Study on epidemiology of brucellosis of human in some districts of Baghdad

Reem Z. Shinashal  Amina N. Al-Thwaini  Abdul Redha M. Al-Abbasi

Presidency of Mosul University  Genetic Engineering and biotechnology institute – university of Baghdad

Received 6/6/2005  Accepted 28/3/2005

الملخص

جُمعت نماذج الدراسة من 100 حالة مرضية في الإنسان شملت 100 نموذج دم و5 نماذج من السائل المخوي والجبل الشوكي ونموذج واحد من السائل الزليلي للمرضى أنفسهم الذين يشكون من أعراض الحمى المتموجة من مناطق في بغداد.

درست الحالة المرضية للمصابين بالحمى المتموجة وتبين أن جميعهم يشكون من أعراض المرض المعروفة فضلا عن ظهور أعراض أكثر حدة وخطورة ألا وهو أعراض neurobrucellosis.

وتبين أن الإصابة بالمرض تظهر في كلا الجنسين إلا أن نسبة إصابة الإناث أكثر من الذكور وهي 60% و 40% على التوالي، وإن الأعمار الأكثر تعرضاً للإصابة هي (6-48) سنة.

اجري فحصا الروزنماك والاسعاع المناعي غير المباشر على نماذج مصلى المرضى و كانت نسبة النتائج الموجبة للفحص الأول 100% و الفحص الثاني 70.2% على التوالي.

وتأثرت نتائج العزل الجرثومي من هذه الحالات بطريقئ وسط كاستنديا الثاني التطوري إلى عزل 23 عزلة جرثومية و هي 18 عزلة نماذج الدم و 4 عزلات لنموذج السائل المخوي و السائل الشوكي و عزلة واحدة لنموذج السائل الزليلي و بتحديد الأنواع والأمراض الحيوية وجد.
Abstract

Occurrence of Brucella was monitored in 100 samples of blood, 5 samples of cerebrospinal fluid and 1 sample of synovial fluid from same patients suffering from sing's of undulant fever in same districts of Baghdad and these cases of undulant fever were studied and all of them were suffering from the diagnostic sings of the disease beside the presence of dangerous sing's of neurobrucellosis. The incidence of disease present in both sexes and the percentage was higher in female than male 60%, 40% respectively and the highest incidence of disease occurred in the range of (6-48) year. The rose bengal test and the indirect Fluorescent antibody test were done on patients serum and positive results appeared as 100% and 70.2% respectively. Bacterial isolation by Castaneda bi - Phasic medium revealed 23 bacterial isolates, 18 isolates, 4 isolates, 1 isolate from blood, cerebrospinal fluid, synovial fluid respectively. The biotypes were determined as showed that 8 isolates were of B. abortus biotype 3rd., 4th. and 15 isolates of B. melitensis of the second and third biotypes. The sensitivity test for the isolates were done by using different antibiotics.

Introduction

Brucellosis is a classical zoonosis (1), there is a over whelming a wareness of the disease in Iraq and neighboring counties where consumption of raw milk and unpasturized milk products are still widely used (Al-Shaarabf and Yahyo, 2, Siddiqui, 3, Best, 4).

B. melitensis is the infective agent of sheep and goats, is widely implicated as a cause of brucellosis in man more than B. abortus which is the infective agent of cattles. (5, 6, 7).

Brucella infected human and cause of undulant fever, clinically the disease might run acute coarse with fever, rigors, sweating, headache with additional signs of bones, joints, nervous systems, genital system and other system (8, 9). The attention of brucellosis in last years specially from WHO and FAO where as the disease attented from many of reasheres, because of infection distribute among animals of over the world and this range of the infections help the disease transmitted to human and lead to human healthy problem the animal source (10, 11, 12).
Materials and Methods

1. Collection of the sample: 100 samples of blood, 5 samples of cerebrospinal fluid from the same patients and 1 sample of synovial fluid were obtained from November 1997 to December 1998 from many hospital and clinics such as Abo-ghrab, medical clinic in Amaria, Baghdad Al-jadeda and Al-Huda lab. in mansour, these samples taken from patients suffering from fever, headache, arthritis. The samples compose of:

A-The Samples of blood:

100 samples from the patients by pull 10 cc of blood by sterile syringe from brachial vein cleaning the place by alcohol, injection 5 cc from blood in castaneda-bi-phasic medium* and abtained of the serum from blood samples in order to rose bengal test and indirect fluorescent antibody test (13, 14),

*This medium is prepared by method of (Castaneda, 15).

B-The samples of cerebrospinal fluid and synovial fluid.

5 samples of cerebrospinal fluid and 1 sample of synovial fluid from the same patients of undulant fever that entering the Ibn-Kateeb hospital and injected in castaneda -bi- phasic medium.

2. The bacterial isolation and diagnosis:

Samples processing techniques and currently recommended procedures for identification of brucella species and biotypes were mentioned elsewhere (16, 17, 18).

The sensitivity test of antibiotic depended upon on (Kirby, et al, 19) to sensitivity test of the antibiotic prepare in market and to treat human and animals.

Results

All patients were suffering from fever, headache, backache, joints, otherwise male were suffering from orchitis beside that appearing some of clinical signs that dangerous upon life of patients were neurobrucellosis (encephalitis, meningitis, brain abscess) that lead to death of two women.

The study showed that the disease is infected both sexes because of brucella is most commonly transmitted to human by consumption of unpasteurized dairy products orginated from infected animals.

The number of female was 60 and of male was 40 making female / male ratio of (1.5:1) figure 1.
Figure (1) Distribution of patients according to age & sex

Rose bengal test and indirect fluorescent antibody test:

The results of this study showed 100 samples of sera by range 100% positive results to R.B.T. was 3 degree of agglutination, number of patients in each of agglutination was 19, 25, 56 respectively where as the results of IFAT showed 74 samples from sera of patients were 52 samples by range 70.2% was positive results and 22 samples by range 29.7% was negative results a titre of positive serum was: 1:16 of 23 patients, 1 : 32 of 18 patients, 1 : 32 of 11 patients, Table 1
Table (1) The relationship between results of blood Culture and results of RBT, IFAT

<table>
<thead>
<tr>
<th>Blood culture</th>
<th>* Rose bengal test</th>
<th>** Indirect fluorescent antibody test</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>+</td>
<td>++</td>
</tr>
<tr>
<td>Positive results</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>Negative results</td>
<td>17</td>
<td>22</td>
</tr>
<tr>
<td>Total</td>
<td>19</td>
<td>25</td>
</tr>
</tbody>
</table>

* measured of titre upon central health Lab./ Baghdad
** + : 1 : 16
++ : 1 : 32
+++ : 1 : 32

Bacterial isolation and diagnosis:

The number of isolates from blood was 18 with a range of 18% from species B.abortus biotype 3rd, 4th, B. melitensis biotype 2nd, 3rd beside that one isolate from synovial fluid species B.melitensis biotype 2nd.

The results of this study showed isolated of brucella from cerebrospinal fluid was 4 isolates species B.abortus biotype 4th and B. melitensis biotype 3rd.

The total of isolates from blood, cerebrospinal fluid, synovial fluid were 23 isolates, 8 isolates from B.abortus by range 34.7%, 15 isolates from B.melitensis by range 65.2%, Table 2.
**Table (2) characterization of brucella isolates**

<table>
<thead>
<tr>
<th>Sample</th>
<th>No. of iso.</th>
<th>Species</th>
<th>Need of CO₂</th>
<th>Product of H₂S</th>
<th>Urea (hours)</th>
<th>Growth in dye (a)</th>
<th>Agglutination (b)</th>
<th>Phages</th>
<th>Biotype</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood 1</td>
<td>1</td>
<td>Abortus</td>
<td>+</td>
<td>+</td>
<td>(1-2)+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>Blood 2</td>
<td>2</td>
<td>Abortus</td>
<td>+</td>
<td>+</td>
<td>(1-2)+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>Blood 3</td>
<td>3</td>
<td>Abortus</td>
<td>+</td>
<td>+</td>
<td>(1-2)+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>Blood 4</td>
<td>4</td>
<td>Abortus</td>
<td>+</td>
<td>+</td>
<td>(1-2)+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Blood 5</td>
<td>5</td>
<td>Abortus</td>
<td>+</td>
<td>+</td>
<td>(1-2)+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Blood 6</td>
<td>6</td>
<td>Abortus</td>
<td>+</td>
<td>+</td>
<td>(1-2)+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Blood 7</td>
<td>7</td>
<td>Abortus</td>
<td>+</td>
<td>+</td>
<td>(1-2)+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Blood 8</td>
<td>8</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V(d)</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Blood 9</td>
<td>9</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Blood 10</td>
<td>10</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Blood 11</td>
<td>11</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Blood 12</td>
<td>12</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Blood 13</td>
<td>13</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Blood 14</td>
<td>14</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Blood 15</td>
<td>15</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Blood 16</td>
<td>16</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Blood 17</td>
<td>17</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Blood 18</td>
<td>18</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Cere. 1</td>
<td>1</td>
<td>Abortus</td>
<td>+</td>
<td>+</td>
<td>(1-2)+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Cere. 2</td>
<td>2</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Cere. 3</td>
<td>3</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Cere. 4</td>
<td>4</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Syno. 1</td>
<td>1</td>
<td>Melitensis</td>
<td>-</td>
<td>-</td>
<td>V</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Total 23</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

+ : positive results  
- : negative results  
(a) Conc. of dye (20) mg/ cc (1:50,000)  
(b) A: monospecific antibody to B. abortus  
M: monospecific antibody to B. melitensis.  
c: routine test dilution  
d: variable
The sensitivity test of antibiotics

The results of this study showed that brucella was sensitive 100% to those Streptomycin, Tetracycline, Gentamicine, Rifampicin, Kanamycin, Trimethoprin and mixed Trimethoprin with Sulfamethoxazole and to a less degree to Doxycycline-hydrochloride and Cephalexin and least sensitive to Erthromycin and Amoxycillin.

Discussion

The milk is considered to be a rich nutrient for human beside that it is the main factor for transmittance of the causative agent of infection to human specially brucella because of brucella is localized in udder and lead to infection and occurred of disease by consumption of unpasteurized dairy products (9).

Brucellosis is named great imitator (20) because clinical signs of this disease is similar the clinical signs of other disease e.g. Influenza, the patients suffer from headache, backache, orchitis, fever, orchitis in male because the testis contain erythritol sugar and considered a rich nutrient for brucella, death two of patients (women) of neurobrucellosis and cause meningitis and encephalitis. (Fathi, 21) said 14 cases of neurobrucellosis in hospital of Baghdad and Mosul from period November 1996 to March 1998.

The study was showed that the infection appeared of ages (6-48) year by percentage (24.40%). The results of this study are in accordance with (Araj, 22) that showed the percentage of infection is high in female than in male by range (1.5:1), therefore the results of the study showed that the infection appears in both sexes male and female.

The results of this study showed the range of R.B.T was 100%, These results differ from (Mohammad, Nooria and Al-Nasiry, 23) and isolates brucella from 18 samples from 100 sera samples table 1, these results belong to blood samples taken from patient in period brucella transmitted from blood to organs and lymph nodes or the patients taken antibiotic before admitted to lab.

The range of positive results to IFAT was 70.2 %, negative results was 29.7%, study of relationship between results of this test, results of blood culture showed that brucella isolates from 14 blood samples from the origin 52 positive to test beside that brucella isolated from 4 blood sample from the original 22 negative to test, these results give clear idea upon distribution of disease but prefer the tests beside that of bacterial isolation, the result differ from (Tawfiq, 24).

Brucella is a fastidious microorganism and difficult for isolation from human cases however it's cultivation is still fairly possible, early in infection and persist in fluid, tissues, organs body e.g Lymph node, brain because of the patient take of antibiotic before admitted to lab. therefore prefer the isolation of brucella in 2nd days from increase of temperature (acute stage) specially bactemia but the isolation was decrease in chromic stage (25).

The Number of isolates from blood is 18 by with a range of 18% species B.abortus biotype 3rd, 4th B.melitensis biotype 2nd, 3rd, first isolate from synovial fluid species B.melitensis 2nd.
This study showed that B. Melitensis much distributed from B. abortus the results of this study in accordance with (Young, 1), and this study showed that isolation of brucella from cerebrospinal fluid was isolate of 4 isolates species B. abortus biotype 4th, B. melitensis 3rd, theses results are accordance with, (Fathi, 21).

The total of isolates from blood, cerebrospinal fluid, synovial fluid was 23 isolates, 8 isolates from B. abortus by range 34.7%, 15 isolates from B. melitensis by range 65.2%.

The isolates were 100% sensitive to Streptomycin, Tetracycline, Gentamicine, Rifampicin, Kanamycin, Trimethoprim and mixed Trimethoprim with Sulfamethoxazole where as less sensitive to other antibiotic otherwise you take the attenuated points for treat the cases before that go make the sensitivity test antibiotics.

References