

CLINICAL AND HISTOLOGICAL ASPECTS IN CHRONIC HEPATITIS C

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Abstract

Hepatitis C affects approximately 160 million people, or about 2.35% of the general population of the globe [Lavanchy, 2011]. In Europe it is estimated about 12 million people infected with C. About 10 % of them are located in Romania, occupying the 1st place in HCV morbidity and 4th place as mortality rate for liver disease. It is estimated that Romania will increase significantly by 2030 the number of patients with hepatitis C cirrhosis and hepatocellular carcinoma and liver disease deaths, thus implying significant social and medical costs [Gheorghe L., 2009]. Although liver biopsy is no longer the gold standard in assessing liver fibrosis continues to be the method used in research on virus C.

Key words: C virus, chronic hepatitis C, liver fibrosis.

INTRODUCTION

Infection with hepatitis C virus (HCV) is a major public health problem worldwide due to asymptomatic evolution to cirrhosis and liver cancer and due to the number of people infected in Romania, where the economic and social deficits makes each one to represent potential sources of infection.

MATERIAL AND METHOD

The study was conducted by prospective epidemiological investigation on clinical and laboratory data recorded in a group of 274 patients diagnosed with chronic hepatitis C. At all patients included in the study was completed survey sheet, were performed functional liver tests (CBC, ALT, AST, GGT, Bi T, Ac HCV), imaging methods, liver biopsy. Were used only biopsy cylinders larger than 5 mm and which have at least 4 porta spaces. The liver tissue fragments samples were fixed in 10 % formalin buffered at neutral pH, followed by inclusion in paraffin. From the obtained paraffin blocks were performed serial sections which were stained in each case Hematoxylin-eosin, Masson trichrome, Van Giesen and were interpreted with the Knodell and Metavir score.

RESULTS AND DISCUSSIONS

The group of the studied patients includes 274 patients aged 19 and 69 years and the average age of 46.03 years \pm 10.02 years, with slight predominance of the female gender (58,76 %), more than half of patients (57,66 %) come from Oradea. About 2/3 of patients (66.06 %), or 181 patients aged 36 to 55 years.

The age group (46-55 years – 33.58%) comprise the patients who in '90 were adults (aged 30 to 45 years) with an increased risk of transmission of virus C infection due to the economical and social factors registered in Romania at the time. Most patients were at the first puncture (88.32 %), 9.49 % in the second puncture, at 5 patients were performed 3 or 4 punctures (1.82%). 15 patients (4.48 %) had previously received treatment with Intron and Ribavirin, but were non-responders. Most patients (24.45 %) had portal fibrosis without septa and mild inflammatory activity. None showed absent fibrosis and severe activity. One patient experienced a portal fibrosis with numerous septa and absent activity (Table 1).

Table 1

Distribution of patients by degree of fibrosis

	A0	%	A1	%	A2	%	A3	%	Total	%	%
F0	8	2,92	7	2,55	2	0,73	0	0	17	6,2	46,71
F1	14	5,11	67	24,45	21	7,66	9	3,28	111	40,51	
F2	1	0,36	28	10,22	54	19,71	17	6,2	100	36,5	53,29
F3	1	0,36	9	3,28	21	7,66	15	5,47	46	16,79	
Total	24	8,76	111	40,51	98	35,77	41	14,96	274	100	100

Were examined between 4 and 24 porta spaces (PS), with an average of 7.8 porta spaces and a standard derivation of 3.82, mostly between 4 and 9 spaces examined. In 36.5 % of patients (100) were studied 6-9 porta spaces, although the biopsy cylinder length was between 1 and 1.5 cm at 42,34 % of them (116 patients). The fewest examined porta spaces (6.06 ± 1.38) were at the patients without fibrosis (F0), and the most examined porta spaces were at patients with severe activity (A3) 10.29 ± 4.22 and severe fibrosis (F3) 8.6 ± 3.43 . At the patients without fibrosis (F0) were not collected the biopsy cylinders over 15 mm length. The minimum biopsy length was greater than 0.5 cm. The biopsy cylinders between 1.6 and 2 inches (34.67 %) were mainly at those with F3 (20 patients), F0 (6 patients) and A1 (40 patients).

The average age was the highest for the degree of fibrosis F3 (46,01 years) and the severe activity (47,68 years). In the chronic HCV infection mild disease (A0F0) the average age was much lower (41.12 ± 9.9 years) than the average age of the severe disease (A3F3) 49.93 ± 9.71 years.

Table 2

Distribution of patients by degree of fibrosis and genres

		Male				Female				Total		Total	
		Nr.	%	Nr.	%	Nr.	%	Nr.	%	Nr.	%	Nr.	%
F0	A0	3	1,09	7	2,55	5	1,82	10	3,65	8	2,92	17	6,2
	A1	3	1,09			4	1,46			7	2,55		
	A2	1	0,36			1	0,36			2	0,73		
F1	A0	6	2,19	47	17,15	8	2,92	54	19,71	14	5,11	111	40,51
	A1	27	9,85			30	10,95			67	24,45		
	A2	8	2,92			13	4,74			21	7,66		
	A3	6	2,19			3	1,09			9	3,28		
F2	A0	0	0,00	42	15,33	1	0,36	58	21,17	1	0,36	100	36,5
	A1	11	4,01			17	6,20			28	10,22		
	A2	27	9,85			27	9,85			54	19,71		
	A3	4	1,46			13	4,74			17	6,20		
F3	A0	1	0,36	17	6,20	0	0,00	29	10,58	1	0,36	46	16,79
	A1	6	2,19			3	1,09			9	3,28		
	A2	5	1,82			16	5,84			21	7,66		
	A3	5	1,82			10	3,65			15	5,47		
Total		113	41,24	113	41,24	161	58,76	161	58,76	274	100	274	100

Of all the 274 patients, 17 patients (6,2%) had no fibrosis, 111 patients (40,51 %) had fibrosis limited at the portal space, without septa (F1), 100 (36,5 %) had portal fibrosis with rare septa (F2), while 16,79 % (46) had numerous septa without cirrhosis (F3). The emphasizing of the degrees of fibrosis occurs in both sexes, maintaining the greater differentiation for female. Statistically there is no significant difference between men and women according to the degree of fibrosis ($\chi^2 = 0.99$, sig = 0.801). There was no increased activity (A0) at 24 patients (8.76 %), 111 (40.51 %) had mild activity (A1), 98 (35.77 %) patients had moderate activity (A2) and 26 patients (14.96 %) had a severe necro-inflammatory activity (A3). The emphasizing of the degrees of fibrosis occurs in both sexes, maintaining the greater differentiation for female. Statistically there is no significant difference between men and women according to the necro-inflammatory activity ($\chi^2 = 0.59$, sig = 0.898).

Table 3

Distribution of necro - inflammatory activity by gender

		Male				Female				Total			
		Nr.	%	Nr.	%	Nr.	%	Nr.	%	Nr.	%	Nr.	%
A0	F0	3	1,09	10	3,69	5	1,82	14	5,11	8	2,92	24	8,76
	F1	6	2,19			8	2,92			14	5,11		
	F2	0	0			1	0,36			1	0,36		
	F3	1	0,36			0	0			1	0,36		
A1	F0	3	1,09	47	17,88	4	1,46	64	23,36	7	2,55	111	40,51
	F1	27	9,85			40	14,6			67	24,5		
	F2	11	4,01			17	6,2			28	10,2		
	F3	6	2,19			3	1,09			9	3,28		
A2	F0	1	0,36	41	14,23	1	0,36	57	20,8	2	0,73	98	35,77
	F1	8	2,92			13	4,74			21	7,66		

	F2	27	9,85			27	9,85			48	17,5		
	F3	5	1,82			16	5,84			27	9,85		
A3	F1	6	2,19	15	5,47	3	1,09	26	9,49	9	3,28	41	14,96
	F2	4	1,46			13	4,74			17	6,2		
	F3	5	1,82			10	3,65			15	5,47		
Total		113	41,24	113	41,24	161	58,76	161	58,76	274	100	274	100

The age group over 66 years, showed no cases of hepatitis without fibrosis or fibrosis stages (F0) and no case without necro-inflammatory activity (A0) and the age group up to 25 years showed no F3 fibrosis any case. The increased degree of fibrosis is associated with the increased age groups. The degree of fibrosis is not associated with the patient age ($\chi^2 = 20.84$, sig = 0.142) and the necro-inflammatory activity is not associated with the patient age group ($\chi^2 = 12.18$, sig = 0.665). Regard to changes in laboratory analyses, the average leukocyte count (6034.49 ± 1094.79), erythrocyte count ($4\ 640000 \pm 490000$) and platelet count (223000 ± 61400) falls within the normal range. The increased degree of fibrosis is accompanied by a slight decrease in the average leukocyte and platelet count, but not in the erythrocyte count. The increased GGT is associated with chronic hepatitis C, because the C virus multiplies including in the bile canaliculi cells.

Table 4

The mean values of ALT and GGT in degrees of fibrosis

	F0	F1	F2	F3
ALAT	46,06±33,96 (15-124)	68,59±55,67 (13-341)	85,28±60,35 (7-255)	102,61±66,76 (36-433)
ASAT	35,47±21,31 (12-82)	50,76±38,82 (12-291)	63,34±51,57 (9-400)	80,41±47,83 (34-285)
GGT	48,65±51,45 (15-230)	62,97±64,54 (5-379)	69,61±59,15 (12-406)	85,8±60,69 (15-362)

The increased degree of fibrosis is associated with the increased average value of hepatocytolysis syndrome and GGT, in the F3 degree of fibrosis was recorded the highest value ALT (433 U / l), then 341 U / l in the F1 degree of fibrosis F1 (Fig. 1).

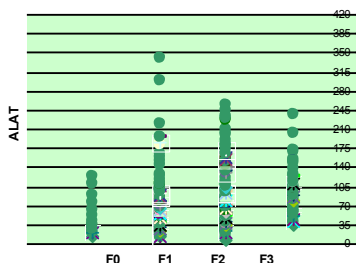


Fig.1 ALT levels and degrees of fibrosis

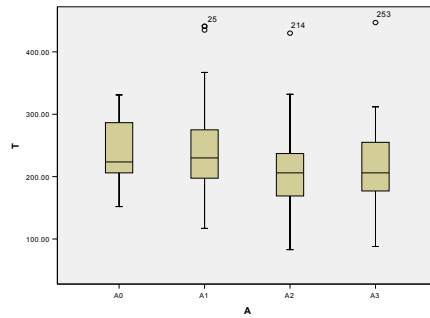


Fig. 2 ALT and necro - inflammatory activity

The increased degree of necro-inflammatory activity is not associated with the increased hepatocytolysis syndrome (Fig. 2) .

The statistical computations reveals the existence of correlations between ALT and GGT, the cholestasis level is directly proportional to ALT. From the statistical point of view, there is no a linear relationship between the values of ALT and GGT, however, the correlation coefficient value ($R^2=0.146$) shows a very little correlation between two variables, compared with the following data (Fig. 3).

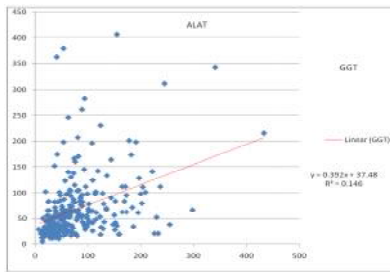


Fig.3 ALAT-GGT

ALT values do not correlate with the leukocytes counts, irrespective of the chosen function type (Fig. 4).

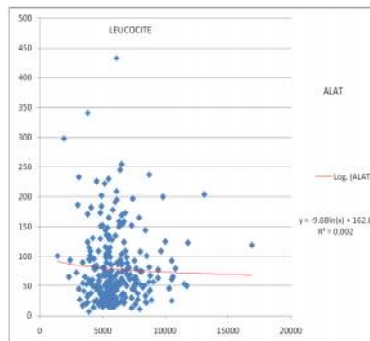


Fig.4 ALAT-WBC number

There is no a linear relationship between ALT values and platelet counts, the correlation coefficient has a value close to 0 (Fig. 5).

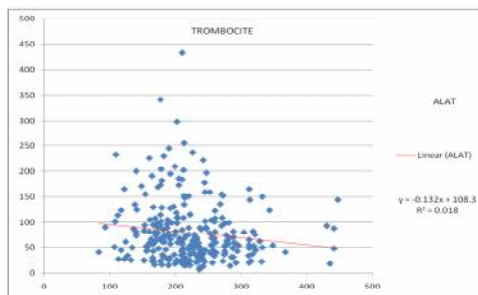


Fig.5 ALAT-platelet number

GGT values do not correlate with the platelet counts (Fig. 6).

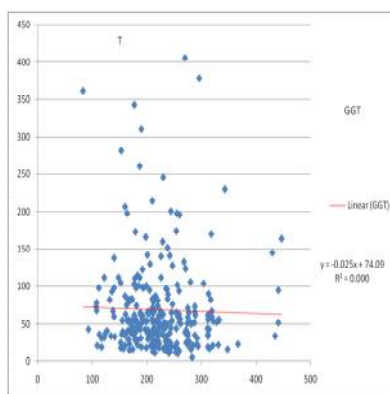


Fig.6 Platelet number-GGT

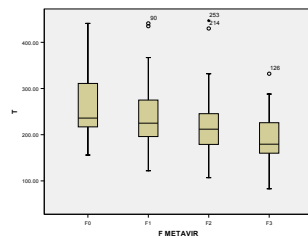


Fig.7 Platelets degrees of fibrosis

The increased degree of fibrosis is associated with the decreased platelet counts (Fig. 7).

Table 5

Histological comparison between minimal and severe fibrosis stage

	Chronic hepatitis C	Chronic hepatitis C
	minimal form	Severe form
	(F0-F1)	(F2-F3)
	128 (46,72)	146 (53,28)
Nr. of portal spaces (media±DS)	14±14,14	7,76±3,72
<11	102 (79,69)	117 (80,14)
≥11	26 (20,31)	29 (19,86)
Lobular necrosis		
0<1 focar	72 (26,28)	34 (12,41)
1 cel puțin un focar	46 (16,79)	81 (29,56)
2 câteva focare	10 (3,65)	31 (11,31)
Piece meal necrosis		
0 absent	86 (31,39)	63 (22,99)
1 focal în câteva	27 (9,85)	47 (17,15)
2-3 difuză în câteva/toate	15 (5,47)	36 (13,14)
Portal inflammation		
0+1 ușoară	56 (20,44)	15 (5,47)
2+3 moderată/severă	72 (26,28)	131 (47,81)
Steatosis		
absentă	62 (22,63)	61 (22,26)
prezentă	66 (24,09)	85 (31,02)
Necro-inflammatory activity		
A0 absent	22 (8,03)	2 (0,73)
A1 ușor	74 (27,01)	37 (13,5)
A2 moderat	23 (8,39)	75 (27,37)
A3 sever	9 (3,28)	32 (11,68)

The chronic hepatitis C is associated with the expansion of the lobular necrosis, the appearance of the diffuse piecemeal necrosis, the moderate and severe portal inflammation, often being severe active (A2, A3). In our group of the studied patients, the hepatic steatosis was present in approximately 55.11 % of cases (151 patients), of which 58.94 % were women, maintaining the difference presented in study. The hepatic steatosis was described as micro-vacuolar in 23 cases, macro-vacuolar in 58 cases and mixed in 70 cases. The emphasizing of the degree of steatosis determines the acceleration of the degree of fibrosis.. The most cases of severe steatosis is accompanied by a high F3 fibrosis.

CONCLUSIONS

- Were examined 274 patients aged 19 to 69 years and the average age of 46.03 years ± 10.02 years, with a slight predominance of the female gender (58.76 %), about two-thirds of patients (66,06 %), i.e. 181 patients with ages between 36 and 55 years.
- The most patients (24.45 %) had a portal fibrosis without septa and a mild inflammatory activity.
- The age group (46-55 years) was the most common (33.58 %)

- The most often was collected between 1.6 and 2 inches (34.67 %), especially at those with F3 (20 patients), F0 (6 patients) and A1 (40 patients).
- The fewest examined porta spaces (6.06 ± 1.38) were at the patients without fibrosis (F0), and the most examined porta spaces were at patients with severe activity (A3) 10.29 ± 4.22 and severe fibrosis (F3) 8.6 ± 3.43 .
- In the chronic HCV infection mild disease (A0F0) the average age was much lower (41.12 ± 9.9 years) than the average age of the severe disease (A3F3) 49.93 ± 9.71 years.
- Statistically there is no significant difference between men and women according to the degree of fibrosis ($\chi^2 = 0.99$, sig = 0.801) or by the necro - inflammatory activity ($\chi^2 = 0.59$, sig = 0.898).
- The increased degree of fibrosis is associated with the increased age groups.
- The increased degree of fibrosis is accompanied by a slight decrease in the average leukocyte and platelet count, but not in the erythrocyte count.
- The increased degree of fibrosis is associated with the increased average value of hepatocytolysis syndrome and GGT.
- The increased degree of necro-inflammatory activity is not associated with the increased hepatocytolysis syndrome
- The increased ALT is statistically correlated with GGT level, but there is no a linear relationship between ALT values and GGT values, there is no a linear relationship between ALT values and platelet counts and GGT values do not correlate with the platelet counts.
- The chronic hepatitis C is associated with the expansion of the lobular necrosis, the appearance of the diffuse piecemeal necrosis, the moderate and severe portal inflammation, often being severe active.

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