

HEPATITIS-B MARKERS IN SERA OF EGYPTIAN HEPATOMA PATIENTS

By

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دلالات الالتهاب الكبدي الوبائي (ب) في مصل مرضي سرطان الكبد المصريين

عبد الباسط الأعصر و نادية زخاري و صبري شعراوي و سلوى الدمرداش
و محمد حمزة و رفعت كامل و ميخائيل ميخائيل

شمل هذا البحث ٥٢ مريضاً مصابون بسرطان الكبد ، من بينهم ٤٦ مريض
مصابون بالنوع المعروف بإسم السرطان الكبدي الخلوي . والهدف من البحث هو إمكانية
وجود علاقة بين الإصابة بفيروس الالتهاب الكبدي الوبائي (ب) والإصابة بسرطان الكبد
في المصريين .

ولتحقيق هذا الهدف تم البحث عن دلالات التهاب الكبدي الوبائي (ب) وبالتحديد
HBsAg و HBsAb في مصل هؤلاء المرضى ومقارنتهم بالأشخاص الأصحاء .

وقد أسفرت الدراسة عن النتائج الآتية :

* ايجابية الإصابة بفيروس الالتهاب الكبدي الوبائي (ب) في مجموعة المرضى
بسرطان الكبد عنه في مجموعة الأصحاء حيث أن الكاشف عن الفيروس المسمى HBsAb
كان ايجابياً في نسبة ٨٠,٨٪ من كل حالات المرضى وبالمثل كانت نسبة الإيجابية في
الكاشف عن الفيروس المسمى HBcAb تبلغ ٨٤,٦٪ ، وأخيراً كانت نسبة الكاشف عن
الفيروس المسمى HBsAg ايجابياً بنسبة ٨٣,٥٪ . هذا بينما كانت هذه الكواشف الثلاثة
السابقة سلبية في جميع حالات مجموعة الأصحاء .

* نسبة الإصابة بفيروس الالتهاب الكبدي الوبائي (ب) أكثر في المرضى المصابون
بالسرطان الكبدي الخلوي عنه في باقي مجموعات سرطان الكبد الأولى الأخرى . وهذا
الفرق ذو دلالة احصائية .

* الدراسة الاحصائية أثبتت أن وجود الكاشف HBsAb في مصل المرضى لايعتمد
على وجود أو عدم وجود الكاشف HBcAb والعكس صحيح .

Key Words: Hepatoma, Hepatocellular carcinoma, Hepatitis B markers.

ABSTRACT

The present study was performed on 52 hepatoma patients; 46 of them were of the histopathological type known as hepatic cell carcinoma (HCC), aiming to investigate the possible role of hepatitis B viral (HBV) infection in the etiology of hepatoma; among Egyptians, with special reference to hepatocellular carcinoma. To fulfill this aim, the three hepatitis B markers namely hepatitis B surface antigen (HBsAg), hepatitis B surface antibody (HBsAb) and hepatitis B core antibody (HBcAb) were estimated in sera of patients and healthy controls. The present study revealed a striking correlation between clinical or subclinical history of HBV infection or even carrier rate, and hepatoma, since 94.2% of the hepatoma patients were positive for one or more of the hepatitis markers studied. The higher prevalence of HBV infection was observed among HCC patients (97%) than among other types of hepatoma patients (66%, $P = 0.03$). HBsAb and HBcAb were positive among 80.8% ($P < 0.01$) and 84.6% ($P < 0.01$) of hepatoma patients; respectively, whereas they were completely absent in sera of controls. On the other hand HBsAg was positive among 13.5% only of the hepatoma patients, revealing no significant changes when compared to controls ($P = 0.133$). Nevertheless, no significant correlation was observed between HBsAb positive patients and those who were positive for HBcAb ($P = 0.35$).

INTRODUCTION

Hepatocellular carcinoma (HCC) is the most common primary malignancy in the liver on a world wide basis. It represents about 83% of all primary malignant hepatic tumour[1]. HCC shows a remarkable geographical variability in world-wide incidence[2]. In portions of Africa and Asia, HCC is the most common malignant tumour, with incidence ranging from 34 per 10,000 men in Singapore to more than 100 per 10,000 men in Mozambique and Taiwan. It occurs less frequently in the United States and North America, with fewer than 10,000 new patients annually, accounting for less than 2% of all malignancies. A similar low incidence is found in Britain, Canada, Australia and South America.

The registry of the National Cancer Institute in Egypt[3] showed that primary liver cancer represented 18.14% of the digestive tract malignancies and 2.5% of total malignancies through out the years 1985 and 1989.

At the same time, the incidence of HCC was 3.7% of the total number of malignant cases received at the Pathology Department, Faculty of Medicine of Cairo University during the period of 1985-1987[4]. HCC occurs in a higher incidence in some subsets of human populations living at specific geographic areas around the world. These include black populations residing south of the Sahara, particularly in South and East Africa, in populations of South East Asia and the Western Pacific, in India, China, and in some other circumscribed areas.

These epidemiologic observation strongly suggest that environmental factors are involved in the etiology of HCC.

Evidence from human and animal data points towards a multicausal etiology, including dietary or environmental contamination with mycotoxin carcinogens, acting in concert with hepatitis B viral infection, and in some areas with malnutrition. Dietary factors that appear to influence susceptibility to HCC include fat, protein, amino acids, vitamin A, selenium and zinc[5].

The importance of chronic hepatitis B viral infection in the development of primary liver cancer has been established by epidemiological studies. Both retrospective and prospective epidemiologic studies have shown an etiological relationship between chronic hepatitis B viral infection and HCC[6].

The present work aimed to find out a relation between the history of hepatitis B viral infection and the incidence of hepatoma, with special reference to hepatocellular carcinoma (HCC).

MATERIALS AND METHODS

The present study was carried out on 52 hepatoma patients from the National Cancer Institute in Cairo (NCI) as well as private hospitals. Their ages ranged from 38 to 72 years. Serum was collected from the patient before receiving any chemotherapeutic, radiotherapeutic or surgical treatments. The control group was represented by fifteen normal volunteers from the medical and paramedical staff. Their ages ranged from 18 to 53 years.

The serum was subjected to the following investigations:

1. Detection of hepatitis B-surface antigen (HBsAg).
2. Detection of hepatitis B-surface antibody (HBsAb).
3. Detection of hepatitis B core antibody (HBcAb). All investigation were carried out using the kits provided by Sorin Biomedical, which was principally using the enzyme linked immunosorbant assay (ELISA) technique, using coated beads.

Statistical analysis were performed using the chi-square test[7].

RESULTS

1. Comparison of Hepatitis B Markers (HBsAg, HBsAb and HBcAb) in sera of patients and controls

Table(1) and Figure (1) showed significant differences in HBsAb and HBcAb incidence among hepatoma patients and controls; being positive in 80.8% ($P < 0.01$) and 84.6% ($P < 0.01$) respectively in sera of hepatoma patients. On the other hand no significant difference was observed between patients and controls, regarding the presence of HBsAg in their sera ($P = 0.133$). On the other hand, the sera of controls were completely free of either of the hepatitis B markers investigated.

Table 1
Hepatitis B Markers in Hepatoma Patients and Controls

| | Control | Hepatoma Patients | P-Value |
|-----------|-----------|-------------------|-----------|
| | n percent | n percent | |
| HBsAg +ve | 0 0 | 7 13.5 | P = 0.133 |
| HBsAg -ve | 15 100 | 45 86.5 | not Sig. |
| HBsAb +ve | 0 0 | 42 80.8 | <0.01 |
| HBsAb -ve | 15 100 | 10 19.2 | Sig. |
| HBcAb +ve | 0 0 | 44 84.6 | <0.01 |
| HBcAb -ve | 15 100 | 8 15.4 | Sig. |
| Total | 15 | 52 | |

n = number of cases

percent = results are represented as percentage taking total number of cases for each group as 100%.

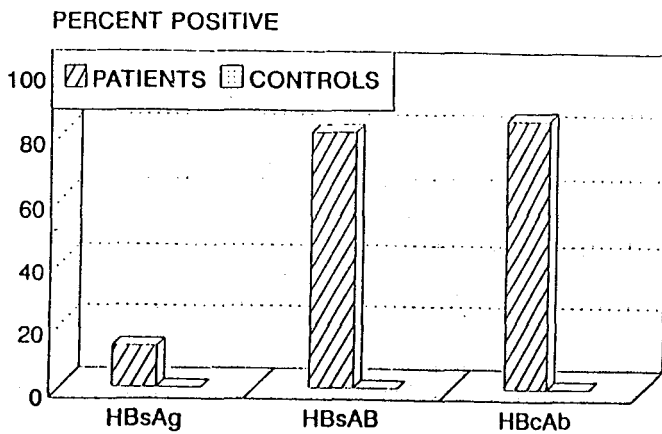


Fig. 1. Hepatitis B Markers in Hepatoma Patients and Control.

2. Hepatitis B Infection among patients with HCC and other types of hepatomas

A significant prevalence of hepatitis B viral infection or history of infection was observed among patients with HCC when compared to those suffering from other types of hepatomas ($P < 0.03$, Table 2 and Figure 2), since 97.8% of HCC patients had history of hepatitis B infection. On the other hand, only 66.7% of patients suffering other types of hepatoma were positive for hepatitis B infection.

Table 2
Hepatitis B Infection Among Patients with HCC and the Other Types of Hepatomas

| | Hepatitis B (+ve) | | Hepatitis B (-ve) | | Total | |
|---------|-------------------|---------|-------------------|---------|-------|---------|
| | n | percent | n | percent | n | percent |
| HCC | 45 | 97.8 | 1 | 2.2 | 46 | 100 |
| Others | 4 | 66.7 | 2 | 33.3 | 6 | 100 |
| Total | 49 | 94.2 | 3 | 5.8 | 52 | 100 |
| P-Value | <0.03 | | significant | | | |

Legends as table (1)

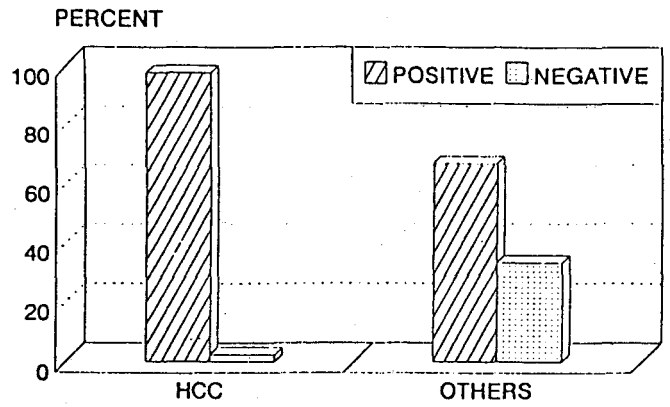


Fig. 2. Hepatitis B Infection Among Patients with HCC and Other Types of Hepatomas.

3. Comparison between HBsAb and HBcAb among hepatoma patients

There was no relation between HBsAb positive patients and those which were positive for HBcAb ($P = 0.035$, Table 3 and Figure 3). Also, 71.2% of the patients were positive for both markers and 5.8% of them were negative for both markers, whereas 23.1% of the patients were positive for one of the markers only and negative for the other.

Table 3
Comparison between HBsAB and HBcAb in serum of hepatoma patients

| Marker | HBcAB | | | | Total | |
|----------|--------------------------|---------|----------|---------|-------|---------|
| | Negative | | Positive | | No. | Percent |
| HBsAB | No. | Percent | No. | Percent | No. | Percent |
| Negative | 3 | 5.8% | 7 | 13.5% | 10 | 19.2% |
| Positive | 5 | 9.6% | 37 | 71.2% | 42 | 80.8% |
| Total | 8 | 15.4% | 44 | 84.6% | 52 | 100.0% |
| P value | = 0.35 = Not significant | | | | | |

Percentage is calculated taking the whole number of hepatoma patients (52) as 100 percent.

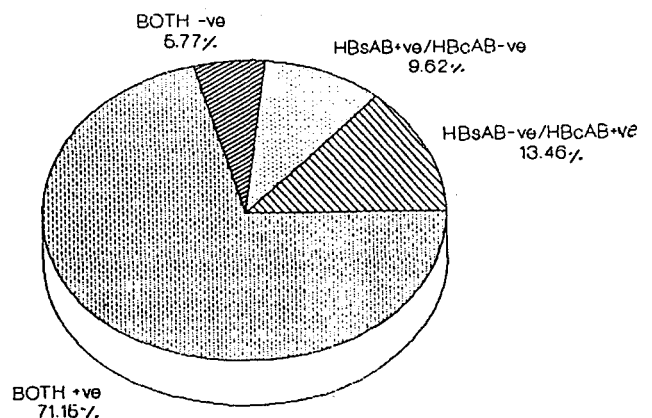


Fig. 3. Relationship Between HBsAB and HBcAB in Hepatoma Patients.

DISCUSSION

Infection with hepatitis B virus leads to the appearance of HBsAg in plasma during the incubation period, about 2-8 weeks before biochemical evidence of liver dysfunction of the onset of jaundice. HBsAg persist during the acute illness and is usually cleared during convalescence. Persistence for more than six months implies a carrier state[8]. Hepatitis B core antigen (HBcAg) is not detectable in a free form in the serum during HBV infection, but an antibody to HBcAg is found in serum at a period of 2-4 weeks after the appearance of the surface antigen and is always detectable during the early acute phase of illness. Core antibody of IgM class usually becomes undetectable within some months of the onset of complicated acute infection. IgG core antibody persists after recovery for many years and possibly for life. IgM core antibody, in the absence of HBsAg, indicates recent infection with HBV, but the finding of IgM core antibody in addition to HBsAg does not discriminate between acute and chronic infection with this virus. Antibody to the surface antigen is the last marker that appears late during convalescence, anti-HBs appears to confirm immunity[9].

The present study revealed a close association between primary hepatic tumour and HBV infection or history of infection. 13.5% of patients suffering different types of hepatomas were positive for HBsAg, 80.8% were positive for HBsAb and 84.6% of patients were positive for HBcAb. Regarding the HCC type of hepatoma, 97.8% were positive for hepatitis or history of HBV infection comparing both HBsAb and HBcAb revealed that 7 patients were positive for HBcAb and negative for HBsAb whereas 5 patients were positive for HBsAb and negative for HBcAb at the same time. There were also 3 patients negative for both HBsAb and HBcAb, whereas 37 patients were positive for both markers. This means that no relationship exists between the results of HBsAb in hepatoma patients. Thus the determination of a single marker is not enough but both markers must be done during the assay for hepatitis history among these patients.

In both high and low hepatocellular incidence areas, previous numerous studies have uniformly and conclusively shown a much higher frequency of HBV markers in patients with HCC than in controls, whether these controls were healthy blood-donors, villagers, office workers, hospital patients with other types of liver disease[2,10]. In high HCC incidence areas, HBsAg is commonly found in sera of patients with HCC with an incidence of 10 to 15 fold greater than control subjects. In low incidence areas HBsAg is found less commonly in patients with HCC, and the HBsAg carrier rate is lower in the control population. The relative risk to hepatoma, remain high as 15 to 20 folds among HBsAg positive individuals[11]. De-Bisceglie et al[12] reported that HCC is quite common in sub-Saharan Africa and the Far East, where most cases are associated with infection with the hepatitis B virus. Ohaki et al[13] reported that HCC is quite common in sub-Saharan Africa and the Far East, where most cases are associated with infection with the hepatitis B virus. Ohaki et al[13] examined 36 cases of HCC in Japanese adult patients for the presence of HBsAg by reverse passive haemagglutination technique. They found that 13 patients (36)% were HBsAg positive. Irie et al[14] examined 35 patients with HCC in Philippine

and they found that 13 patients out of 35 (37%) showed histological HBsAg positivity. Data from Hong Kong, Japan, Philippine and the United States indicated that between 40% and 45% of patients dying from B viral cirrhosis, were also suffering from HCC[1]. Sjoren et al[15] tested 124 cases of HCC in black American adult patients for the presence of immunoglobulin G antibody against HBcAg. They found that HBcAg was present in 85 out of 124 patients (68.8%).

In France, where there is a low prevalence of hepatitis, hepatitis markers were studied in the cirrhotic patients with HCC. The results were as follows, HBsAg was 0%, HBsAb was 36.4% and HBcAb was 36.4% Chung et al[16] reported that hepatitis B viral infection has an etiological role in cirrhotic patients who are negative for HBsAg, but positive for antibody to hepatitis B surface or core antigen and in these patients, persistent HBV replication may play a role in the pathogenesis of liver damage. Bassendine[10] suggested that the replication system of HBV becomes more and more defective during the sequence of hepatitis to cirrhosis and to carcinoma.

Besides the HBV infection as etiological factors for HCC, there was also hepatitis C viral (HCV) infection which is considered as an important etiological factor in HCC etiology[17]. Kaklamani et al[18] reported that there were interaction between HCV infection and HBV infection in the origin of HCC.

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