

Evaluating the prevalence of specific Treponemal antibodies in Iraqi blood d

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### Abstract

Detection of specific antibodies to *Treponema pallidum* in serum is correlated with Syphilis. The study aimed to know the seroprevalence of Specific treponemal antibodies among Iraqi blood donors in relation to age and sex. This study was conducted on 8,147 subjects attending National Center for Blood Transfusion/Baghdad for donation of blood during January 2009 to May 2010. A total of 8,147 blood donors were screened to detect the presence of specific antibodies against *Treponema pallidum* by *Treponema pallidum* Haemagglutination Test (TPHA), according to standard test kit protocol, initially reactive samples were reconfirmed by repeat testing and repeatedly reactive samples were considered seropositive for Syphilis. The results of this study showed that seroprevalence of specific antibodies to *Treponema pallidum* among blood donors were found 1.75%. Moreover, the seroprevalence in males was 1.53% and in females was 0.22% .Furthermore, this study showed increase in seroprevalence was found among persons within second to third decade. The study revealed considerably higher Specific treponemal antibodies among the male donors than Female donors. In conclusion, our results indicate that syphilis is found among healthy donors in Iraq and that there is need to introduce *Treponema pallidum* Haemagglutination Test for the screening of donated blood for specific treponemal antibody in all blood transfusion centers in Iraqi governorates.

**Key words:** Specific treponemal antibodies, Syphilis, Blood donors, Seroprevalence.

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### الخلاصة

أن التحري المصلي عن الأجسام المضادة الخاصة ضد اللولبية الشاحبة يكون مرتبط بمرض الزهري . هدفتُ الدراسة لمعرفة الانتشار المصلي للأجسام المضادة الخاصة ضد اللولبية الشاحبة في العراقيين المتبرعين للدم مع الأخذ بالنظر عامل العمر والجنس. أجريت هذه الدراسة على ٨١٤٧ شخص من المتبرعين للدم في المركز الوطني لنقل الدم في بغداد للفترة الممتدة من كانون الثاني ٢٠٠٩ إلى أيار ٢٠١٠. ما مجموعه ٨,١٤٧ متبرع بالدم عُرضوا لاكتشاف التحري عن الأجسام المضادة ضد اللولبية الشاحبة باستخدام اختبار التلازن الدموي للولبية الشاحبة . طبقاً لنظام عُدّة الاختبار القياسية ، فإن العينات التي تعطي تفاعل أولي يجب إعادة الاختبار عليها والعينات التي تعطي تفاعل ثاني تعتبر مصلية موجبة بالنسبة لمرض الزهري. أظهرت نتائج الدراسة بأن الانتشار المصلي للأجسام المضادة الخاصة ضد اللولبية الشاحبة بين المتبرعين بالدم كان (١.٧٥%) . أضافه إلى ذلك كان الانتشار المصلي في الذكور حوالي (١.٥٣%) و في الإناث كان (٠.٢٢%) . علاوة على ذلك، أظهرت هذه الدراسة زيادة في الانتشار المصلي في الأشخاص ضمن العقد الثاني والثالث من العمر. كُشفتُ الدراسة بأن الأجسام المضادة الخاصة ضد اللولبية الشاحبة أعلى بين المتبرعين الذكور من المتبرعين الإناث . نتيجة لذلك، نتائجا تُشيرُ بأن مرض الزهري موجود بين المتبرعين الأصحاء في العراق وبأن هناك حاجة لإدخال اختبار التلازن الدموي للولبية الشاحبة لفحص دم المتبرعين للأجسام المضادة الخاصة ضد اللولبية الشاحبة في كل مراكز نقل الدم في المحافظات العراقية

### Introduction

Blood transfusion has life-saving benefits, but also carries risks. Currently, prevention of transfusion-associated infection depends upon proper, pre-donation selection of donors, followed by serologic testing for infectious pathogens, including HBV, HCV , HIV and Treponema pallidum (1). Treponema pallidum is a species of spirochaete bacterium with subspecies that cause treponemal diseases such as syphilis, bejel, pinta and yaws. It is not seen on a Gram stained smear because the organism is too thin. T. pallidum is a motile spirochete that is generally acquired by close sexual contact, entering the host via breaches in squamous or columnar epithelium (2). Syphilis is a complex disease which is normally

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sexually transmitted, the causative organism, *Treponema pallidum* cannot be grown on conventional laboratory culture media or in the conventional tissue culture, Infection is normally diagnosed by detecting antibodies specific for *Treponema pallidum* in the patient's serum or cerebral spinal fluid (CSF), antibody become detectable at about 3-4 weeks following exposure, and remain at detectable levels for long periods after treatment (3).

The treponemal test: *Treponema pallidum* Haemagglutination Test (TPHA) have good sensitivity at all stages of disease and indicate the evidence of Syphilis, TPHA is a specific, sensitive passive haemagglutination test for the detection of antibodies to *Treponema pallidum* in serum or CSF (4). Syphilis is major public health problem world wide, the world health organization (WHO) estimates that 12 million new cases of syphilis occur each year and the greatest number of cases was estimated to have occurred in Asia (5). Syphilis can be transmitted by sexual contact, transplacental transfer and through indirect routes: contaminated objects, tattoos and blood transfusions (6, 10). One of the significant routes for transmission of the infection is through blood transfusion, the prevalence of antibodies to syphilis in Asian and African blood donors are relatively mild to moderate (7). In India, the seroprevalence of antibody to syphilis among blood donors was (1.0%). A similar prevalence rate (1.2%) of syphilis antibody has recently been reported in blood donors from Dar -Salaam, Tanzania (8,9). The literature also notes that syphilis can occur in blood donors (11, 12, and 13). Even so, there is very little information on the prevalence rate of Specific treponemal antibodies among blood donors in Iraq. This study is expected to be useful to evaluating the prevalence of specific Treponemal antibodies in Iraqi blood donor and would generate the data regarding sero-positivity of Specific treponemal antibodies in healthy looking general population.

### Subject, Materials and Methods

#### Subject group

A total of (8,147) subjects attending National Center for Blood Transfusion/Baghdad for donation of blood during January 2009 to May 2010. Aged from 18-57 years with male to female ratio of 5:1. Blood donors undergo a clinical screening, which involves a questionnaire and a routine medical examination and only those found to be

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Healthy individuals are used as donors, Donated blood is routinely screened For HIV, HBs Ag and HCV .In this study, an additional blood sample was also taken from blood donors for detect specific treponemal antibody.

### Sample collection

From each individual included in this study, 5-10 ml of blood was drawn by vein puncture using disposable syringes. The blood was placed in plastic disposable tubes, it was left to stand at room temperature (20-25°C) to allow it to clot, then the sera was separated by centrifugation for 5 minutes, and divided into aliquots (250 µl) and stored at -20°C till examination. Each aliquot of the serum was used once to avoid thawing and freezing. All sera and reagents were allowed to stand at room temperature before use in the test.

### Methods

#### Detection of specific Treponemal antibody by TPHA

##### Principle of the test

The TPHA comprises Treponema pallidum sensitized formalized tanned fowl erythrocytes; unsensitized formalized tanned fowl erythrocytes, diluents and control sera. When diluted positive samples are mixed with sensitized erythrocytes, antibody to the sensitizing antigen causes agglutination of the cells .The cells form a characteristic pattern of cells in the bottom of a microtitration plate well. In the absence of antibodies, they form a compact button in the well. This test has been calibrated to WHO reference serum for serodiagnostic for Treponemal infectious (4, 10). The detailed procedure was carried out as has been suggested in the leaflet supplied with the test kit (Omega, UK).

### Results and Discussion

At the conclusion of the study, a total of 8147 Iraqi blood donors, aged from 18-57 years were found to have been screened for syphilis, there were the number of males was 6799 and females was 1348 with male to female ratio of 5:1 (table 1) and majority of the donors were in the age group between 18 to 37years i.e. second to third decade of life (Table 2).

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Table 1: Gender distribution of Donors

| Gender | Frequency | %     | Ratio |
|--------|-----------|-------|-------|
| Male   | 6799      | 83.5  | 5     |
| Female | 1348      | 16.5  | 1     |
| Total  | 8147      | 100.0 | ----  |

Table 2 :-Distribution of donors according to gender within different age groups.

| Age (years) | Male (No.) | %     | Female(No.) | %     |
|-------------|------------|-------|-------------|-------|
| 18-27       | 1860       | 27.35 | 49          | 3.63  |
| 28-37       | 3650       | 53.68 | 741         | 54.98 |
| 38-47       | 830        | 12.2  | 381         | 28.26 |
| 48-57       | 459        | 6.75  | 177         | 13.13 |
| total       | 6799       | 100.0 | 1348        | 100.0 |

However, there is scanty information on the occurrence of Specific treponemal antibodies among blood donors in Iraq with which to compare. From this study, the age range of blood donors was 18 to 57 years. This is similar to that in the study of Khan et al (14) in Peshawar, Pakistan who found that their blood donors were in the age range of 18 to 60 years. It is also similar to the findings of Muktar et al (15) in Zaria, North-western, Nigeria in which their donors had a mean age of 33 years even though their age ranged from 19 to 52 years. Additionally, the donors in Jos-North central Nigeria were in the age range 21 to 50 years according to Egah et al (16). Most of the blood donors in this study were males, 83.5% (table 1). This is similar to the 85% in the study of Egah et al (16), moreover, Muktar et al (15) found that 98% of their donors were males, while Nwokediuko et al (17) in their study in Enugu, South-eastern, Nigeria found that 91.8% of their donors were males. However all the donors were males in the study of Elfaki et al (12) among Sudanese donors and the study of Khan et al (14). Furthermore, The results of this study showed that seroprevalence of specific antibodies to *Treponema pallidum* among blood donors was found 1.75%. besides, the

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seroprevalence in males was 1.53% and in females was 0.22% , as well as ,the majority of the infected donors are in the sexually active age groups, 18-27 and 28-37 years (Table 3 , 4).

**Table 3: Distribution of specific antibody to *Treponema pallidum* according to male within different age groups**

| Age (years) | No. positive / tested | % Positive |
|-------------|-----------------------|------------|
| 18-27       | 23/ 1860              | 1.27       |
| 28-37       | 69/ 3650              | 1.9        |
| 38-47       | 8/ 830                | 0.97       |
| 48-57       | 4/ 459                | 0.78       |
| total       | 104 / 6799            | 1.53       |

**Table 4: Distribution of specific antibody to *Treponema pallidum* according to female within different age groups**

| Age (years) | No. positive/ tested | % Positive |
|-------------|----------------------|------------|
| 18-27       | 0/49                 | ----       |
| 28-37       | 2/741                | 0.27       |
| 38-47       | 1/381                | 0.26       |
| 48-57       | 0/177                | -----      |
| total       | 3/1348               | 0.22       |

From this study, a specific treponemal antibody to syphilis among blood donors was 1.75% using TPHA. This figure is similar to the 1.72% found by Abdalla et al (18) in their study among Kenyan donors. It is also similar to the 1.71% found by Fiekumo et al (19) in Osogbo, South-western, Nigeria. It is however higher than the 0.85% found by Gupta et al (20) in Indian donors .The finding of 1.75% of syphilis among blood donors in this study was lower than the 3.6% found by Chikwem et al (21) in Maiduguri, North-eastern Nigeria ; the 7.5% found by Adjei et al (22) in Ghanaian donors and the 15.0% found by Elfaki et al (12) among Sudanese donors. The wide differences in the specific treponemal antibodies rate among the blood donors in the different regions may be due to the differences in geographical

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locations, age range of blood donors, sample sizes, the period of time the studies were carried out, and the different socio-cultural practices such as sexual behavior, marriage practices etc which take place in these regions. The implication of syphilis in voluntary blood donors is the risk of transmission of this infection to recipients of blood and blood products. This can contribute to the ever-widening pool of infection in the wider population. Syphilis has also acquired a new potential for morbidity and mortality through association with increased risk for HIV infection (10) thus making safe blood more difficult to get. We recommend the screening of all prospective blood donors for all transfusion transmissible infections. Blood that is positive for syphilis should be discarded, and the affected donor treated appropriately. A strict selection criteria for blood donors to exclude those with multiple sexual partners, and that blood transfusion should be restricted.

In conclusion, Even though the seroprevalence of specific Treponemal antibodies to syphilis infection is relatively low among voluntary blood donors in Iraq, safe blood will still be more difficult to get.

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