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إشراف

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Abstract

Toxoplasma gondii is an obligatory intracellular protozoal parasite which causes a disease called toxoplasmosis which infects approximately one-third of the world's population and a wide range of animals. Household cockroaches are among widespread insects that constitute a significant public health threat due to their capacity of transmitting medically-important parasites and other pathogens.

The first objective of this study was to evaluate the seroprevalence of toxoplasmosis among female university students living in Diyala Province, middle of Iraq. Blood samples from each participant for serological analysis were obtained and the complete blood count (CBC) has been measured. The anti-*T. gondii* IgG antibodies were detected by using an enzyme-linked immunosorbent assay (ELISA). The results revealed that 44 female students out of 243 (18.1%) were found seropositive for anti-*T. gondii* IgG antibodies, while 199 students (81.9%) were found seronegative. The results showed an odds ratio of 1.85, indicating that the odds of *T. gondii* seropositivity were 1.85 times higher in the female students studying health sciences (MHC) than in the students studying Islamic studies (IS).

The second objective made a comparison between healthy control female students (Toxoplasma-negative) and female students who were Toxoplasma-positive regarding the haematological indices and also regarding the possible correlation between the infection with *T. gondii* (Toxoplasmosis) and six selected hemogram parameters [monocyte-to-lymphocyte ratio (MLR), neutrophil-to-lymphocyte ratio (NLR), platelet-to-lymphocyte ratio (PLR), red cell distribution width (RDW), platelet distribution width (PDW), and mean platelet volume (MPV)] that could be served as markers (diagnostic tools) for the occurrence of *T. gondii* infection.. From each participant, blood has been collected and the complete blood count (CBC) has been measured. The results revealed significant differences in most of the haematological profiles between *T. gondii*-

seropositive and *T. gondii*-seronegative female university students. The total numbers of lymphocytes and neutrophils decreased significantly ($P < 0.05$), while the numbers of monocytes, eosinophils, and platelets ($P = 0.05 - 0.0001$) increased significantly in the bloods of seropositive female students in comparison with the seronegative females. Our results revealed for the first time that the levels of RDW, PDW-CV, MPV, and procalcitonin (PCT) were significantly higher ($P = 0.05 - <0.0001$) in the seropositive females than in their seronegative counterparts. In addition, the results revealed for the first time that the levels of MLR and PLR ($P = 0.05 - <0.0001$), but not NLR were significantly higher ($P = 0.05 - <0.0001$) in the bloods of female students who were infected with *T. gondii* than in their *T. gondii*-negative counterparts.

More future studies are necessary in order to ascertain the potential clinical use of these simple and inexpensive haematological markers in diagnosis of toxoplasmosis.

The third objective of this study was to evaluate the role of Iraqi household cockroaches as potential vectors of medically-important parasites. For this part, fifty cockroaches were collected from different dwelling places of Baquba City, Diyala Province during the period from October to December 2022. The contents of their gut and external body surfaces were examined for the presence of medically-important parasites. The results of this part showed that the overall prevalence rate of parasitic infestation on the external body surfaces of the cockroaches was 60.0%, while the overall prevalence rate of parasitic infection in the guts was 56.0%. Based on capture sites, cockroaches trapped in the toilets had more parasites than those caught from kitchens and rooms. Four species of medically important parasites were isolated from the external body surfaces of the cockroaches; three species were identified as protozoa and only one species was helminth. The protozoan parasites were identified as *Blastocystis spp.* (36.0%), *Giardia spp.* (14.0%) and *Cryptosporidium spp.* (4.0%). In addition, five species of medically important parasites were isolated from the gut contents; four species were identified as protozoa and

only one species was helminth. The protozoan parasites were identified as *Blastocystis* spp. (24.0%), *Giardia* spp. (10.0%), *Cryptosporidium* spp. (8.0%), and *Entamoeba histolytica/ E. dispar* (2.0%). *Enterobius vermicularis* was the only helminth parasite isolated from both gut contents (2.0%) and external body surfaces (2.0%) of cockroaches.

In conclusion, the results showed that cockroaches were found to harbor intestinal parasites of public health importance. Consequently, awareness on the potential role of these insects in the transmission of human intestinal parasites needs to be established.



Chapter One: Introduction

1. Introduction

T. gondii is a protozoan parasite that causes toxoplasmosis, a common disease around the world (Duby, 2013). It is regarded as one of the most "successful" parasites due to its ability to parasitize nearly any warm-blooded creature (Remington *et al.*, 1995; Delgado *et al.*, 2022). Although *T. gondii* infects over two billion individuals annually (Stelzer *et al.*, 2019), the majority of infected people are asymptomatic or only have mild symptoms, with only a small percentage of infected women experiencing severe symptoms (Wilson, 1990). Toxoplasmosis is likely one of the deadliest zoonotic infections in the world due to the parasite's frequency and its effects on pregnant women, which can cause major brain damage that potentially led to the death of the fetus (Wilson, 1990).

Domestic cats and other members of the Felidae family (e.g., tigers, lions, leopards, and cheetahs) are the only final hosts of *T. gondii*, implying that the parasite can only complete its sexual life cycle in the presence of these animals (Frenkel *et al.*, 1970). Postnatally, the disease can be transmitted by eating undercooked or raw meat containing tissue cysts or by ingesting water or food contaminated with infected oocysts from cat feces. A cat suffering from severe feline disease may release up to 100 million parasites every day. These sporozoite-containing oocysts are very infectious and may live for a long time in the soil (Mittal and Ichhpujani, 2011). People must thus restrict their pet cats indoors to prevent *T. gondii* from spreading at night and to limit their proclivity for hunting and urinating outside. Furthermore, uncooked meat should be avoided, and frozen meat should be kept at -12 °C or lower for at least two days before cooking. Furthermore, after coming into contact with any form of raw food, cutting boards, countertops, cutlery, crockery, and other kitchen items must all be properly cleaned (Smith, 2021).

T. gondii antibodies detected in patients may assist in diagnosis. For the detection of humoral antibodies, several serological procedures are available, including the Sabin-

Feldman dye test, the indirect hemagglutination assay, the indirect fluorescent antibody assay (IFA), the direct agglutination test, the latex agglutination test (LAT), the enzyme-linked immunosorbent assay (ELISA), the immunosorbent agglutination assay test (IAAT) (Robert-Gangneux *et al.*, 2012), and the polymerase chain reaction (Savva *et al.*, 1990).

Due to the fact that available drugs are not fully effective, the treatment of toxoplasmosis is a significant challenge for science. Firstly, these drugs are not capable of reaching the bradyzoites inside the tissue cysts found in the muscles and nervous tissues. Secondly, many of these drugs come with severe side effects (Montoya and Liesenfeld, 2004; Peyron *et al.*, 2019; Molan and Ismail, 2021). As a result, continuing health education and laboratory tests for detection appear to be the most reliable strategies for prevention, diagnosis, and early treatment of infection, particularly in pregnant women (a). It is worth noting that previous studies have demonstrated the effectiveness of health education programs in reducing the prevalence of toxoplasmosis and improving knowledge about preventive practices (Bertran-Luengo *et al.*, 2021).

A complete blood count (CBC) is a standard blood test used to assess overall health and investigate various disorders, such as anemia, infection, and leukemia. It is widely available and reasonably priced. However, all the tests mentioned earlier that are used to detect the presence of toxoplasma in the sample (the most commonly used test in Iraq being ELISA) require specific kits and are more time-consuming. Therefore, using blood factors to predict the risk of toxoplasmosis can be a more cost-effective approach and can help mitigate the consequences of the disease, including numerous prenatal infections.

Cockroaches, being one of the most efficient mechanical vectors for a wide range of hazardous pathogens, including fungi, bacteria, viruses, and parasites (Weiss *et al.*, 2009), some of which can cause fatal infections in both domesticated animals and humans, represent a severe public health concern in cities. Several countries have reported a high

prevalence of cockroach-borne human illnesses, with variations in the number of reported cases (Wilson, 2010; Desta, 2021). Although there are over 4,000 distinct species of cockroaches worldwide, only a few of them are considered pests by humans (Salehzadeh *et al.*, 2007). Cockroaches are particularly common in urban regions of countries with tropical and subtropical climates (Galván, 2008).

Objectives of the study:

1. The first objective was evaluating the seroprevalence of toxoplasmosis among female university students in Diyala Province, middle of Iraq.
2. The second objective was investigating the possibility of using some haematological indices as indicators for the presence of *T. gondii* infection by comparing these indices between females' university students infected with *T. gondii* (*Toxoplasma*-positive) and who were not infected with *T. gondii* (*Toxoplasma*-negative).
3. The third objective was to assess the possible difference in infection with *T. gonidii* between female students who studying subjects related to health sciences and those studying Islamic studies
4. The fourth objective was to examine the possible relationship between toxoplasma infection and various demographic factors, such as age, marital status, and residency (urban or rural).
5. The fifth objective was detecting the role of cockroaches in the transmission of parasites and specifically *T. gonidii*.