

A Comprehensive Survey for Toxoplasmosis for the period (1991-2007) and the Using ELISA for Diagnosis of Toxoplasmosis in Pregnant Women in Nineveh Governorate

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Abstract

The current study deals with two major aspects: the first is analysing statistical information documented at Health Care Department in Mosul from 1991 up to 2007. The second is evaluating serological test immunoglobuline (IgM) by (ELISA) test in order to identify toxoplasmosis. The result of the study showed the highest level of parasite toxoplasma *gondii* infection was through the years 2000, 2001, and 2002, with an average of infection 76.42, 62.67, 56.67 respectively, While the results of (ELISA) test indicated that (14) cases were positive out of (242) cases, and its ratio was 5.78%. As concern age group the highest range of infection is between (15-20) years old, where the percentage reached at 32% and 18% for the age between (21-25) years old, 12% for the ages between (26-30) years old, 10% for the ages between (31-35) years old and 8% for the ages between (36-40) years old, then it is 20% for the ages between (41-45) years old.

Introduction

The protozoa parasitic *Toxoplasma gondii* is an obligate intracellular parasite in the phylum Apicomplexa. Its life cycle can be completed only in cats and other felids, which are the definitive hosts. However, *T. gondii* also infects a wide variety of intermediate hosts, including humans, land and sea mammals like sea otters, and various bird species [1]. This parasite is the only known member species of the genus *Toxoplasma* [2]. Many people carry the parasite in their bodies but may not know because they have no symptoms. Acute stage toxoplasmosis infection can be a symptomatic, but often gives flu-like symptoms with early acute stage, and flu-like can become fetal in very rare cases [3].

Nicolle and Manceaux first described by their observation the parasite existence in blood, spleen and liver of North Africa rodent (*Ctenodactylus gondii* from which parasite was isolated in 1909) [2].

Pregnant women can pass the infection to their unborn baby if they get infected for the first time during or just before pregnancy, causing abortion or congenital infection leading to blindness, mental retardation and hydrocephaly [4]. If infection is acquired during pregnancy, the infection can spread to the fetus, the probability of its occurrence the occurring increases with the trimester of pregnancy, 17% in the first, 24% in the second, 62% in the third [5]. Also toxoplasmosis is one of the main reasons of abortion in sheep and goats in many countries, including Australia and United States [6].

Members of cat family (Felidae) are the only known definitive host for the sexual stage of *T. gondii* and thus the main reservoirs of infection is acquired by ingestion of viable by cat that contaminate the environment [7].

Cats are important in spreading toxoplasmosis, because they can shed the parasite in their feces; also cat become infected with *T. gondii* by eating infected mice and it is the only primary host at *T. gondii* [7].

The life cycle of *T. gondii* has two phases the sexual part of the life cycle takes place only in member of Felidae family (domestic and wild cats) which is the primary and definitive host for the sexual stage *T. gondii*. The asexual

part of life cycle takes places in any warm-blooded animal including felines, mice, birds, domestic livestock and humans serving as potential intermediate hosts. In these intermediate hosts, the parasite invades cells, forming intracellular and so-called parasitophorous vacuoles containing bradyzoites, (encysted bradyzoites) which grows slowly and subsists within the host throughout its life, vacuoles from tissue cysts mainly within the muscles and the brain.

Since they are within cells, the host immune system does not detect these cyst. Within these vacuoles of *T. gondii* propagates by a series of binary fission until the infected cell eventually bursts and tachyzoites are released. Tachyzoites are the motile asexually reproduced from of the parasite, unlike the bradyzoites, the free tachyzoites are usually efficiently cleared by the hosts' immune response, tissue cysts are ingested by cat through feeding on an infected mouse, the cysts survive passage through the stomach of the cat, and the parasites infect epithelial cells of the small intestine where they undergo sexual reproduction and oocyst formation. Oocysts are shed with the feces. Animals and humans that ingest oocyst by eating unwashed vegetable or tissue cysts or improperly cooked meat get infected. The parasite enters macrophages in the intestinal lining and distributed via blood stream through the body.

Oocysts can survive several months and remarkably resist disinfectants, freezing and drying but are killed by heating to 70°C⁰ for 10 minutes or -14 min. [8]

In Europe, congenital toxoplasmosis affects between 1 and 10 in 10000 new born babies, of whom 1% to 2% develop learning difficulties or die and 4% develop choroidal lesion leading to permanent unilateral impairment vision.

In United Kingdom 10% infection in pregnant women and Norway to around 55% [9]

In various places through the world, it has been shown that up to 95% of some population have been infected with Toxoplasmosis and is often highest in areas of world that have hot humid climates and lower altitudes. [10]

Toxoplasmosis is not passed from person to person, except in instances of mother to child (congenital) transmission and blood transfusing or organ transplantation. People typically become infected by three principal routes of transmission:

1. mother – to – child (congenital).
2. Animal – to – human (Zoonotic).
3. Food borne.
4. Rare instances.[11,12,13,14].

Recent epidemiologic studies indicate that infections agent may contribute in some psychotic disorder such as schizophrenia [15]. The parasite, *Toxoplasma gondii*, can alter behavior and neurotransmitter function in human such as decreased novelty – seeking behavior, slower reactions, feeling of insecurity and neuroticism [16]. Numerous studies indicate that, although the symptoms of schizophrenia generally do not manifest until late adolescence or early adulthood, the disease process has its origin in earlier stages of brain development. The ability of *T. gondii* to infect the perinatal brain is thus consistent with this aspect of schizophrenia pathogenesis. [17]

A recent study has indicated toxoplasmosis is also correlated strongly with an increase in boy births in humans, leading to an alteration of human sex proportion depending on the antibody concentration, the probability of the birth of a boy can increase up to a value of 0.72 which means that for every 260 boys born, 100 girls are born. [18]

The parasite in Iraq was recorded by machattie in 1939, after that a survey study was achieved by using skin test by Nizi in 1976 [19], then followed by some studies including epidemiology and diagnosis by using IFAT achieved by Fattuhi 1985 [20] and Aldujaili 1998 [21] successively.

As for Ninevah Governorate, studies were achieved by Al-Khaffaf in 2001[22], then followed by an experimental study was made by Al-Hayali in 2002 [23], concerning with the isolation and diagnosis of *T. gondii* from human placenta as preliminary trial to initiate toxoplasmosis and the efficacy of some antibiotics in its treatment induced in Mice Ninevah governorate. As for Aldulaimi, she made an epidemiological, immunological and pathological study in 2002 [24]. Also Ahmed (2006) [25]. She study Toxoplasmosis and its relationship to free radical in serum of women and mice

The aim of present research is to analyze the statistic study for documented information at health care department of the city of Mosul from 1991-2007, and determine the prevalence of IgM antibody to *T. gondii* in

pregnant Mosul women. It is worth mentioning that the current study is a pioneering one in diagnosis this disease by using ELISA test.

Methods

The current study deals with two major aspects: the first is analysing statistical information documented at Health Care Department in Mosul from 1991 up to 2007. For correction of statistic study (1991,2008) were neglected due to incompleteness of record and this is due to the sanctions against Iraq. The second is evaluating serological test by 242 serum sample for immunoglobuline (IgM) by (ELISA) test in order to identify toxoplasmosis, by the kit Toxoplasma IgM Enzyme Immunoassay Test Kit Catalog Number: BC-1087 [26].

Also data were collected by interview after diagnosis of infection by toxoplasmosis using standard questionnaire, Women were first asked how they could avoid toxoplasmosis infection to assess their knowledge about sources of infection, age, parity, educational level, high risk occupations, environmental exposures, contact with cats, diet or provision of piped water, consumption of raw or under cooked meat, raw sausage locally produced dry cured meat any tasting raw meat, eggs and milk while cooking, were recorded for each patient.

Results and Discussion

The results of the statistic study as shown in Fig.(1) represent the distribution of *Toxoplasma gondii* infection during 1991 when the highest infection emerged within the three months of summer: July, August and October. It was decreased to 50% in the other months of the year. The uprising of infection average during the three summer months may be attributed with degree of temp. might have affected abortion through the suggested relationship between protein and calcium which form calmodulin Ca-binding protein (CaM) and there adsorption of fertile egg by increasing or decreasing in c^0 temp. Thus produced calmodulin mainly plays a part in cyclic adenosine monophosphate (cAMP) metabolism.

CaM→Adrenalin(Ad)then→noradrenalin (NA) that has a high level in schizophrenia [27], NA is receptors for cAMP which also has a high level in schizophrenia [28]. Also this increase which takes place in June and July can be caused by cockroaches and flies which are very active in such period of time, they serve as transporting hosts for *T. gondii* carrying cats' fecal material on their bodies [29]. On the other hand, Iraq's bad condition, bad services and weak abilities of society in performing hygiene standard.

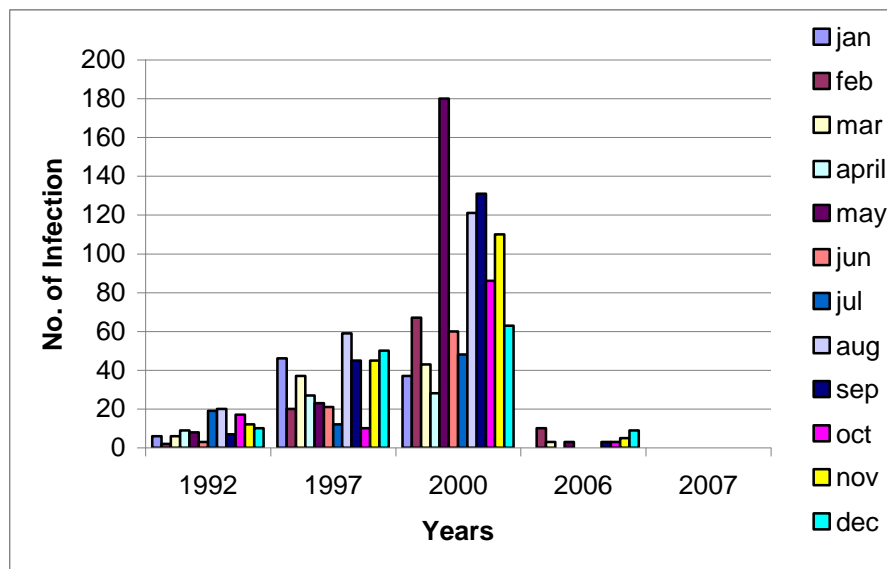


Fig.(1) Represents number of infection from year (1992, 1997, 2000, 2006, 2007)

As for the year of 1997; the average of the monthly infection upraised significantly as shown in figure (2), reached 32.9 percent monthly. it is also noticed that the highest infection was in 2000, as it reached 76.42 and declined in the following years. This difference may be attributed to many social factors such as getting married in certain months, pregnancy and it is related to fertilization and the psychological conditions of Iraqi people. This comes in accordance with what is stated in [30]. As in some cases of a cute toxoplasmosis in adults is associated with psychiatric symptoms such as delusion and hallucination, [15] and some of these findings may be related to the preparation of war condition, the matter which contributes in appearing the infection especially when tests of *T.gondii* are not examined until the appearance of health problems and complications with the pregnant women depending on physician speculations and suggestions Fig.(2) shows

that the highest infection was in the years 2000, 2001 and 2002 this occurred though Iraq witnessed an improvement in individuals incomes, because of the (activation of oil for food and medicine) agreement, while the main public services was still in low levels, in addition to the relative stability and attempts of many civilians to grow domestic animals which shared in passing on and deploying the disease. These high levels of reading between 2000,2002 may be attributed to inaccuracy in making the lab test (private and public labs) or the spread of disease causes overlapping with Toxoplasmosis such as Rubella (German measles), or being infected with cytomegaly (CMV) virus, and herpes simplex (HSV) virus. These three infection can cause a cluster of symptomatic birth defect in newborns. This group of defect is sometimes called TORCH syndrome [31].

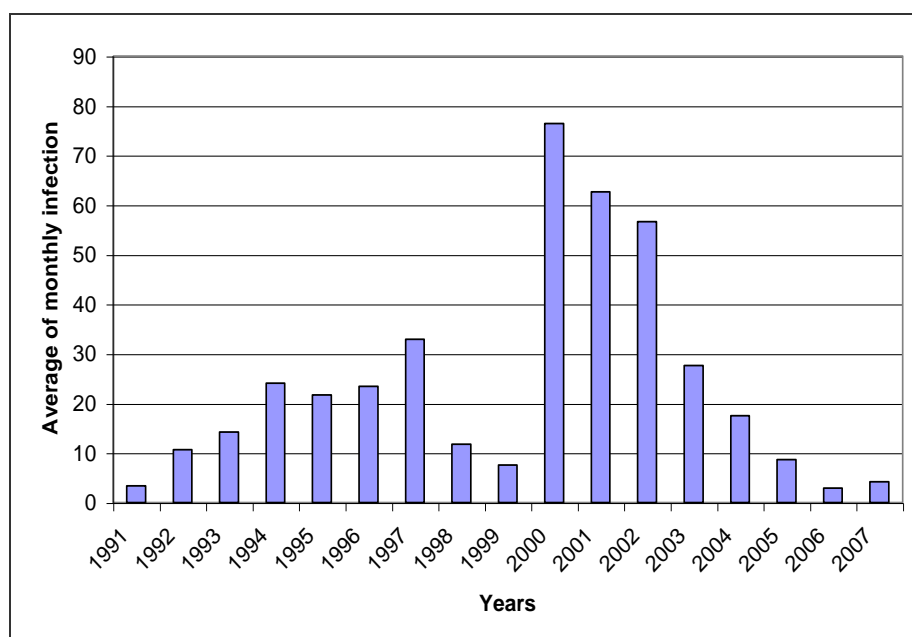


Fig.(2) The average of monthly infection with Toxoplasmosis during the years (1991-2007)

Part two of the study as shown in Fig.(3) shows that 242 sample examined by ELISA test to investigate (IgM) in patients of Al-Salam Hospital in Mosul as suspected of being infected with *T.gondii* revealed that (14) cases

were positive of ELISA examination out of (242). This may be due to overlapping with toxoplasmosis as explain previously in TORCH test

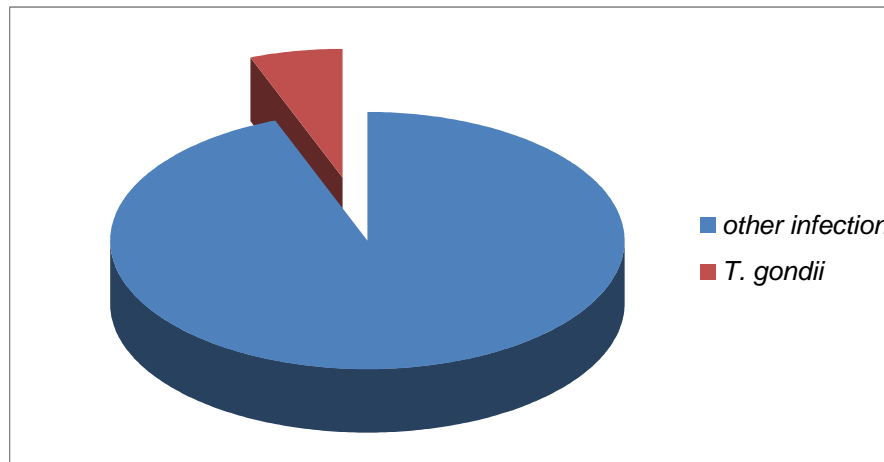


Fig. (3) represent percentage of toxoplasmosis among other overlapping expected infections

Fig (4) show the factional distribution of the infected cases associated with the ages of the pregnant women who carry the disease. Also Fig. reveals that the highest average ratio of the infection is restricted to ages between (15-20) years old were the percentage reached 32% and 18% for the ages between (21-25) years old. Then it decreased to 12% for the ages (26-30) year old, 10% for ages between (31-35) years old. Then the percentage upraise to 20% for the ages between (41-45) years old this result comes in accordance with what is stated in [32] and this is attributed to level of Hygiene,

educational environmental levels and the availability of cleaning facilities and stuff, home sterilization and the psychological state of mother. Meanwhile mothers with higher than 33 years of ages are adhered to high levels of cleanliness more than the others, then the percent decreases in the following ages who suffer from fatigue caused by indoor activities than old women exercise, the matter which affect the levels of hygiene due to that mothers dependencies upon lesser in age and knowledge of preserving cleanliness that share mother duties

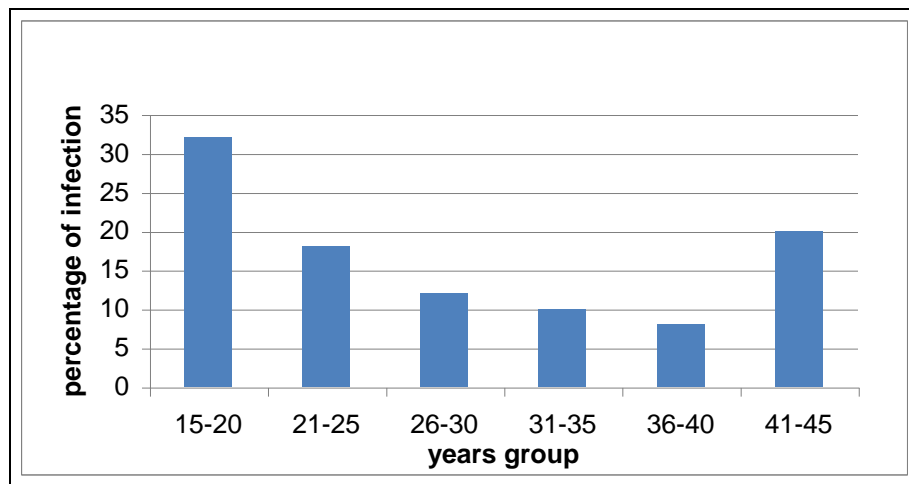


Fig. (4) show the functional distribution of infected cases associated with the ages of the pregnant women carry the disease

The risk of toxoplasma infection was increased in women who reported tasting meat while preparing meals or eating undercooked liver and raw sausage, consumption of un pasteurized milk on milk product the percentage is 40% - 63% also contaminated food with knives, utensil, cutting boards or other foods that have had contact with raw meat.

A percentage of 17% to 37% of infection were attributed to soil contact or unwashed vegetables or gardening which allows contact with infective oocysts deposited

by any recently infected cat. While oocysts take one to five days to become infective, they can remain infective in soil for up to one year [33].

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الملخص

شملت الدراسة الحالية محورين أساسيين، الأول دراسة إحصائية للمعلومات الموثقة لدى قسم الرعاية الصحية في محافظة نينوى للأعوام ١٩٩١ ولغاية ٢٠٠٧، والمحور الثاني تقييم الفحوصات المصلية للتحري عن الكلوبولين المناعي (Immunoglobuline(IgM)، عن طريق فحص الاليزا (ELISA) لتشخيص داء المقوسات Toxoplasmosis. أظهرت النتائج الحالية إن أعلى إصابة لطفيلي *Toxoplasma gondii* كانت للأعوام ٢٠٠٢، ٢٠٠١، ٢٠٠٠، حيث بلغت معدلات الإصابة ٥٦،٦٧، ٦٢، ٤٢، ٧٦ على التوالي. أما نتائج الفحوصات المصلية لداء المقوسات بفحص الاليزا التي أجريت على ٢٤٢ عينة، فكانت ١٤ عينة منها موجبة، إذ بلغ معدل الإصابة ٥،٧٨ %. كما تم تحديد معدل الإصابة للفئات العمرية، وكان أعلى معدل إصابة عند الفئة العمرية (١٥-٢٠) سنة، إذ بلغ ٣٢ %، وبلغ ١٨ % للأعمار (٢١-٢٥) سنة، ١٢ % للأعمار (٢٦-٣٠) سنة والى ١٠ % للأعمار (٣١-٣٥) سنة والى ٨ % للأعمار (٣٦-٤٠) سنة، في حين وصل المعدل إلى ٢٠ % للأعمار (٣٦-٤٠) سنة.