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Epidemiological study of Hepatitis C virus in Misan governorate

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Abstract
Hepatitis C virus (HCV) is a RNA virus classified as a febrile disease virus that infects liver specifically. The illness that caused by this virus can be asymptomatic and in long term of infection might lead to damage the liver and then potentially life threatening of the virus carriers. The main goal of this research is to HCV diagnossis in suspected patients in Misan province. Here, immunological assay, which is Enzyme Linked Immunosorbent Assay (ELISA) were utilized for HCV detection. The sera of the patients with different in ages and sex were obtained by collaboration with Central Health laboratory in Misan governorate to be use for these purposes. The findings revealed that the HCV infection is an existent in Misan province, as the ELISA test confirmed 36 (3.4%) positive cases and 1077 (96.6%) negative cases in 1115 specimens for 15 months from January 2023 to March 2024. In conclusion, this research has been demonstrated that the incidence rate of HCV in Misan is low and it potentially increased if any a lack of local health system and/or emerging a new virulent strain of the virus.

Keywords: HCV, ELISA, Hepatocyte, Misan province

1. Introduction

Hepatitis C virus (HCV) is a virus that refers to *Flaviviridea*, along with Dengue virus, West Nile virus and Yellow fever virus. These viruses are distributing globally and cause illness for both human and animals [1]. HCV belonging to the *Hepacivirus* genus, and there are six sub-species of HCV include 1a, 1b, 2, 3, 4, and 6 that distribute around the globe[2]. The HCV invades hepatocytes and in chronic infection might leads to liver cirrhosis and hepatocellular carcinoma [3]. The virion is enveloped, spherical, around 50 nm in diameter and has virus-encoded membrane proteins, which is E1 and E2 and capsid protein [4–7]. The genome is single stranded RNA, positive sense and composed from around 9600 nucleotides [8–10]. HCV genome contains one open reading frame that composed from around 300 amino acids and terminated by untranslated region (UTR) at 5' terminal, as the replication and translations starts from this region [11]. The nucleocapsid is globular, reaches around 30 nm and flanked by lipid envelope that the HCV acquires it from endoplasmic reticulum (ER) of target cells [12]. Additionally, there are three-structure protein (sp) and seven non-structure protein (nsp) with the genome, which have significant role in replication and assembly strategies of the virus [11]. HCV virus causes a cute and chronic infection and it depends on the immunity of the infected persons. In weak immune response patients, HCV infection develops to chronic and leads to steatosis [13–15] fibrosis [16,17] and cirrhosis [18,19] of liver. More over, these chronic lesions can lead to Hepatocellular carcinoma as the researches revealed that it the main cause of the death worldwide [20]. The reports declared that there is up to 180 millions persons were infected or carrier with HCV around the world most of them in United States, as those patients account 3% of total world population [21]. For instance, in Pakistan, there are around 19 millions persons were infected with HCV as they account around 10% of the total population of Pakistan [22,23]. In Iraq, many researches stated that many patients

were infected with HCV in Iraqi governorates including Kurdistan region [24–27]. The reports revealed that the HCV infection is high in Baghdad (Resafa region), Diwaniya and Sulaymaniyah governorates [24]. HCV can be detected by many methods, in Iraq, the ELISA technique is a widely used for detection HCV in patients in both research and health institutions [27].

2. Materials and Methods

2.1 Collection of blood specimens

The blood samples were obtained from patients attended Central Health laboratory in Misan province from January 2023 to March 2024. At this study, 1115 blood specimens (491 males and 441 females from January 2023 to December 2024, 93 males and 90 females from January 2024 to March 2024) were accumulated from in different ages. Total of 584 samples from males aged between 20 and 60 years and 531 samples from females aged between 20-60 years.

2.2 Elisa

In this research, Hepatitis C virus (HCV) was tested by ELISA apparatus (BIoTek) using Anti HCV ELISA 480 Test (HMG) kit according to manufacture's instructions. The procedure was done in isolated condition to avoid any expected contamination and the serums were prepared from obtained blood samples. At first, the reagents were reached at room temperature for 15 minutes. Then, three wells of each positive and negative control were prepared on each plate with one well as blank control. Secondly, 10 ml of sera, 100 ml positive control and 100 ml negative control were added into the wells. After, the wells were sealed, shacked gently and incubated at 37 °C for one hour. Next, the wells were washed five times for 20 seconds. Again, 50 ml of conjugates were added to the wells, and then incubated at 37 °C for 30 minutes. After that, the wells were washed five times by washing buffer for 20 seconds. Meanwhile, 50 ml of both substrate A and B were added to the wells. Eventually, 50 ml of stopping buffer was added to terminate the reactions. As results, the final reactions were red by ELISA reader (BIoTek).

3. Statistical analysis

The collected data were analysed statistically and presented by utilizing SPSS V 28.0 software.

3. Results:

At this study, ELISA technique was used for Hepatitis C virus (HCV) detection in Misan province. The results obtained from the preliminary analysis are set out in Table 3.1, 3.2, 3.3. In particular, as shown in Table 3.1, in 2023, 10 (2.0%) males were positive (infected) and 481 males (98.0%) were negative (non-infected) from total 491 males. In females, there are 2 (0.5%) females were positive and 439 (99.5%) females were negative from total 441 females. The statistical analysis (Chi-Square test), carried out by SPSS indicates that there are statistical significant in infection between males and females as p value < 0.05. In first quarter of 2024, as shown in Table 3.2, there are 9 males (9.7%) were positive and 481 (98.0%) males were negative from total 93 males. In females, there are 17 (18.9%) females were positive and 73 (81.1%) females were negative from total 90 females. The most surprising aspect of the data is that the incidence rate of HCV infections in first quarter of 2024 is greater than whole year of 2023 in both males and females. In total, there are 1115 tested samples from January 2023-March 2024, it is apparent from Table 3.3 that 19 (1.7%) males patients were infected with HCV, were 584 (96.7%) males were negative from total 584 tested samples. A part from this, the results also shows that there are 19 (3.6%) females were positive and 512 (96.4%) females were negative from total 531 females. What is more, the Chi-Square test indicating that there are non-statistical significant in infection between males and females as p value > 0.05 as shown in Table 3.2 and 3.3. These finding matches with the result of other studies carried out other Iraqi provinces, which reported the incidence rate of HCV is low [24,26,27].

Therefore, this research has demonstrated that HCV infection rate are low relatively in Misan province, as there are a few numbers of infected patients from up to a thousand of tested specimens and thus future researches on the present topic are recommended.

Table 3.1: Hepatitis C virus (HCV) incidence from January 2023 to December 2024 in Misan province.

Sex	Age (Year)	Variables	Cases		Total
			Positive	Negative	
Male	20-60	Count	10	481	491
		% within sex	2.0%	98.0%	100.0%
		% within infected	83.3%	52.3%	52.7%
		% of Total	1.1%	51.6%	52.7%
Female		Count	2	439	441
		% within sex	0.5%	99.5%	100.0%
		% within Infected	16.7%	47.7%	47.3%
		% of Total	0.2%	47.1%	47.3%
Total		Count	12	920	932
		% within Sex	1.3%	98.7%	100.0%
		% within Infected	100.0%	100.0%	100.0%
		% of Total	1.3%	98.7%	100.0%

*Chi-Square= 4.582, p value < 0.05.

Table 3.2: Hepatitis C virus (HCV) prevalence from January 2024 to March 2024 in Misan province.

Sex	Age (Year)	Variables	Cases		Total
			Positive	Negative	
Male	20-60	Count	9	84	93
		% within sex	9.7%	90.3%	100.0%
		% within infected	34.6%	53.5%	50.8%
		% of Total	4.9%	45.9%	50.8%
Female		Count	17	73	90
		% within sex	18.9%	81.1%	100.0%
		% within Infected	65.4%	46.5%	49.2%
		% of Total	9.3%	39.9%	49.2%
Total		Count	26	157	183
		% within Sex	14.2%	85.8%	100.0%
		% within Infected	100.0%	100.0%	100.0%
		% of Total	14.2%	85.8%	100.0%

*Chi-Square= 3.184, p value > 0.05.

Table 3.3: The total number of Hepatitis C virus (HCV) prevalence from January 2023 to March 2024 in Misan province.

Sex	Age (Year)	Variables	Cases		Total
			Positive	Negative	
Male	20-60	Count	19	565	584
		% within sex	3.3%	96.7%	100.0%
		% within infected	50.0%	52.5%	52.4%
		% of Total	1.7%	50.7%	52.4%
Female		Count	19	512	531
		% within sex	3.6%	96.4%	100.0%
		% within Infected	50.0%	47.5%	47.6%
		% of Total	1.7%	45.9%	47.6%
Total		Count	38	1077	1115
		% within Sex	3.4%	96.6%	100.0%
		% within Infected	100.0%	100.0%	100.0%
		% of Total	3.4%	96.6%	100.0%

*Chi-Square= 0.089, p value > 0.05.

4. Conclusions

The main purpose of the current research is for Hepatitis C virus (HCV) investigation and incidence rate evaluation in Misan city, Iraq. The study was set out to test blood specimens from suspected patients by collaboration with Central Health laboratory in Misan. The most significant finding to emerge from this research is that there is existence of HCV in Misan province. Moreover, the results of this investigation confirmed that the incidence rate of HCV is low, which is similar to the reports by other researches that carried out in other Iraqi governorates[24–27]. Taken together, it seems that the HCV is endemic virus in Misan and other Iraqi cities, therefore, the Iraqi health authorities should well prepared in order to control and eliminate the virus transmission. As there is no vaccine available for this virus, the most important way to prevent the HCV infection is to avoid contact with the virus and additional care should be utilized in health care setting and for the patients that they infected with HCV. The key strengths of this research are its used sensitive technique for virus detection and its acquire long duration to be completed and it is unique research on HCV carried out in Misan province. Further surveys and experimental investigations are recommended in future to determine virus distribution and for evaluation the incidence rate caused by HCV infections in Misan and other Iraqi governorates.

5. Conflict of interests

The authors confirmed that there is no competing of interests related to this research.

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