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Occult Hepatitis (B) Virus Infection among Patients Who Have Received Direct-Acting Antiviral Drugs for Hepatitis (C) Virus infection

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Abstract

Background: Occult HBV infection (OBI) is defined as detecting HBV DNA (in blood and/or liver tissue) without detectable HBsAg with or without anti-HBc or anti-HBs. The clinical importance of occult HBV infection may be noticed in the following clinical contexts: transmission of HBV infection after blood transfusion and organ transplantation, reactivation of chronic hepatitis B with exposure to immunosuppression, and implication in chronic liver diseases (CLD), especially with cryptogenic chronic liver diseases, HCV-related chronic liver diseases, and hepatocellular carcinoma.

Aim of the Study: To determine the prevalence of occult HBV infection in HCV related chronic liver disease and to investigate the possible role of occult HBV infection in patients who have received Direct Acting Antivirals (DAAs) for HCV related chronic liver disease by clarifying the clinical and laboratory features of occult HBV infection in HCV related chronic liver disease.

Patients and Methods: The present study was conducted in the gastroenterological Department, Cairo Fatemic Hospital. This study included 198 patients who have received treatment for the hepatitis C virus. All patients were subjected to the following: thorough history taking, careful clinical examination, abdominal ultrasonography, complete blood picture, liver function tests (total and direct bilirubin, total plasma proteins and albumin, and liver enzymes, and coagulation tests

Results: We found that occult HBV correlated with the clinical picture, laboratory investigations for all the studied patients.

Conclusion: Occult hepatitis B was detected in a significant number of patients using HBc IgG so check HBc IG status before initiating pan-oral DAAs therapy is important, as occult HBV may contribute to chronic liver damage, development of HCC and flare of liver enzyme and role of OBI on the outcome of DAAs treatment for HC OBI is a life-threatening public health problem worldwide.

Keywords: Occult; Hepatitis (B); Antiviral

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1. Introduction

Occult hepatitis B (OBI) was defined as the detection of hepatitis B virus (HBV) DNA in the liver (with or without HBV DNA in serum) without HBsAg **[1].**

The prevalence of OBI varies from region to region worldwide. This variability relies upon the sensitivity of HBV DNA detection assays, the sample size, and the detection of HBV DNA in liver tissue and serum by nested PCR or real-time PCR. The prevalence of OBI varies from 1% to 87% in different regions of the world **[2],** there is no standard assay for diagnosis of OBI in liver tissue or in serum, and the only reliable method is the detection of HBV DNA by nested PCR or real-time PCR **[3].** The HBsAg gene mutations have been observed among patients coinfected with hepatitis C virus (HCV) **[4].**

It has been described that about one-third of patients with chronic HCV infection had detectable serum HBV DNA but undetectable HBsAg **[5].** When the coexistence of both HBV and HCV genomes occurs in the same hepatocyte, the replication of HBV is inhibited due to the interference of HCV molecules, which therefore results in the creation of OBI with low replication of HBV DNA **[6].**

The presence of OBI in chronic HCV-infected patients increases the risk of HCC **[7].** Blood transfusion is the leading risk factor for transmission of OBI, and the prevalence of OBI among blood donors varies from country to country, provided that the screening of blood donors is done with less security **[8].**

It is recommended that all the patients on Hemodialysis (HD) be routinely screened for viral blood-borne infections (HBV, HIV, and HCV), including OBI, which is evaluated by evaluation of quantitative HB DNA that was found to be the most efficient method to evaluate OBI in HD patients **[9].**

Occult hepatitis B virus infection (OBI) has been regarded as an additional risk factor for the progression of liver cirrhosis and HCC **[10].**

The rate of cryptogenic liver diseases varies significantly in different regions of the world. Patients with long-term persistent ALT abnormality or with the lack of overt viral detection and autoimmune markers have been shown to be positive for HBV DNA (OBI) **[11].**

The prevalence of OBI among cirrhotic patients varies from region to region worldwide and The mechanism of liver damage due to OBI is still not well elucidated, but there are some data that described the persistence and transcription of HBV cccDNA in hepatocytes and, subsequently, production of cytokines, such as TNF-α and interferon-γ may result in damage to hepatocytes **[12].**

Most findings described that OBI is an important risk factor for hastening the progression of liver disease and the development of cirrhosis and HCC **[13].** In

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addition, HBV DNA has been found to be integrated within the host chromosomes of individuals with HCC **[14].**

Liver transplantation is the only option for patients with end-stage chronic liver disease. But in liver transplant recipients with OBI, the reactivation of HBV is enhanced by the induced immunosuppression factors and rapidly leads to graft failure and death **[15].**

For the management and prevention of the consequences of OBI in organ transplant recipients, it is suggested that the screening of HBV DNA be carried out in both donors and organ transplant recipients by highly sensitive molecular means as (highly sensitive nested PCR or by a real-time PCR technique that can detect fewer than 10 copies of HBV DNA using the oligonucleotide primers specific for different HBV genomic regions and complementary to highly conserved nucleotide sequences)**[16].** Also, Health care workers are more often at high risk of HBV infection/OBI than the general population**.** Most of the individuals with OBI are clinically asymptomatic and remain undiagnosed unless a sudden development of cirrhosis or HCC occurred **[17].**

2. Patients and Methods

2.1. Patients:

2.1.1. The site of the study

The ethical committee of our institution approved this study to be conducted at Cairo Fatemic Hospital Cairo governorate, Egypt. It included 198 chronic HCV patients who have completed treatment of HCV with DAAs.
The diagnosis of HCV was confirmed by the detection of anti-HCV antibodies and HCV RNA.

2.1.2. Inclusion criteria

We included patients with such criteria HBsAg: Negative, Patients, with elevated liver enzymes, Positive for anti-HCV and HCV RNA positive, Signed written informed consent for the study.

2.1.3. Exclusion criteria

We excluded patients with such criteria; Patients who refused to be enrolled in the study, Any other cause of chronic liver disease other than HCV, Overt HBV Co-infection, Drug-induced liver disease., Ischemic cardiovascular insult within the last six months., Immunologically mediated diseases, Patients with organ transplants., Substance abuse (abstention for the last 12months) and Immunosuppressive drugs.

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All the studied patients were subjected to the following:

1. History taking, with special emphasis on history suggestive chronic liver disease attack of hematemesis, Hepatic melena encephalopathy. BMI, physical examination.

2. Clinical examination

Signs of chronic liver disease such as jaundice, lower limb edema, etc. Abdominal examination ultrasonography of the liver, splenomegaly, prescience of ascites or not

3. Biochemical assessment

Liver function tests, kidney functions, complete blood counts, anti-HB c total antibodies were detected by a rapid immunoassay, while anti-HBs antibodies were determined by the electrochemiluminescence immune assay "ECLIA" technique using commercially available kits

Patient Monitoring

All the patients attended the Viral Hepatitis outpatient Clinic for monitoring during treatment. Patients were assessed at weeks 0, 2, 4,8, and week 12 of treatment

At each review, laboratory tests were performed, including serum ALT and AST, bilirubin, full blood count, and serum creatinine. Bodyweight and symptom checklist were recorded at each visit. Quantitative serum HCV-RNA was determined at baseline and at week 12. Qualitative HCV-RNA was determined at weeks 24 and 48.

2.2. Methods:

This observational retrospective cross-sectional study from May 2020 to October 2020 evaluates 198 consecutive adults treated with pan oral DAAs, including Sofosbuvir, Daclatasvir, and Ribavirin (SVR).

SVR at 12 weeks after the end of treatment was achieved in all patients, apart from hepatitis, no significant serious adverse events were reported in all patients treated with DAAs

2.2.1. Patient groups

This study was done on 198 patients who have received the new treatment for HCV

2.2.2. Statistical Analysis

Data collected throughout history, essential clinical examination, laboratory investigation, and results coded, entered, and analyzed using Microsoft Excel Software. Data were then imported into a statistical package for social science (SPSS) for analysis. Data are represented in tables either as mean ± standard

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deviation and range or the number of cases (percentage of the total count of the instances (n %). Reported results significance level was set to $P < 0.05$

Qualitative data were expressed as frequency and percentage. The following tests were done: Independent-samples t-test of significance was used when comparing two means. Chi-square (x2) test of significance was used to compare proportions between qualitative parameters. Pearson's correlation coefficient (r) test was used to assess the degree of association between two sets of variables. the p-value was considered significant as Probability (P-value): P-value 0.05 was deemed to be insignificant

P-value <0.05 was considered significant.

P-value <0.001 was considered highly significant.

P-value >0.05 was considered insignificant.

3. Results

The baseline characters of all patients are described in:

Table (5): Description of sex, age &BMI in all studied patients.

		Studied patients (N = 198)	
Sex	**Male**	110	55.6%
	Female	88	44.4%
Age (years)	**Mean ±SD**	52.7 ± 9.9	
	Min – Max	22 – 74	
BMI (kg/m^2)	**Mean ±SD**	27.2 ± 3.8	
	Min – Max	19 – 38	

This table shows the description of sex, age & BMI in all studied patients. As regard sex, there were 110 males (55.6%) and 88 females (44.4%) in all studied patients. The mean age in studied patients was 52.7 ± 9.9 years with a minimum age of 22 years and maximum age of 74 years. As regards BMI, the mean BMI in studied patients was 27.2 ± 3.8 kg/m^2 with a minimum BMI of 19 kg/m^2 and a maximum BMI of 38 kg/m^2.

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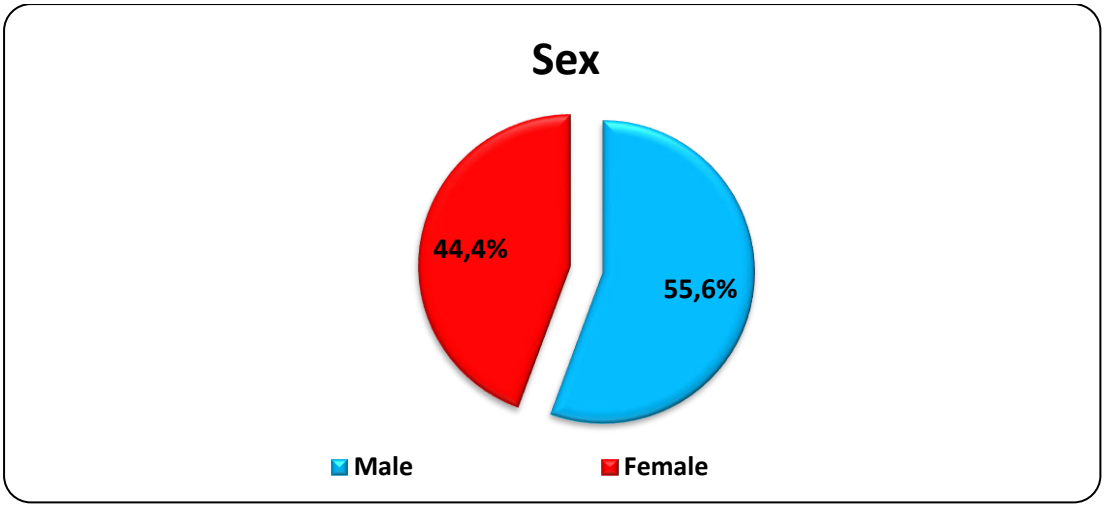

Figure (6): Distribution of sex in all studied patients.

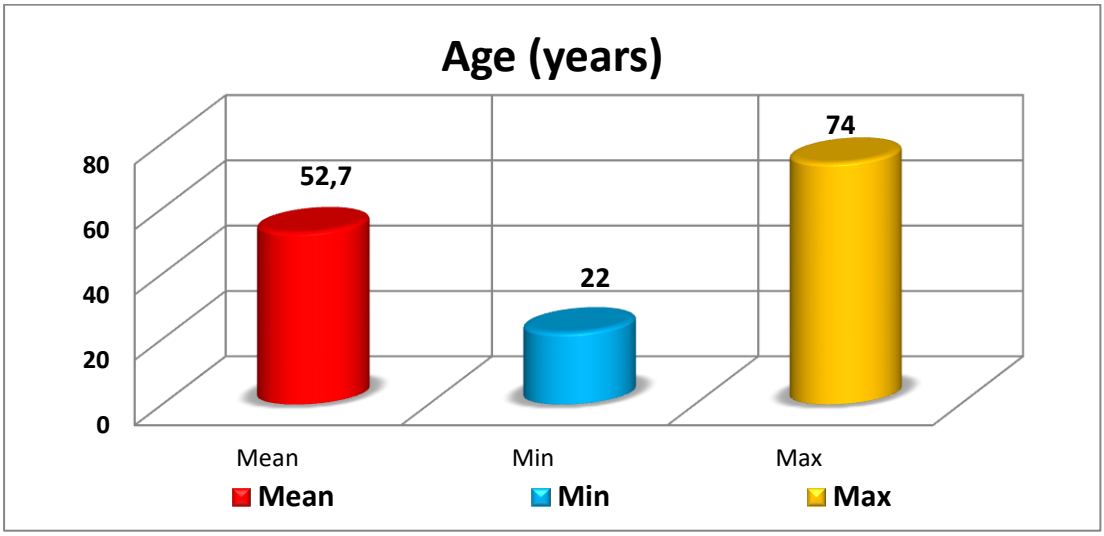

Figure (7): Distribution of age in all studied patients.

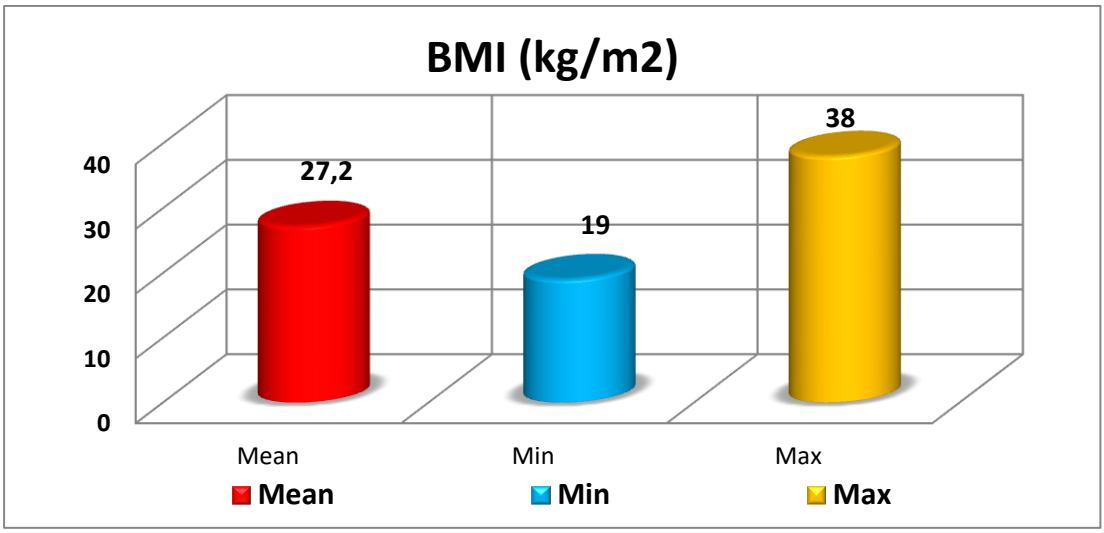

Figure (8): BMI in all studied patients.

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Table (6): complain in all studied patients.

		Studied patients (N = 198)	
Complain	**Follow up**	74	37.4%
	Abdominal pain	58	29.3%
	Epigastric pain	6	3%
	Hematemesis	12	6.1%
	Jaundice	22	11.1%
	Easy fatigability	12	6.1%
	Weight loss	8	4%
	Melena	4	2%
	Dyspnea	2	1%

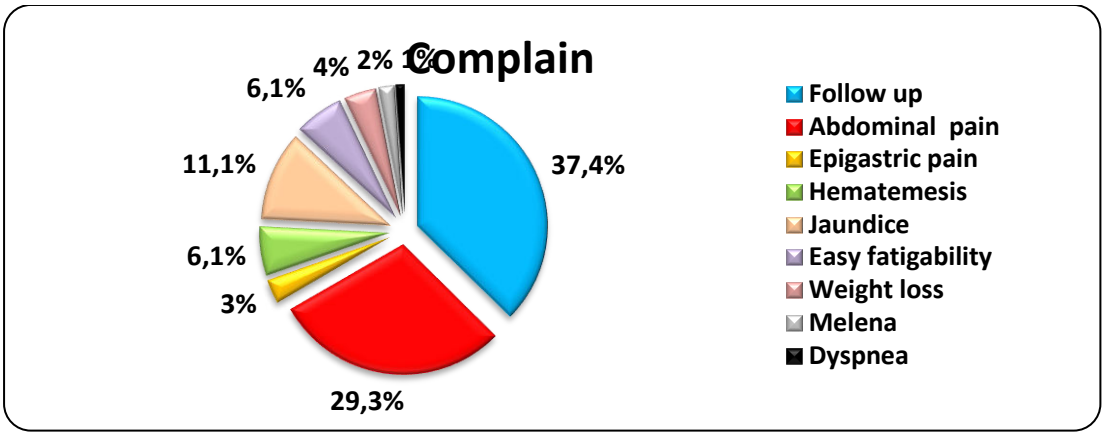

Figure (9): Complain in all studied patients.

Table (7): Symptoms in all studied patients.

Symptoms		Studied patients (N = 198)	
		Number	%
Hematemesis	**No**	**162**	**81.8%**
	Yes	**36**	**18.2%**
Melena	**No**	**158**	**79.8%**
	Yes	**40**	**20.2%**
Hepatic encephalopathy	**No**	**182**	**91.9%**
	Yes	**16**	**8.1%**
Lower limb edema	**No**	**142**	**71.7%**
	Yes	**56**	**28.3%**
Abdominal pain	**No**	**100**	**50.5%**
	Yes	**98**	**49.5%**

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Jaundice	**No**	**156**	**78.8%**
	Yes	**42**	**21.2%**

This table shows the description of symptoms in all studied patients. There were 36 patients (18.2%) with hematemesis, 40 patients (20.2%) with melena, 16 patients (8.1%) with hepatic encephalopathy, 56 patients (28.3%) with lower limb edema, 98 patients (49.5%) with abdominal pain and 42 patients (21.2%) with jaundice in the studied patients.

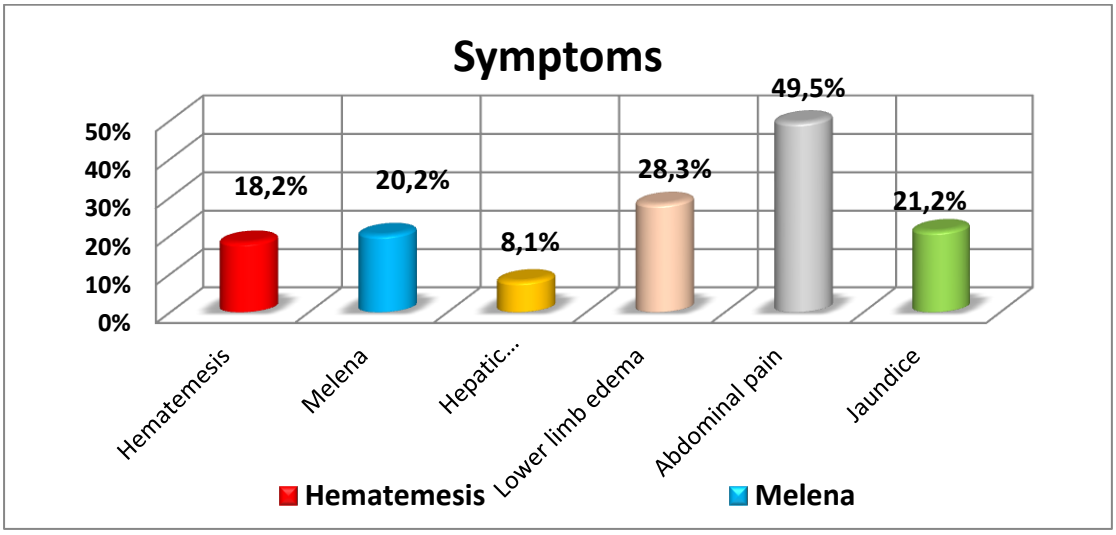

Figure (10): Symptoms in all studied patients.

Table (8): Comorbidities in all studied patients.

Co-morbidities		**Studied patients (N = 198)**	
DM	**No**	161	81.3%
	Yes	37	18.7%
HTN	**No**	120	60.6%
	Yes	78	39.4%
Blood transfusion	**No**	110	55.6%
	Yes	88	44.4%
Surgery	**No**	94	47.5%
	Yes	104	52.5%

This table shows the description of comorbidities in all studied patients. There were 37 patients (18.7%) with DM, 78 patients (39.4%) with HTN, 88 patients (44.4%) with a history of blood transfusion, and 104 patients (52.5%) with a history of previous surgery in the studied patients.

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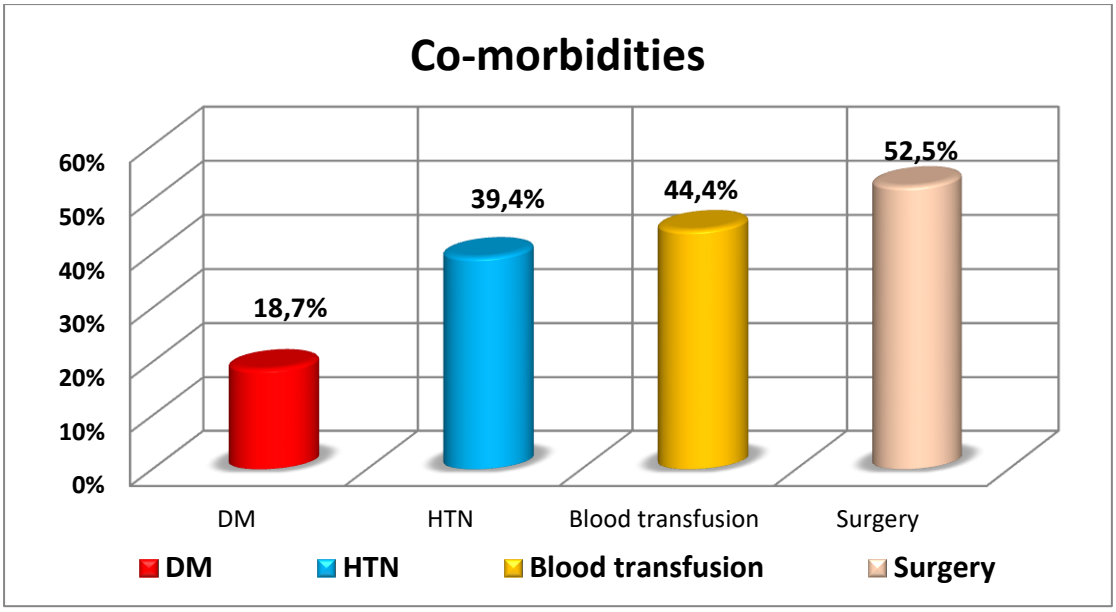

Figure (11): Comorbidities in all studied patients.

Table (9): Laboratory data in all studied patients.

Hb	6.5	16.5	11.7	1.8
HCT	24	56	38.4	5.4
WBCs	2	17.1	6.2	2.6
PLTs	44	377	169.8	83.8
PT	12	28	15.4	2.9
INR	0.8	2.58	1.3	0.3
NA	125	147	135.6	4.0
K	2.7	5	3.9	0.4
Urea	13	165	37.2	22.2
Creat	0.4	3.5	0.9	0.4
Total bilirubin	0.4	13	2.3	2.4
Direct bilirubin	0.1	8.93	1.2	1.4
AST	19	379	69.9	51.5
ALT	20	776	90.5	110.0
ALP	28	217	84.1	42.4

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GGT	14	323	79.0	70.1
ALB	2.1	4.7	3.6	0.7
TP	4.7	7.6	6.0	1.1
AFP	2	1600	66.7	196.3

Table (10): Abdominal U/S in all studied patients

Abdominal U/S		**Studied patients (N = 198)**	
Liver status	Normal	68	34.3%
	Bright	46	23.2%
	Cirrhosis	84	42.4%
Splenomegaly	No	98	49.5%
	Yes	100	50.5%
Ascites	No	136	68.7%
	Mild	14	7.1%
	Moderate	34	17.2%
	Marked	14	7.1%
PV diameter	Mean ±SD	13.2 ± 1.6	
	Min - Max	10 – 16.7	
Focal lesion	No	160	80.8%
	Yes	38	19.2%

This table shows the description of abdominal U/S in all studied patients. **As regard liver status,** there were 68 patients (34.3%) of normal liver, 46 patients (23.2%) of the bright liver, and 84 patients (42.4%) of the cirrhotic liver in the studied patients. **As regard splenomegaly,** there were 100 patients (50.5%) with splenomegaly. **As regard ascites,** there were 14 patients (7.1%) with mild ascites, 34 patients (17.2%) with moderate ascites, and 14 patients (7.1%) with marked ascites, while there were 136 patients (68.7%) had no ascites. **As regards PV diameter,** the mean PV diameter in studied patients was 13.2 ± 1.6

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mm with a minimum PV diameter of 10 mm and a maximum PV diameter of 16.7 mm. There were 38 patients (19.2%) with a focal lesion in the studied patients regarding focal lesions.

Table (11): Description of HBC Ig in all studied patients

		Studied patients (N = 198)	
HBC Ig	**Negative**	146	73.7%
	Positive	52	26.3%

This table shows the description of HBC Ig in all studied patients. There were 146 patients (73.7%) HBC Ig negative and 52 patients (26.3%) HBC Ig positive in the studied patients.

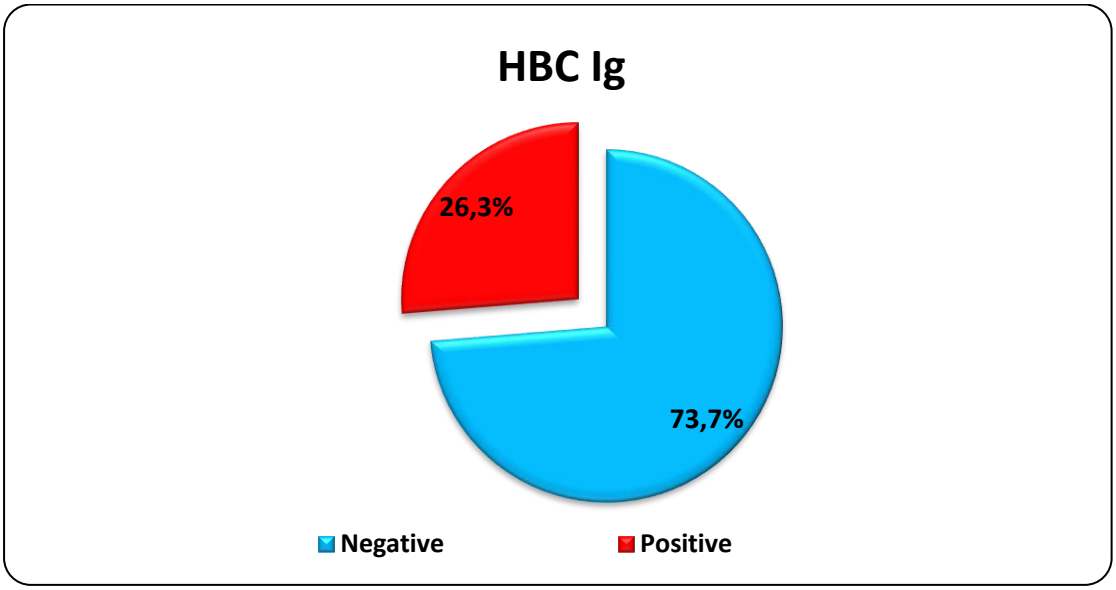

Figure (12): Description of HBC Ig in all studied patients.

There were 146 patients (73.7%) HBC Ig negative and 52 (26.3%) HBC Ig positive in the studied patients.

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Table (12): Comparison of demographic data as regard HBC Ig result.

		HBC Ig		Stat. test	P-value
		Negative (N = 146)	Positive (N = 52)		
Age (years)	Median	52	58.5	MW = 2292	< 0.001 HS
	IQR	45.8 – 58.3	53 – 64		
Sex	Male	82 56.2 %	28 53.8 %	X^2 = 0.08	0.773 NS
	Female	64 43.8 %	24 46.2 %		
BMI (kg/m^2)	Median	27	26	MW = 3244	0.118 NS
	IQR	24 - 31	24 – 28		

MW: Mann-Whitney U. HS: p-value < 0.001 is considered highly significant.
X^2: Chi-square test. NS: p-value > 0.05 is considered non-significant.

This table shows, No statistically significant difference **(p-value > 0.05)** of sex and BMI as regard HBC Ig results and a Highly statistically significant difference **(p-value < 0.001)** between HBC Ig positive and HBC Ig negative patients as regard age.

Table (13): Comparison of CBC as regard HBC Ig result.

		HBC Ig			
		Negative (N = 146)	Positive (N = 52)	Stat. test	P-value
Hb (g/dl)	Median	12	10.5	MW = 2174	< 0.001 HS
	IQR	10.8 – 13.6	9.3 – 12.1		
HCT (%)	Median	40	36	MW = 1585	< 0.001 HS
	IQR	37.7 – 43	32 – 38		
WBCs (x10^3/ul)	Median	6	4.6	MW = 2980	0.021 S
	IQR	4.5 – 7.6	3.5 – 7.9		
PLTs (x10^3/ul)	Median	185	93.5	MW = 1954	< 0.001 HS
	IQR	124.8 – 225.3	75 – 144		

MW: Mann-Whitney U. HS: p-value < 0.001 is considered highly significant. S: p-value < 0.05 is considered significant.

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This table shows, Statistically significant difference **(p-value < 0.05)** between HBC Ig positive and HBC Ig negative patients as regard WBCs AND a Highly statistically significant difference **(p-value < 0.001)** between HBC Ig positive and HBC Ig negative patients as regard Hb, HCT & PLTs.

Table (14): Comparison of Kidney function tests as regard HBC Ig resultt

		HBC Ig			
		Negative (N = 146)	**Positive (N = 52)**	**Stat. test**	**P-value**
Na (mmol/L)	**Median**	**138**	**133**	**MW = 1248**	**< 0.001 HS**
	IQR	**135.7 – 139**	**129 – 134**		
K (mmol/L)	**Median**	**4**	**3.5**	**MW = 1384**	**< 0.001 HS**
	IQR	**3.8 – 4.3**	**3.3 – 3.8**		
Urea (mg/dl)	**Median**	**33**	**34**	**MW = 3324**	**0.183 NS**
	IQR	**26 – 38.3**	**28 – 44**		
Creat (mg/dl)	**Median**	**0.8**	**0.9**	**MW = 3354**	**0.212 NS**
	IQR	**0.7 – 1.09**	**0.7 – 1.25**		

MW: Mann-Whitney U. HS: p-value < 0.001 is considered highly significant. NS: p-value > 0.05 is considered non-significant.

This table shows**,** No statistically significant difference **(p-value > 0.05)** between HBC Ig positive and HBC Ig negative patients regarding urea & Creat.
Highly statistically significant difference **(p-value < 0.001)** between HBC Ig positive and HBC Ig negative patients as regard Na & K.

Table (15): Comparison of liver function tests as regard HBC Ig result.

		HBC Ig			
		Negative (N = 146)	**Positive (N = 52)**	**Stat. test**	**P-value**
PT (sec)	**Median**	14	16.1	**MW = 2032**	**< 0.001 HS**
	IQR	13 – 16	15 – 18		
INR	**Median**	1.2	1.4	**MW = 2062**	**< 0.001 HS**
	IQR	1.02 – 1.3	1.25 – 1.7		
T. Bil (mg/dl)	**Median**	1.2	2.4	**MW =**	**< 0.001 HS**

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	IQR	0.9 – 1.5	1.95 – 4.3	**1096**	
D. Bil (mg/dl)	**Median**	0.7	1.35	**MW =**	**< 0.001 HS**
	IQR	0.4 – 0.9	0.9 – 3.05	**1372**	
AST (U/L)	**Median**	53	78.5	**MW =**	**< 0.001 HS**
	IQR	39.3 – 69.5	52 – 94	**2460**	
ALT (U/L)	**Median**	58	87.5	**MW =**	**< 0.001 HS**
	IQR	36.8 – 78.8	53 – 140	**2460**	
ALP (U/L)	**Median**	69	86	**MW =**	**0.009 S**
	IQR	53.8 – 97.3	59 – 121	**2874**	
GGT (U/L)	**Median**	44	81.5	**MW =**	**0.007 S**
	IQR	32 – 87	39 – 154	**2844**	
ALB (g/dl)	**Median**	3.9	2.8	**MW =**	**< 0.001 HS**
	IQR	3.6 – 4.2	2.5 – 3.2	**640**	
TP (g/dl)	**Median**	6.5	5.2	**MW =**	**< 0.001 HS**
	IQR	6 – 6.8	4.5 – 5.6	**1128**	
AFP (ng/ml)	**Median**	6	123.5	**MW =**	**< 0.001 HS**
	IQR	4 – 8.25	8 – 322	**888**	

MW: Mann-Whitney U. HS: p-value < 0.001 is considered highly significant.

S: p-value < 0.05 is considered significant.

This table shows:

- Statistically significant difference **(p-value < 0.05)** between HBC Ig positive and HBC Ig negative patients as regard ALP & GGT.
- Highly statistically significant difference **(p-value < 0.001)** between HBC Ig positive and HBC Ig negative patients as regard PT, INR, bilirubin, AST, ALT, ALB, TP & AFP.

Table (16): Comparison of abdominal U/S as regard HBC Ig result.

		HBC Ig Negative (N = 146)		**Positive (N = 52)**		**Stat. test**	**P-value**
	Normal	68	46.6%	0	0%		
Liver status	**Bright**	44	30.1%	2	3.8%	**X^2 = 83.6**	**< 0.001 HS**
	Cirrhosis	34	23.3%	50	96.2%		
Splenomegaly	**No**	94	64.4%	4	7.7%	**X^2 = 49.3**	**< 0.001 HS**
	Yes	52	35.6%	48	92.3%		
	No	132	90.4%	4	7.7%		
Ascites	**Mild**	2	1.4%	12	23.1%	**X^2 = 123.8**	**< 0.001 HS**
	Moderate	10	6.8%	24	46.2%		
	Marked	2	1.4%	12	23.1%		
Focal lesion	**No**	140	95.9%	20	38.5%	**X^2 = 81.5**	**< 0.001 HS**

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	Yes	6	4.1%	32	61.5%		
PV diameter	**Median**	12.8		14.2		**MW = 1444**	**< 0.001 HS**
	IQR	11.5 - 14		14 – 15.1			

MW: Mann-Whitney U. HS: p-value < 0.001 is considered highly significant. X^2: chi-square test.

This table shows a highly statistically significant difference **(p-value < 0.001)** between HBC Ig positive and HBC Ig negative patients regarding abdominal U/S.

Table (17): Comparison of symptoms as regard HBC Ig result.

		HBC Ig Negative (N = 146)		**Positive (N = 52)**		**X^2**	**P-value**
Hematemesis	**No**	136	93.2%	26	50%	**47.9**	**< 0.001 HS**
	Yes	10	6.8%	26	50%		
Melena	**No**	132	90.4%	26	50%	**38.8**	**< 0.001 HS**
	Yes	14	9.6%	26	50%		
Hepatic encephalopathy	**No**	146	100%	36	69.2%	**48.9**	**< 0.001 HS**
	Yes	0	0%	16	30.8%		
Lower limb edema	**No**	128	87.7%	14	26.9%	**69.8**	**< 0.001 HS**
	Yes	18	12.3%	38	73.1%		
Abdominal pain	**No**	90	61.6%	10	19.2%	**27.5**	**< 0.001 HS**
	Yes	56	38.4%	42	80.8%		
Jaundice	**No**	128	87.7%	28	53.8%	**26.3**	**< 0.001 HS**
	Yes	18	12.3%	24	46.2%		

X^2: chi-square test. HS: p-value < 0.001 is considered highly significant.

This table shows a highly statistically significant difference **(p-value < 0.001)** between HBC Ig positive and HBC Ig negative patients regarding symptoms.

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Table (18): Comparison of comorbidities as regard HBC Ig result.

		HBC Ig Negative (N = 146)		Positive (N = 52)		X^2	P-value
Blood Transfusion	**No**	98	67.1%	12	23.1%	**30.1**	**< 0.001 HS**
	Yes	48	32.9%	40	76.9%		
Previous surgery	**No**	72	49.3%	22	42.3%	0.75	0.385 NS
	Yes	74	50.7%	30	57.7%		
DM	**No**	133	91.1%	28	53.8%	**35.01**	**< 0.001 HS**
	Yes	13	8.9%	24	46.2%		
HTN	**No**	104	71.2%	16	30.8%	**26.3**	**< 0.001 HS**
	Yes	42	28.8%	36	69.2%		

NS: p-value > 0.05 is considered non-significant.
X^2: chi-square test. HS: p-value < 0.001 is considered highly significant.

This table shows**,** Highly statistically significant difference **(p-value < 0.001)** between HBC Ig positive and HBC Ig negative patients as regard DM, blood transfusion & previous surgery, and no statistically significant difference **(p-value > 0.05)** between HBC Ig positive and HBC Ig negative patients as regard HTN.

4. Discussion

HBV disease is a significant medical issue, and around two billion individuals are currently infected despite effective vaccination. There are 350 million HBV carriers worldwide, and approximately one million die annually from HBV-related liver disease **[18].** Occult hepatitis B infection (OBI) is one of the most challenging topics in viral hepatitis **[19].**

OBI is defined by the presence of HBV DNA in the liver (with detectable or undetectable HBV DNA in the serum) in patients with serological markers of the previous infection (anti-HBc and anti-HBs positive) or in patients without serological markers (anti-HBc and anti-HBs negative) **[20].**

Occult HBV infection has frequently been identified in patients with chronic HCV infection; This occult infection may be associated with more severe liver damage and even the development of hepatocellular carcinoma (HCC**)[21].**

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Concerns have been raised regarding hepatitis due to HBV reactivation after successful clearance of HCV with pan oral DAAs in HBV/HCV coinfected patients. **[22]** this study represents the cross-sectional study of patients treated with pan oral DAAs.

The underlying mechanisms of HBV reactivation during pan oral DAA therapy for CHC remain speculative. Previously, several reports have documented that de novo HCV superinfection in the setting of chronic hepatitis B can result in HBeAg seroconversion and in some cases, clearance of HBsAg **[23].**

This shows that HCV can suppress HBV replication by host immune responses. Hence, clearance of HCV infection with effective anti-HCV therapy could then ameliorate immune control on HBV replication and result in HBV reactivation **[24].**

Life-threatening fulminant hepatitis due to HBV reactivation has also been reported in CHC patients with OBI treated with NS3/4A protease inhibitors–containing regimen **[25].**

Multiple studies have reported HBV reactivation in HCV/HBV coinfected patients treated with DAA. Still, the reactivation rate is unclear, and the clinical outcome can range from ALT flares to liver failure and even death **[26].**

HBV reactivation occurred more frequently, and the clinical outcome was more severe in patients treated with DAAs. This effect may be explained as follows: HBV viral replication can be suppressed in patients with HCV infection, and the rapid suppression of HCV viral load by DAA treatment may create a permissive environment for HBV replication, resulting in HBV reactivation **[27].**

Anti-HBc is the first antibody produced after HBV infection, and it is the primary distinguishable marker in the window period. Isolated anti-HBc refers to the presence of anti-HBc in the serum without HBs Ag or HBs Ab. Isolated anti-HBc may be due to resolved HBV infection. HBsAb had declined to an imperceptible level, testing during the window period, or chronic contamination, in which HBsAg cannot be identified because of protein change, makes it imperceptible by certain analytic measures **[28].**

In our study, HBc IG was mainly used to determine the presence of occult Hepatitis B infection in a large cross-sectional adult Patients who have received DAAs, and HBV PCR performed to patients who have anti-HBc positive Patients with occult HBV infection, who lack detectable HBsAg, may have an infection only indicated by anti-HBc and HBV-DNA **[28].**

In this study, we have investigated the prevalence of occult HBV infection in a population of HCV patients had treated by DAAs at different stages of the disease, from chronic hepatitis to liver cirrhosis. The study included 198 patients who have received the new treatment (DAAs) for HCV with negative viral markers for hepatitis B surface antigen in serum samples. We found 52 patients (26.3%) of

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198 patients had evidence of hepatitis B viral infection. Some studies on occult HBV in hepatitis C patients reported highly variable prevalence, from 0% to 52% **[29].** The prevalence of occult HBV infection varies depending on the hepatitis B risk factors **[30].**

Another reason for the heterogeneity of the results on the prevalence of occult hepatitis B is the significant variability in methodological approaches to its detection. A previous study was done by **(Fernandes et al. 2015[31]),** who found higher levels of biochemical parameters as well as a greater degree of liver fibrosis in HCV/occult HBV patients when compared with patients with HCV alone **[32]**. Moreover, several studies **(Miura,et al [7]**) emphasized the clinical impact of silent HBV in patients suffering from chronic liver disease as a result of HCV and reported that higher levels of disease severity were seen in the liver.

Also, we found there are elevated liver enzymes in patients with HBc IG positive. according to their levels of ALT, occult hepatitis B virus infection was significantly more frequent in patients with ALT flare than patients with normal or slightly high ALT

A relationship between the occult HBV and chronic HCV disease and high aminotransferases levels has been suggested by some studies **(Fukuda et al. [32])** found higher serum ALT levels in occult HBV-infected patients. Similarly**, Kannangai et al. [33]** reported that HBV genomes were detectable in all cases with ALT/AST flares found that 20% of chronic HCV-infected patients had occult HBV infection 66.7% of them had significant ALT elevation. The flare in liver enzymes may be due to the flare in HBV DNA replication, immune activation, and subsequent liver injury **[34]**

In agreement with most reports, our study found an association between the prevalence of OBI and flare of liver enzymes. A possible hypothesis for this finding is that HCV infection may block the circulating viral expression of HBV, but anti-HBc in the serum and HBV DNA in the hepatocytes may persist. Many healthcare exposures are associated with HBV, including residence, HCV infection, surgical history, blood donation, blood transfusion, and hemodialysis **[35].** In our study, a history of previous blood transfusion was observed in a significant number of cases. (24 patients out of 52 had OBI) have a history of blood transfusion. Also, 36 OBI patients have a history of previous surgery. However, our study agrees with However, and other studies reported that blood transfusion was an important risk factor for acquiring HBV infection **[36].**

Anti HBc positive are at increased risk for HCC in our study; we found that 32 patients of 52 who have anti-HBc positive (61.5%) but in a patient with anti-HBc negative HCC were six patients of 146 (4.1%)

In other studies, from Asia and Europe, the prevalence of OBI in patients with chronic HCV infection was 15–49% in those without HCC compared to 73% in those with HCC **[37].**

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5. Conclusion

In our study occult, hepatitis B was detected in a significant number of patients using HBc IgG, so check HBc IG status before initiating pan-oral DAAs therapy is essential, as occult HBV may contribute to chronic liver damage, development of HCC, and flare of liver enzyme and role of OBI on the outcome of DAAs treatment for HCV. OBI is a life-threatening public health problem worldwide. The detection of OBI is costly, especially for developing countries; therefore, many patients with OBI may remain undiagnosed. OBI can be controlled in high-risk groups, provided that the implementation of highly sensitive molecular means is used to detect HBV DNA as a preventive measure.

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