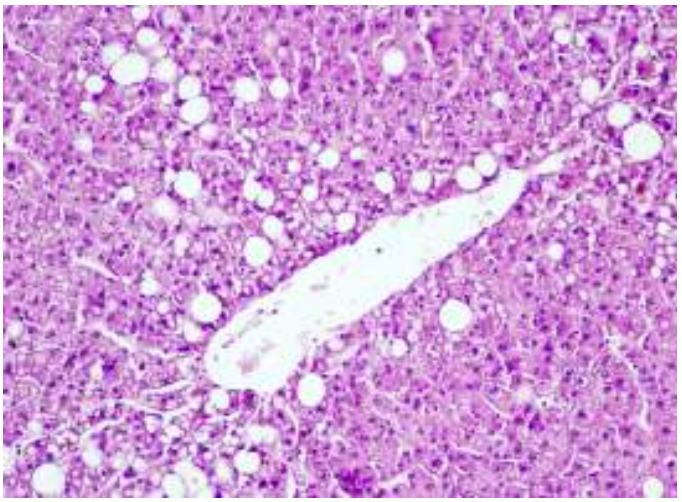
In our center HCV-chronically infected patients with hepatosteatosis grade 2 (5-25%) of steatosis liver lesions prevailed (Fig 1-3).



Ryc. 1. Stłuszczenie stopnia 1 (skala Dixona) barwienie H-E /preparat badany w Katedrze Patomorfologii AM we Wrocławiu/

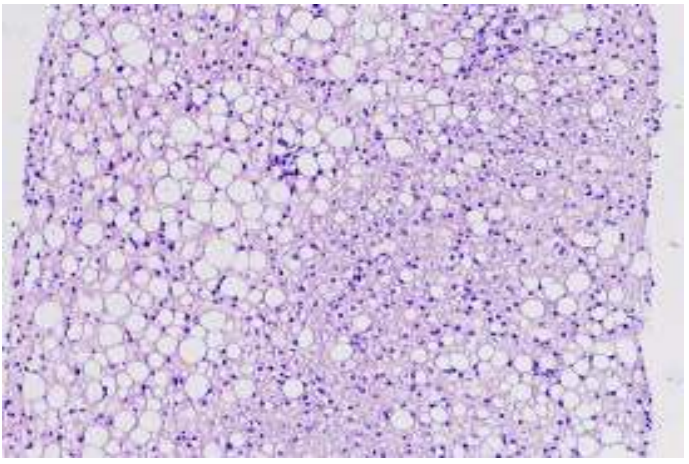
Fig 1. Steatosis 1 (Dixon's scale) H-E methods /sample analyzed at Patomorphology Department, Medical University of Wroclaw/

Patients with no steatosis were significantly younger than patients of other groups (40.7 vs 43.8-45.8 yrs) (Fig 5). There were hardly any age differences between patient groups with steatosis.



Ryc.. 2. Stłuszczenie stopnia 2 (skala Dixona) barwienie H-E /preparat badany w Katedrze Patomorfologii AM we Wrocławiu/

Fig. 2. Steatosis 2 (Dixon's scale) H-E methods /sample analyzed at Patomorphology Department, Medical University of Wroclaw/

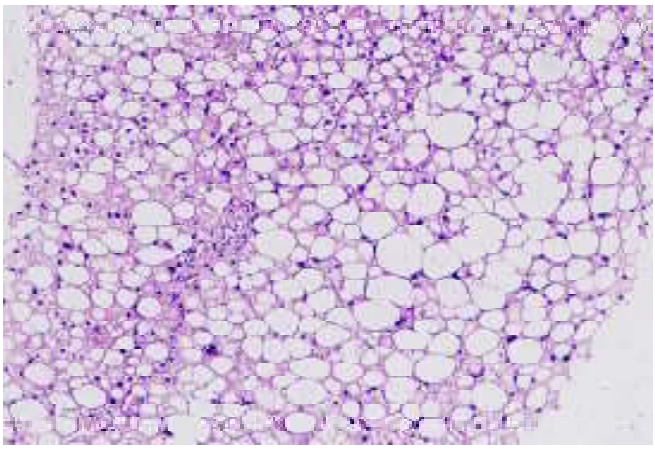


Ryc. 3. Stłuszczenie stopnia 3 (skala Dixona) barwienie H-E /preparat badany w Katedrze Patomorfologii AM we Wrocławiu/

Fig. 3. Steatosis 3 (Dixon's scale) H-E methods /sample analyzed at Patomorphology Department, Medical University of Wroclaw/

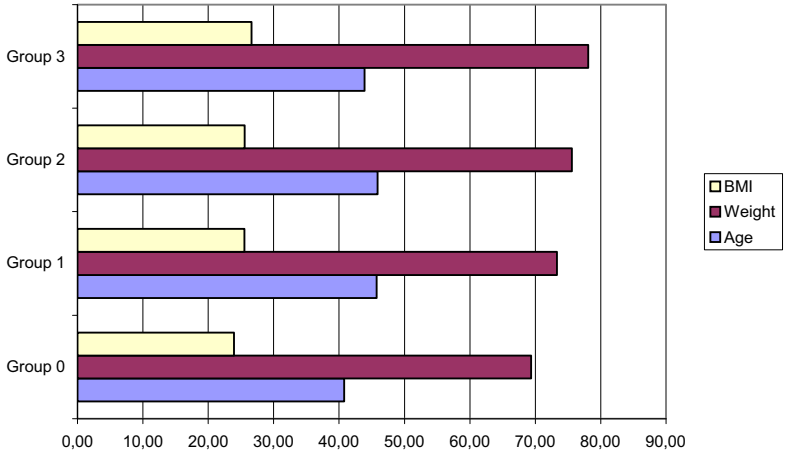
Fibrosis stage was significantly high in group 3 compared to the control group (2.25 and 1.26 respectively, p=0.0001). However, similar staging score was in group 1, 2 and 3 (2.24, 1.12 and 2.25 respectively, NS).

Relatively low grading score was in the control group unlike groups with steatosis (e.g. 1, 18 and 1, 87 respectively for group 0 and group 2), table I.



Ryc. 4. Stłuszczenie stopnia 4 (skala Dixona) barwienie H-E /nieobecne w analizie, preparat badany w Katedrze Patomorfologii AM we Wrocławiu/

Fig.4 . Steatosis 4 (Dixon's scale) H-E methods /not present in the analysis, sample analyzed at Patomorphology Department, Medical University of Wroclaw/.



Ryc.5. Porównanie podgrup pod względem wieku, wagi i wskaźnika BMI.

Fig .5. Differentiation of BMI, weight and age in each group

Tabela I. Średnia arytmetyczna wyników badanych parametrów biochemicznych u pacjentów z pwzw C w każdej z badanych grup

Table I. Mean arithmetic values of biochemical parameters in each analyzed group

	Number	ALAT (IU)	GGTP (IU)	Bilirubin (mg%)	Cholesterol (mg %)	HDL (mg %)	Triglyceride (mg %)
Group 0	23	117.0	69.9	1.16	161.2	47.6	79.1
Group 1	27	122.8	108.8	1.56	167.8	46.4	93.3
Group 2	43	127.5	93.5	1.03	170.3	40.1	110.3
Group 3	26	122.7	95.1	1.23	156.0	51.7	94.8

No significance was found between the liver enzymatic activity (ALT, GGTP), serum bilirubin concentration as well as the lipid metabolism parameters and the referent values of the "no steatosis" group, table II.

Tabela II. Zaawansowanie histopatologicznych zmian wątroby w badanych grupach (wg skali METAVIR i Dixona)
Table II. Histology differentiation in each group (METAVIR and Dixon`s scale).

	Zapalenie Grading	Włóknienie Staging	Stłuszczenie Steatosis
Group 0	1.19	1.26	0.00
Group 1	1.98	2.24	0.98
Group 2	1.87	2.12	1.97
Group 3	1.84	2.25	2.92

Patients of group 3 had the highest BMI value compared to other groups and the "no steatosis" group (26, 6 and 23,9 respectively, p=0,04).

Neither viremia load nor the HCV genotype was different between the "no steatosis" group and steatosis groups (p>0,05).

Fibrosis was significantly high when comparison between control and steatosis 2 and 3 was done. More advanced liver changes of fibrosis in patients with hepatosteatosis made the subpopulations more frequently be treated with PegIFN+RBV therapy than the control one. However, control group percentage responding to the therapy was relatively higher than group 3 (1.75 and 1.62, respectively).

DISCUSSION

It has already been widely proved that progression of steatosis lesions should be precisely diagnosed and effectively treated as some of patients with underlying hepatosteatosis develop its further consequences like steatohepatitis, cryptogenic cirrhosis or even HCC in course of time. Some primary hepatotropic viruses as HCV have a direct propensity to its induction, as well. In the case, hepatosteatosis has certain prognostic and therapeutic implications which are widely of much interest. The steatosis lesions are highly prevalent in chronic hepatitis C patients and may be both the effect of the HCV activity as well as cause of the HCV therapy resistance. In contrast to this, reversion of steatohepatosis by means of effective antiviral therapy is also widely discussed. Steatosis prevalence differs in chronic HCV population. Most of the influencing factors are associated with metabolic disturbances as high BMI, diabetic history as well as lipid abnormalities ([23, 10, 11, 12]). However, there also seems to be a strong viral-dependent impact on steatosis outcome as some genetic viral factors has already been found (core protein of HCV) ([13]). Consequently, HCV genotype 1 is much more associated with steatosis as well as metabolic disturbances whereas genotype 3 directly induces steatosis lesions dependent on high viral load ([14]). In our study steatohepatosis was relatively frequent in the group of patients at a moderate degree which supports previous studies ([3, 15]). In the overall observation, hepatosteatosis ranging 5-25% of the histopathology image was dominant regardless of any other analyzed factors. However, unlike the studies, we did not observe any diabetic or obese patients (relatively low BMI=

25) in the overall group. This might also stand for the fact of no extreme steatosis (>60%) seen in our study. Consistently, we did not find any relations of viral dependent or patient self-dependent factors with steatosis degree between the "no steatosis" group and the others. There was no difference in steatosis prevalence between all groups when genotype or viral load was concerned. Although, viral load in all the population ranged millions of copies not in all cases HCV viremia was measured which might have affected low credibility of the factor. In the previous studies HCV viremia played a large part in steatosis prevalence usually in strong liaison with genotype 3 .

Liver enzyme activity revealed poor association to steatosis advance. This fact confirms some previous studies` results of the paired populations: HCV alone and HCV with superimposed steatosis (non-alcoholic steatosis) ([16]). However, in another study higher transaminases activity was seen in all HCV-stetatosis group ([17]). The discrepancy still leaves an issue of the diagnostic role of ALT level in NASH unclear. What is more, unlike the studies we did not find any significant lipid or glucose abnormalities in all the subpopulations. However, a group of steatosis 3 patients had the highest BMI which suggested that the factor has some influence on the lesions development.

Alcohol intake may play a large part in steatosis outcome what has already been largely found (3). However, contrary to these findings some authors suggest alcohol may play a minor part in the process even when high degree alcohol intake was noted (17). In the study we also tried to analyze potential alcohol consumption influence upon the steatosis development. Unfortunately, since this was a retrospective study we did not manage to draw the credible history of alcohol consumption out of the medical record. Thus, we are not able to unequivocally refer our groups to the influence of alcohol consumption on steatosis .

In our study highly significant relation of steatosis and fibrosis was found what supports others` results ([18],[19],[20]). However, in contrast to the analyses we found significant relation between degree of the inflammatory infiltrates (grading) and steatosis within all groups. The fact may suggest influence of other factors affecting inflammatory response in our population (inducing steatohepatitis) as concomitant medications or alcohol intake (poor credibility of medical record data). In our center, histology fibrosis staging is the most important factor qualifying patients to start antiviral therapy. Consequently, as staging score was higher in patients with high degree of steatosis, the patients with any steatosis degree were more frequently treated. However, unlike similar studies there was no difference in the anti-HCV therapy response observed when we compared the groups with steatosis to the control group. The result is not consistent as in large cohort studies there was strong association found between steatosis and response to the therapy regardless of fibrosis stage ([21,18,22]). The revelation needs further observation on a larger group of treated patients.

CONCLUSIONS

In the analyzed group the most advanced hepatosteatosis changes were observed among patients with higher BMI score. We did not find any significant abnormalities in sex and age of patients. No significant lipid metabolism abnormalities seen in our population may suggest there hardly were any overlaps of HCV and NAFL when a relation to the parameters was done. Differences between our Silesian group results and literature data patients from other countries calls for a larger prospective study in the Polish population.

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PRZEGLĄD PACJENTÓW ZE STŁUSZCZENIEM WĄTROBY I PRZEWLEKŁYM WIRUSOWYM ZAPALENIEM WĄTROBY TYPU C

STRESZCZENIE

Dane epidemiologiczne wskazują, że stłuszczenie wątroby spotykane jest często i dotyczy ponad 20 % populacji krajów wysoko i średnio rozwiniętych, a współistnienie przewlekłego wirusowego zapalenia wątroby typu C jest czynnikiem szczególnie predysponującym do jego rozwinięcia. W badaniu analizowaliśmy rodzaj zaburzeń biochemicznych, metabolicznych i danych wirusologicznych pacjentów z przewlekłym wirusowym zapaleniem wątroby typu C zależnie od obecnych zmian stłuszczeniowych wątroby według obowiązujących systemów ich oceny.

Materiał i metody: Obserwacją objęto pacjentów z potwierdzonym dodatnim wynikiem HCV-RNA w surowicy oraz różnego stopnia zaawansowaniem zmian stłuszczeniowych wątroby hospitalizowanych w Klinice Chorób Zakaźnych, Wątroby i Nabytych Niedoborów Immunologicznych, hospitalizowanych w latach 2003-2006 .

Populacja została podzielona na 4 grupy w zależności od stopnia zaawansowania zmian stłuszczeniowych w biopsji wątroby ocenionej w skali Dixona i analizowana retrospektywnie w zakresie stwierdzanych zaburzeń biochemicznych, metabolicznych i poszczególnych parametrów wirusologicznych (w tym podjętych decyzji terapeutycznych i odpowiedzi na leczenie) w wyodrębnionych podgrupach. W analizie statystycznej stosowano test T- studenta, testy korelacji programu STATISTICA PL v 5.1.

Wyniki. W analizowanej grupie odsetek pacjentów z przewlekłym wirusowym zapaleniem wątroby i stłuszczeniem wątroby wyniósł 80.76%, przy czym przeważali pacjenci z 2. stopniem stłuszczenia (36,1%) . Nie stwierdzono istotnych różnic w aktywności enzymów wątrobowych (ALAT, GGTP), stężeniu bilirubiny i lipidów pomiędzy grupami badanymi i grupa kontrolną. Pacjenci z grupy 3. charakteryzowali się najwyższym wskaźnikiem BMI (p=0.04). W podgrupie tej występował też istotnie najwyższy stopień zaawansowania włóknienia w wątrobie (p= 0.0001)

Wnioski. Stopień zaawansowania zmian stłuszczeniowych w wątrobie może wiązać się z szybciej postępującym włóknieniem u pacjentów zakażonych HCV. W naszym badaniu nie wykazano jednak istotnych zaburzeń metabolicznych, ani obciążającego wywiadu chorobowego przewlekłych chorób układu krążenia u osób z bardziej zaawansowanymi zmianami stłuszczeniowymi. Może to sugerować pewne odrębności naszej populacji, co jednak wymaga analizy na większym materiale.

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