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Serological Study of the Relationship Between Toxoplasmosis Patients and Dopamine and Adrenaline Levels

A Thesis

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By

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قَالَ رَبِّ اشْرَحْ لِي صَدْرِي ﴿٢٥﴾ ويَسِّرْ لِي أَمْرِي ﴿٢٦﴾ وَاحْلُلْ عُقْدَةً مِنْ لِسَانِي ﴿٢٧﴾ يَفْقَهُوا قَوْلِي ﴿٢٨﴾

صَدَقَ اللَّهُ الْعَلِيُّ الْعَظِيمُ

سورة طه

Dedication

This thesis is dedicated to:

The great martyr, my brother **AHMED**.

The pure souls of my father and mother.

My support in life….. my husband.

My beloved brother and sisters.

My hope in life…… my children.

My big family and my friends.

I dedicate this research.

Zahraa

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Finally, I wish to express my thanks to all individuals who cooperated with me, and without their help, this work would not have been accomplished.

Supervisor Certification

I certify that this thesis which is entitled

(Seroepidemiological Study of Toxoplasmosis and its Relationship with the Neurotransmitters: Dopamine and Adrenaline)

Presented by (Zahraa Khalid Mijbel) was prepared under my supervision in the Department of Biology / College of Science as partial fulfilment of the requirements for the Degree of Master in Biology.

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Date: / / 2022

Head of Department Advice

In view of the recommendations available, I forward this thesis to the examination committee.

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Date: / / 2022

Summary

Toxoplasma gondii is a globally widespread parasite that infected about 30–50% of the world's population and caused toxoplasmosis, which infected more than 300 species of warm-blooded animals, including humans (according to World Health Organization data) , which are considered intermediate hosts, and cats, which are the definitive hosts.

The current study was carried out in Misan Province, Southern Iraq, to investigate the spread of toxoplasmosis and to determine its relationship with levels of the neurotransmitters dopamine and adrenaline. 174 venous blood samples (153 males, 21 females) were collected from individuals who visited Al-Shaheed Al-Sadder teaching hospital and the main blood bank in Misan from December (2020) to October (2021).

The enzyme-linked immunosorbent assay (ELISA) was used to determine the levels of the immunoglobulin as anti-*Toxoplasma* Immunoglobulin M (IgM) and immunoglobulin G (IgG), as well as to determine the levels of dopamine and adrenaline in the blood of the participant individuals. The Complete Blood Count (CBC) test was used to identify the effect of the *Toxoplasma gondii* on the contents of this test.

Some socio-demographic factors of the participant individuals were recorded in a questionnaire form that was designed to evaluate their association with toxoplasmosis.

The results are summarized in the following points:

1- The percentage of the toxoplasmosis-seropositive is 52.3%, including 32.97% is positive for IgM alone, 38.46% for IgG, and 28.57% for IgM and IgG together.
2- The infection rate for females is 66.67% and for males is 50.33%.
3- There are significant differences between the dopamine level of toxoplasmosis positive and negative sera. It shows the dopamine level of toxoplasmosis-infected individuals is higher at about 163.347% than uninfected individuals.
4- The level of adrenaline in *Toxoplasma*-seropositivity is higher than that of *Toxoplasma*-seronegativity with significant differences between them.
5- There is a statistically significant decrease in the hemoglobin level and RBCs count in the blood of toxoplasmosis-infected individuals compared with uninfected individuals.
6- The counts of WBCs and platelets in the blood toxoplasmosis-infected individuals are decreased compared with uninfected individuals but no significant differences between them have been noticed.
7- There is a statistically significant association between the level of education and income rate and toxoplasmosis.
8- There is a statistically significant relationship between the history of surgery, blood transfusion, fever, headache, muscle pain, fatigue and toxoplasmosis.
9- There is a statistically significant relationship between contact with cats and the infection with toxoplasmosis.
10- There is no significant relationship between *Toxoplasma*-seropositivity and vision problems and gland dysfunction.

11- There is no significant relationship between *Toxoplasma*-seropositivity and some behavior like drinking unpasteurized milk or eating dairy products from street vendors and cleaning the house garden.

12- There are statistically significant relationships between the *Toxoplasma*-seropositivity and late pregnancy, abortion, and women whose children were born with birth defects.

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List of Abbreviations

Abbreviation	Full name
ACTH	Adrenocorticotropic Hormone
AD	Adrenaline
AIDS	Acquired Immune Deficiency Syndrome
BSA	Bovine Serum Albumin
C	Celsius
CNS	Central Nervous System
DA	Dopamine
EIA	Enzyme Immunoassay
ELISA	Enzyme linked Immunosorbent Assay
EPI	Epinephrine
HCT	Hematocrit Test
HIV	Human Immunodeficiency Virus
HRP	Horseradish Peroxidase
IFAT	Immunofluorescence Antibody Test
IFN	Interferon
IgG	Immunoglobulin Gama
IgM	Immunoglobulin Mega
IL	Interleukin
IU	International Units
L-DOPA	Levodopa
MAT	Miller Analysis Test
OD	Optical Density
PBS	Phosphate Buffered Saline
PDW	Platelet Distribution Width

PLT	Platelet Count
PNMT	Phenyl ethanolamine N methyl transferase
RBC	Red Blood Corpuscle
SA	Spinal Anaesthetic
SAM	S-Adenosyl Methionine
SPSS	Statistical Package for the Social Sciences
SD	Standard Deviation
TMB	Tetramethylbenzidine
TNF	Tumor Necrosis Factor
WBC	White Blood Cells
WHO	World Health Organization

CHAPTER ONE

INTRODUCTION

AND

LITERATURES

REVIEW

1.1 Introduction

Toxoplasmosis is an example of the One Health concept because the causative parasite *Toxoplasma gondii* affects nearly all warm-blooded species, including humans. As a result, *T. gondii* is among the most prolific parasites on the planet, affecting up to a third of the global populace (Djurković-Djaković *et al.*,2019).

Primary infection is frequently asymptomatic or accompanied by self-limited symptoms such as fever, malaise, and cervical lymphadenopathy in immunocompetent people. Transmission of *T. gondii* to the fetus is frequently related to the infection acquired during pregnancy, resulting in congenital illness (Al-Mayahi 2011). *T. gondii* infection causes severe symptoms in immunocompromised people, including splenomegaly, chorioretinitis, pneumonitis, encephalopathy, multisystem organ failure, and even death (Montoya and Liesenfeld, 2004).

Humans obtained toxoplasmosis either from the environment or through the placenta, through the mother to her fetus which is resulting in congenital toxoplasmosis (Mahmud *et al*., 2017), clinical toxoplasmosis can be either congenital or acquired, and symptoms vary according to the immune status of the infected person, in immunosuppressive diseases such as AIDS, toxoplasmosis infection is one of the most important challenges facing its victims, which is often fatal. In some individuals, toxoplasmosis may show up some days or extend for a long time as years after being born, and the symptoms of congenital toxoplasmosis may be seen in weeks, or extend for long years after being. Retinitis, intracerebral calcification, psychopathy,

and hydrocephalopathy are some of the signs and symptoms of congenital toxoplasmosis (Mahmud *et al*., 2017).

In the last years, the prevalence of *Toxoplasma* has dropped. Simultaneously, in certain Asian countries, the prevalence has risen from around 5% to over 10%. (Flegr *et al*.,2014). *Toxoplasma* oocysts in water, food, or soil contaminated by cat feces, or *Toxoplasma* cysts found in raw or undercooked meat, infect humans. (Robert-Gangneux *et al.*, 2012). Typically, the musculoskeletal system, brain, and eyes are where parasites are detected in humans (Mahmud *et al*.,2017).

Toxoplasma gondii, a common parasite, manipulates the behavior of its host. The behavioral alterations in infected rodents improve the chances of the parasite being transmitted back to its definitive cat host, which is an important phase in the parasite's life cycle (Prandovszky *et al.*,2011), although the mechanism causing behavioral changes in the host is unknown, two lines of evidence point to parasite neurotransmitter signal transduction: disruption of parasite-induced behavioral changes with psychiatric medications (specifically dopamine antagonists) and identification of a tyrosine hydroxylase encoded in the parasite genome (Prandovszky *et al.*,2011). *Toxoplasma* has been shown to alter not just the behavior of its intermediate animal hosts, but also the behavior and personality of infected people. increasing in dopamine and testosterone in males, as well as hypomethylation of particular regulatory regions of critical genes in the infected host's amygdala, are the most likely mechanisms responsible for the observed behavioral abnormalities (Flegr 2015).

Dopamine is a monoamine neurotransmitter that is most known for its involvement in the bodies when compared to other neurotransmitters (Basu and Dasgupta 2000).

Adrenaline is a monoamine that belongs to a group of monoamines known as catecholamines (Lieberman *et al*., 2013).

1.2 Aim of the study

The study aimed to:

1- Determine the seroprevalence and intensity 0f *Toxoplasma gondii* infection by using the ELISA test to determine the levels of IgG and IgM antibodies in Al- Amara city.

2- Study the relationship between toxoplasmosis and the levels of some neurotransmitters, such as dopamine and adrenaline in humans by using the ELISA test.

3- Study the impact of *T. gondii* on some parameters of blood picture (complete blood count 'CBC').

4- Study the relation between toxoplasmosis and some sociodemographic factors.

Literature Review

1.3 Toxoplasmosis:

1.3.1 Historical view:

In the laboratory of Charles Nicolle at the Pasteur Institute in Tunis, a protozoan was found in the tissues of a rodent used to study leishmaniasis known as the *gundi*, *Ctenodactylus gundi*, by Nicolle and Manceaux in 1908. At first, Nicolle thought the parasite was a piroplasm (Ajioka and Soldati 2007), and then *Leishmania*, but quickly realized he had found a new species, so he gave it the names *T. gondii* and *T. gondii* based on the host and morphology (mod. L. toxo = arc or bow, plasma=shape) (Nicolle and Manceaux 1909). Consequently, its full name is *T. gondii* (Nicolle and Manceaux 1909). The following 30 years saw the discovery of *T. gondii*-like entities in a range of hosts, notably bird species (Sabin and Olitsky, 1937), (Dubey 2002).

Isolated *T. gondii* for the first time, and used cross-protection to verify that it was similar to the human isolate of *T. gondii*. *Toxoplasma. gondii* protection turned revealed to be a complicated process involving both innate and specific immunity (Sabin and Feldman 1948). Humoral antibodies were discovered in the 1940s to attack external but not intracellular tachyzoites (Sabin and Olitsky 1937; Sabin and Feldman 1948). Over the next 50 years, immunological lymphoid cells were discovered to have a major role in protective immunity (Frenkel 1967; Suzuki *et al*., 1988; Gazzinelli *et al*., 1991). Despite the fact that *T. gondii* has a worldwide distribution and possibly the broadest host range of any parasite, the genus *Toxoplasma* has only one species, *gondii*. It's unclear why some hosts get clinical toxoplasmosis and

others don't. Methods for recognizing genetic changes between *T. gondii* isolates from humans and animals were established during the 1980s and 1990s (Howe and Sibley 1995).

1.3.2 Classification of *T. gondii*:

Toxoplasma gondii is a coccidian parasite with cats as the definitive host, and warm-blooded animals as intermediate hosts. It belongs to:

Kingdom: Animalia

Phylum: Apicomplexa; Levine,1970

Class: Sporozoasida; Leukart, 1879

Subclass: Coccidiasina; Leukart, 1879

Order: Eimeriorina; Leger, 1956

Family: Toxoplasmatidae; Biocca, 1956

Genus: *Toxoplasma;* Nicolle and Manceaux, 1909

Species: *Toxoplasma gondii* (Dubey 2010).

1.3.3 Morphology:

Oocyst, Tachyzoite, Bradyzoite and Tissue cysts are the forms of *Toxoplasma gondii* (Fig 1-1, Fig 1-6). Cats are the definitive host and all the forms exist in cats. The intermediate hosts have tachyzoites and tissue cysts. All of these forms are contagious to humans (Mahmud *et al*., 2017).

1.3.3.1 Oocyst

The parasitic apicomplexan *Toxoplasma gondii* can survive in the environment as an infectious stage known as an oocyst (Shapiro *et al*.,

2019) (Freppel *et al*., 2019). *T. gondii* oocysts, the zoonotic parasite's environmentally resistant stage, play a critical part in the parasite's epidemiology (Dumetre *et al*., 2012), the global distribution of *T. gondii* and how it has developed to be one of the most prevalent infectious agents of animals and humans may be explained further by the biological characteristics of oocysts (Dubey, 2004; Dumetre *et al*., 2012).

Many birds and mammalian species, including humans, are susceptible to oocyst infection, following a 1–2week sporulation phase, oocysts are deposited in cat feces and become infectious. Sporulated oocysts are ∼13 × 11μm long and contain two sporocysts, each with four potentially infective sporozoites shielded by the sporocysts and oocyst walls from harsh environmental conditions (Shapiro *et al.*, 2019).

By gametogony (sexual reproduction), this form of the parasite develops in the epithelial cells of the final host (cat's) gut (Paniker, 2002). Infected household cats or wild felines could contaminate the environment with *Toxoplasma* oocysts. A single cat can shed over 100 million oocysts into the environment after primary infection (Tenter *et al*., 2000). If they receive sufficient aeration, humidity, and heat, oocysts can sporulate and become infectious in less than a day. Depending on the *Toxoplasma* strain, ingesting not less than 10 sporulated oocysts might cause the disease in intermediate hosts (Dubey *et al*., 1996). Unsporulated oocysts discharged in feline feces are not immediately infectious (Tenter *et al*., 2000). As a result, *Toxoplasma* infections are rare when people come into direct touch with cats (Tenter *et al*., 2000). On the other hand, the environment may include sporulated oocysts that could infect humans and other

intermediary hosts (Fig 1-4), in several case-control research on human infections with *Toxoplasma*, consuming undercooked meat was identified as the main risk factor (Cook *et al*., 2000).

Toxoplasma sporulated oocysts are extremely resistant to environmental conditions. They can endure short periods of cold and dehydration, and they can live for up to 18 months in moist soil or sand (Frenkel, 2000). sporulated oocysts survived storage at 4°C for up to 54 months and freezing at - 10°C for 106 days in the lab. Heat reaching 55- 60°C kills them in 1-2 minutes (Dubey, 1998). The highly impermeable nature of sporulated oocysts makes them resistant to disinfectants (Tenter *et al*., 2000).

1.3.3.2 Tachyzoite

Tachyzoites are obligate intracellular parasites that can penetrate, replicate, and multiply in the cytoplasm of parasitophorous vacuoles in all mammalian cells except anuclear erythrocytes by endodyogeny (Kasper, 2005). Frenkel (1973) created the word "tachyzoite" (tachos = speed in Greek) to designate the stage that replicated quickly in any cell of the intermediate host and in the non-intestinal epithelial cells of the definitive host. tachyzoites are also known as endodyozoites or endozoites (Dubey *et al*., 1998).

The crescent-shaped trophozoites (tachyzoites) are (2 - 3) µm wide and (4 - 8) µm long (Garcia 2007), one end has a more rounded shape than the other (Garcia 2007). Tachyzoites can move by gliding, bending, undulating, and spinning, although they lack apparent locomotion mechanisms like cilia, flagella, and pseudopodia (Fig1-5) (Frenkel, 1973). They are encased in a membrane of the host cell known as a

pseudo cyst, and they have a spherical appearance. Up until the host cell lyses or a tissue cyst forms, intracellular proliferation persists (Kasper, 2005). Numerous intermediate hosts, including cows, sheep, and goats, have been reported to carry *Toxoplasma* tachyzoites in their milk, semen, saliva urine, tears, and sputum (Tenter *et al*., 2000).

Tachyzoites are vulnerable to proteolytic enzymes and are typically destroyed by gastric digestion; however, it has been hypothesized that on rare occasions, they may enter the host through mucosal tissue penetration and do so in order to access the host's circulation or lymphatic system before reaching the stomach (Johnson, 1997).

1.3.3.3 Tissue cysts

In the intermediate host, tissue cysts represent the final step of the life cycle and are immediately infectious. They can live for the rest of the host's life in some intermediate host species, such as most livestock. The parasite invades cells of intermediate hosts (as well as the definitive host, felines) and forms intracellular parasitophorous vacuoles harboring bradyzoites, the parasite's slowly replicating form (Dubey *et al*., 1998).

Tissue cysts show a strong preference for neurological and muscle tissues in a wide range of hosts, they're mostly found in the central nervous system (CNS), the eye, and the skeletal and cardiac muscles, they can also be found in visceral organs such as the lungs, liver, and kidneys, though to a lesser level (Dubey *et al*., 1998).

Bradyzoites are a long-lived, slow growing form of *Toxoplasma gondii* found during the chronic phase of toxoplasmosis, the intracellular bradyzoites form cysts in tissues (Fig1-2), such as muscles and the brain

where they can evade the immune response (Elsheikha &JarroII 2017). Frenkel (1973) created the name "bradyzoite" (Brady = slow in Greek) to characterize the organism that multiplies slowly within a tissue cyst. Bradyzoites are also known as cystozoid (Dubey *et al*. ,1998). The cyst is oval or circular in shape and ranges in diameter from 10 to 200 µm, it contains bradyzoites, which are slow-growing trophozoites. It's discovered during a persistent latent (asymptomatic) infection and can last for years in human tissue (Maclean, 2005).

After infection, the cysts stay alive and survive in practically every tissue for months or even years, but skeletal and cardiac muscles, as well as the central nervous system (CNS), appear to be the most prevalent sites of chronic infection (Willis *et al*., 2002).

Some *Toxoplasma* strains are allegedly resistant to freezing, despite the fact that most tissue cysts perish around -12°C or less (Dubey, 2000). It's worth noting that cooking for a prolonged period of time in a home environment may be essential to achieve the temperatures necessary to eliminate *Toxoplasma* tissue cysts in the meat (Karem 2007). Certain tissue cysts will continue to be contagious if uneven heating techniques, including microwave heating, are used during the cooking process (Lundén *et al*., 1992). Therefore, meat that is raw or undercooked needs to be fully cooked (to a temperature of 67°C or more) before consumption. While deep-freezing meat (at a temperature of -12°C or lower) before cooking can help reduce the risk of infection, freezing by itself does not guarantee that all tissue cysts will become non-infectious (Paul, 1998; Cook *et al*., 2000).

1.3.4 Life cycle

The *Toxoplasma gondii* life cycle is complicated, with oocysts produced after a final sexual phase of reproduction in the intestines of Felidae family members (definitive hosts) (Fig 1-4). The parasite reproduces asexually in a wide variety of intermediate hosts. *Toxoplasma gondii* survives in the intermediate host by converting from the proliferative tachyzoite stage to quiescent encysted bradyzoites (Djurković-Djaković *et al.*,2019).

Tachyzoites, bradyzoites found in tissue cysts, and sporozoites found in sporulated oocysts are the three phases of the life cycle that can infect any host, including humans (Montoya and Liesenfeld, 2004). The sexual stages are only produced available by members of the Felidae family (Dubey, 2009), and therefore they are considered definitive hosts. Recently, linoleic acid supplementation and the reduction of murine-delta-6-desaturase activity in the gut allowed researchers to examine the parasite's sexual development in mouse intestinal cells (Maner and Moosavi 2022)).

Cats throw out oocysts in their feces (Mahmud *et al*., 2017). The unsporulated oocyst must sporulate for 1 to 5 days before it can become infectious after elimination (Ruelhmann, 2010). Infection occurs in animals and humans who consume oocysts (for example, by eating unwashed vegetables) or tissue cysts in inadequately cooked meat (Karem 2007). After ingestion, sporozoites exit from the sporocyst and oocyst walls and penetrate host enterocytes, lamina propria macrophages, and dendritic cells before transforming into tachyzoites (Delgado Betancourt *et al*., 2019). Proteolytic enzymes break down the

cyst walls, releasing bradyzoites (Ortega, 2007; Jones and Dubey, 2010).

When sporozoites and bradyzoites infect humans, they enter the intestinal mucosa and proliferate asexually through endodyogeny to produce tachyzoites, by way of the circulatory system, tachyzoites are carried to various extraintestinal organs where they develop into tissue cysts, many different organs can develop cysts (Mahmud *et al*.,2017). If a definitive host consumes tissue cysts, the bradyzoites enter a second asexual phase of growth in small intestine epithelial cells (Karem 2007). The cysts survive transit through the cat's stomach (Fig 1-3), and the parasites infect epithelial cells of the small intestine, where they reproduce sexually and asexually and generate oocysts (Remington *et al*., 2004). In the small intestine, sporozoites escape from sporocysts and oocysts. Some sporozoites reach epithelial cells and remain to commence an enteroepithelial cycle in cats, whereas others pierce the mucosa to begin development in the lamina propria, mesenteric lymph nodes, and other distant organs, as well as white blood cells. There is no enteroepithelial growth in hosts other than cats; sporozoites enter host cells and multiply by endodyogeny. Tachyzoites are the rapidly dividing stages in acute infections. Before a host cell's parasitophorous vacuole disintegrates, eight to 32 tachyzoites collect within the parasitophorous vacuole, releasing parasites to infect new cells (Schmidt *et al*.,2005). Merozoites are produced by the asexual multiplication (schizogony) of the zoites secreted in the small intestine. Some Merozoites travel outside of the intestine and settle in tissues or organs, where they develop tissue cysts. Some Merozoites mature into male and female gametocytes that produce microgametes and

macrogametes to start the sexual cycle (gametogony), a macrogamete that has been fertilized by a motile microgamete grows into an oocyst that sporulates in the soil after being released in the cat's feces. An oocyst with eight sporozoites serves as the infectious form (Mahmud *et al.*,2017).

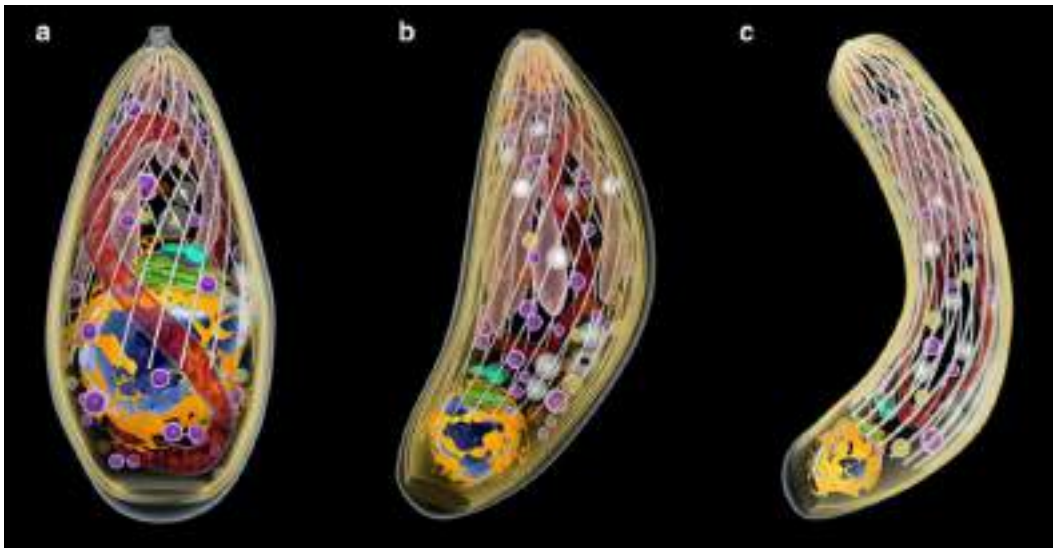


Fig. (1-1) The infective stages of *Toxoplasma gondii* (a) tachyzoite, (b) bradyzoite, (c) and sporozoite (Attias *et a*l.,2020).

Fig. (1-2) Tissue cyst of *Toxoplasma gondii* (Elsheikha &JarroII 2017).

Fig. (1-3) The life-cycle of *Toxoplasma gondii* in cat (Attias *et al*.,2020).

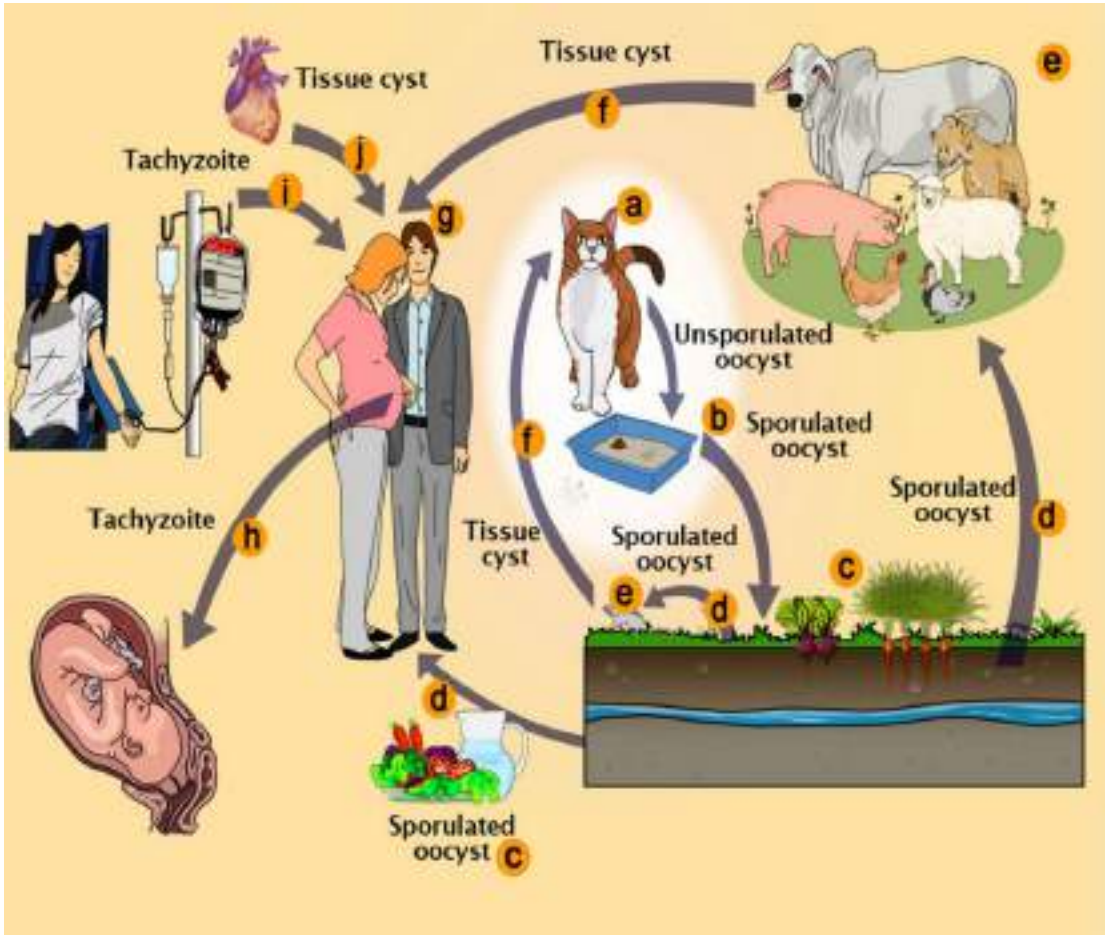


Fig. (1-4) Methods of transmission of *Toxoplasma gondii* (Attias *et al.*,2020).

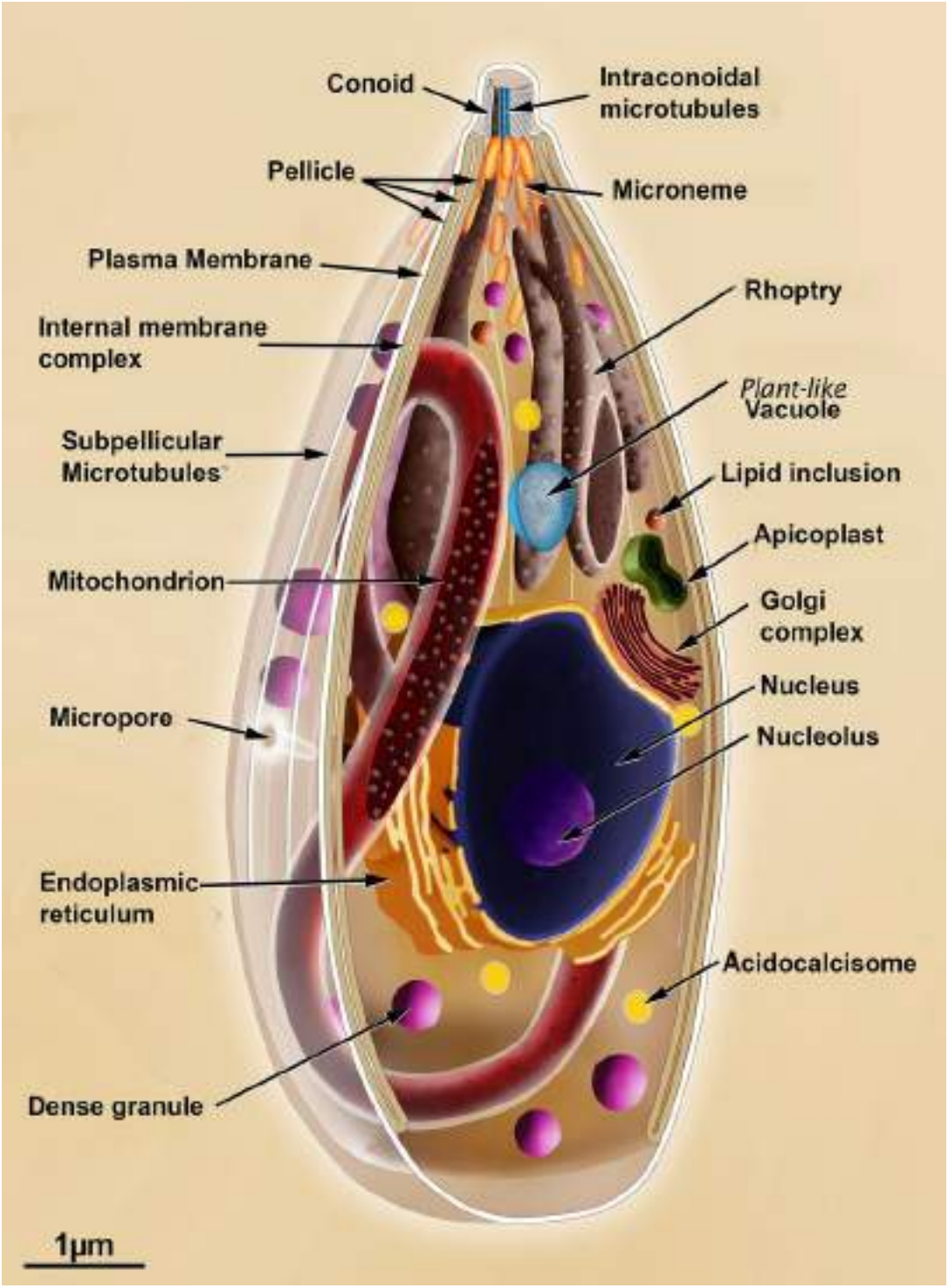


Fig. (1-5) The main components and organelles in the longitudinal section of the *Toxoplasma gondii* (tachyzoite form) (Attias *et al*., 2020).

1.4 Infectiousness and Virulence

Pathogenicity and infectiousness of *Toxoplasma gondii* depend on interactions between the parasite and the host's immune system (Dubremetz and Lebrun, 2012). The population structure of *T. gondii* is composed of genotypes I, II, and III as well as a wide variety of genotypes (Su *et al.*,2012). In mice, genotype I is typically associated with highly virulent strains, while genotype II and III count for the vast majority of non-virulent strains (Sibley and Boothroyd, 1992). The virulent characteristics of the genotype I strain in mice may also impact humans, given that it has been linked to serious eye illness in otherwise immune-competent people (Boothroyd and Grigg, 2002). Less research has been done and more reliance is placed on epidemiological data when it comes to strain-specific pathogenicity in humans. According to Howe and Sibley (1995), the majority of human cases are caused by genotype II, which is most likely a result of this genotype's overrepresentation in animals in the USA and Europe, where the bulk of human cases have been detected (Boothroyd and Grigg, 2002). Small genetic changes result in unique behavioural changes in pathogenicity and infectivity because of the close genetic links among the clonal lineages (Sibley and Ajioka, 2008).

Recombinant and atypical genotypes are poorly understood. With little knowledge of human infection, most virulence research has concentrated on genotypes I, II, and III and has usually assessed virulence in mice pathogenicity (Dubremetz and Lebrun, 2012). Recent cases of severe sickness in immunocompromised individuals, foetuses and otherwise healthy individuals in French Guiana and Brazil were brought on by extremely virulent atypical genotypes (Dubey *et al.*,

2013). Atypical and type II-like strains from native Australian wildlife have been isolated. All of the Australian isolates examined thus far were not virulent in mice bioassays (Parameswaran *et al*., 2010).

1.5 Epidemiology:

Toxoplasma gondii is found all across the world, however, the prevalence of the parasite varies widely (Jones *et al.*, 2001). In European countries such as France and Italy, seroprevalences of 43 % and 28.3 %, respectively, have been observed (Berger, *et al*; 2009; De Paschale *et al.*, 2010).

Toxoplasmosis seroprevalence was reported to be high in Asian countries, ranging from (41.6% - 45 %), and in Thai pregnant women 29.1% (Nissapatorn *et al.*, 2011). In Brazil, there was a 77.5% seroprevalence of toxoplasmosis (Flegr 2013). In Canada (0.2%) and (40%) in the United States of America (Bigna *et al.*, 2020). According to data from a number of African countries, the prevalence of Toxoplasmosis during pregnancy varies. In Côte d'Ivoire and Nigeria, for example, seroprevalence rates of 60% and 43.7 percent have been reported (Adou- Bryn, 2004). Morocco has a seroprevalence rate of 50.6 % for toxoplasmosis (El Mansouri *et al.*, 2007). In Ghana (92.5%) (Pappas *et al.*, 2009), in Senegal, has been reported34.5% of *Toxoplasma* seroprevalence (Ndiaye *et al.*, 2011).

The large prevalence range could be attributed to a number of variables that affect how likely it is for people to become infected with *T. gondii*. Aspects to take into count include the environment, healthcare resources, socioeconomic conditions, geographic location, and personal traits including spending habits and behaviour (Bigna *et al*., 2020). The

large differences in incidence among regions and countries may be due to geographical factors such as climate (rainfall, temperature). Heat and humidity, for example, are regarded to be crucial elements in oocyst conservation in the soil, and hence in sustaining a high frequency. This disparity in prevalence has also been linked to eating patterns (Cenci-Goga *et al.,* 2011).

Seroprevalence in the United States ranged from 29.2 percent in the northeast to 20.5 percent and 17.5 percent in the South-Midwest and west, respectively. Similarly, the prevalence of toxoplasmosis appears to differ greatly between HIV-positive and HIV-negative adults. For example, a study of HIV-positive and HIV-negative people in Zambia found a seroprevalence of 7% (Zumla *et al.,* 1991).

Another study on a comparable cohort in Ethiopia in 1991 found an 80 percent seroprevalence (Woldemichael *et al.,* 1998). The cause for this may not be evident, but it could be due to a variety of factors including socioeconomic position, education, sanitary conditions, and dietary preferences. In Rwanda, there is a scarcity of historical knowledge about the disease. In a study conducted in two remote communities, N genda and N yarutovu, currently in Bugesera district eastern province, 50 percent of the adults in both areas tested positive for *T. gondii* antibodies (Gascon *et al.,* 1989).

Toxoplasma gondii seroprevalence varies by age in Germany, with about 20% for 20-year-olds, 30% for 30-year-olds, and so on (Gross,1994). *Toxoplasma gondii* usually causes no or very minor symptoms in immunocompetent persons. Reactivation of lingering bradyzoites in immunocompromised people, such as AIDS patients or those on immunosuppressive medication, can result in life-threatening

toxoplasmosis with encephalitis, CNS abnormalities, or chorioretinitis (Ferreira *et al.,* 2002).

A study in Iraq found that the high prevalence of toxoplasmosis among the investigated high-risk women in the Al-Hawija and Al-Baiji districts was due to a number of risk factors, including age, a number of deliveries, contact with host animals (small ruminants), contact with raw meat, drinking raw sheep or goat milk, as well as a lack of studies on the disease, poor health education, ineffective medication, no surveys, and possible environmental pollution with the rat. The disease was heightened following the Iraq occupation, with a frequency of infection of more than 40%, due to environmental pollution with organisms caused by sanctions and a succession of wars that targeted the country (Al- Jebouri *et al.,* 2013).

1.6 Clinical manifestations:

1.6.1 Toxoplasmosis in immunocompetent patients:

In the vast majority of adults and children, *Toxoplasma gondii* infection is asymptomatic (Remington *et al.,* 2005). The most common symptom in (10%–20%) of normally immune-competent individuals with a symptomatic initial *T. gondii* infection is lymphadenopathy (Montoya and Remington, 2000). About 80%–90% of the clinical indications in this group are asymptomatic and self-limited after a few weeks to months (Schmidt *et al*., 2005) Lymphadenopathy symptoms include a maculopapular rash, fatigue, weakness, pharyngitis, malaise, sore throat, hepatosplenomegaly, myalgia, and distinctive lymphocytosis (Cox and John-Alde, 2005).

1.6.2 Toxoplasmosis in immunodeficient patients:

Immunocompromised patients are more vulnerable to toxoplasmosis, for instance those with hematologic malignancies, solid organ transplants, bone marrow transplants, or AIDS. The most prevalent toxoplasmosis symptom in those with impaired immune systems is toxoplasmic encephalitis (Remington *et al*., 1983). Patients with toxoplasmic encephalitis exhibit a wide range of clinical symptoms, such as altered mental status, seizures, paralysis, cranial nerve disturbances, sensory abnormalities, cerebellar indications, meningismus, movement difficulties, and neuropsychiatric manifestations (Liesenfeld *et al*., 1997).

1.6.3 Congenital toxoplasmosis:

Congenital toxoplasmosis is brought on by *T. gondii* infection of the placenta during pregnancy. More than 90% of pregnant women who contract a primary infection do so without showing any symptoms (Montoya and Rosso, 2005). If they contract the infection prior to being pregnant, the organism will be in the cyst form and the placenta won't be able to accommodate any trophozoites. If a mother has an illness again while pregnant but has immunity from an earlier infection, the organism might not pass from mother to fetus (Warren, 2006). In comparison to the first and second trimesters, the probability of developing an infection increases in the final trimester (Black and Boothroyed, 2000).

1.6.4 Ocular toxoplasmosis:

In roughly 25% of cases, ocular toxoplasmosis causes permanent vision loss in the affected eyes (Bhopale, 2003). Although a sizable portion of

children with congenital infections develops an ocular disease, the conventional wisdom that congenital infection is the primary cause of most cases has been disputed (Gilbert and Stanford, 2000). Infected infants may have retinochoroiditis, which is inflammation of the retina and choroid with accompanying vitritis, when they are born (Mcleod *et al.,* 2006).

The majority of instances of ocular toxoplasmosis are caused by latent infection reactivation rather than the initial infection, according to epidemiological data. Many factors influence disease progression, including the host's immunological response, the parasite's virulence, and environmental conditions. Even in the absence of treatment, ocular toxoplasmosis can cure on its own after two to three months. A study of the ophthalmic literature reveals that massive multicenter clinical studies have failed to prove any conventional therapy (Silveira, *et al*., 2002).

In a population-based study in Britain, the lifetime risk of symptomatic *T. gondii* associated ocular disease was determined to be 18 in 100,000 individuals (Gilbert *et al.,* 1999). In Germany, 4.2% of uveitis cases are thought to be due to *T. gondii* infections (Maenz *et al.,* 2014). Acute and reactivating infections in immunocompromised persons (e.g. AIDS patients or transplant recipients) can affect the central nervous system. An incidence of 3% of cerebral toxoplasmosis, associated with poor prognosis, was found among allogeneic hematopoietic stem cell transplant recipients (Schmidt *et al.,* 2013), (Hakko *et al.*,2013).

1.7 Immune Responses:

The early removal of *T. gondii* from peripheral blood during acute infection is owing to a coordinated and efficient systemic immunological reaction (Mordue and Sibley, 1997). In the infected host, *Toxoplasma gondii* causes a powerful humoral and cellular response (Lori *et al.,* 2002).

1.7.1 Humoral Immunity:

The immune response to infection changes depending on the stage of infection, so some antibodies present at one stage may be missing at later stages, and vice versa. This necessitates the presence of several epitopes from various antigens in an immunoassay to identify antibodies present in various disease states (Pelloux *et al.,* 2006). Antibodies directed against *Toxoplasma* antigens at a high level (IgM, IgA, and IgG) (Roitt *et al.,* 2001).

Patients with toxoplasmosis did not have an increase in Ig E (Jones, 2006). In the early stages of infection, IgM and IgA levels rise in lockstep; later, IgG develops. IgM and IgA levels normally fall and vanish after 2–8 months (Correa *et al.,* 2007). IgM antibodies might last for over a year in some cases (Flori *et al.,* 2008). When a specific antibody is coupled with a compliment, the extracellular tachyzoites are lysed (Mahalakshmi, 2006). Antibodies inhibited the ability of tachyzoites to penetrate different cells, and they were easily absorbed by macrophages (Nash *et al.,* 2005). Elevated *T. gondii* IgG antibodies indicate a past infection, even though elevated *Toxoplasma*-specific IgG levels might last for several years after primary infection. (Nazan, 2008).

1.7.2 Cellular Immunity:

The high incidence of toxoplasmosis in the human immunodeficiency virus-infected population prior to the introduction of highly active antiretroviral therapy demonstrates the importance of cellular immunity against *T. gondii* (Luft and Remington, 1992). The parasite's resistance to this parasite is largely due to cellular immunity; the parasite's encystment is aided by the cellular immune response (Charles, 2009). *T. gondii*-infected cells can be lysed by both immunological CD4+ and CD8+ cells (Montoya and Remington, 1996). These T-cell subsets collaborate with NK cells, macrophages, and lymphokine-activated killer cells during defensive activities (Robinson *et al*., 2009). Cytokines are significant in the pathogenesis of toxoplasmosis and toxoplasmic encephalitis because they play an important role in infection defense (Bonfioli and Orefice, 2005).

Following sensitization with *T. gondii* the macrophages produce Interleukin - 12 (1L – 12), which together with Interleukin 13 (1L- 13), Interleukin 18 (1L- 18), and Tumor necrosis factor (TNF- α) stimulate NK cells to produce interferon Gamma IFN-γ. IFN- γ acts in synergy with the TNF- α, which induces the production of nitric oxide which kills intracellular *T. gondii* (Robinson *et al.,* 2009). As a result, cytokines that activate macrophage function, such as IFN- and TNF, are critical for limiting tachyzoites multiplication throughout both the acute and chronic phases of infection. During the chronic period, T-Lymphocytes are the main producers of these cytokines (David *et al.*, 2008).

1.8 Neurotransmitter

A neurotransmitter is signalling chemical released by a neuron and transmitted across a synapse to affect another cell, another neuron, a gland, or a muscle cell could be the cell that receives the signal from any main body component or target cell (Lodish *et al.*,2000). Synaptic vesicles release neurotransmitters into the synaptic cleft, where they can interact with neurotransmitter receptors on the target cell, the receptor to which the neurotransmitter binds determines the neurotransmitter's action on the target cell, many neurotransmitters are made from simple and abundant precursors like amino acids, which are easily available and only require a few biosynthetic steps, the function of complex neural systems is dependent on neurotransmitters, although the exact number of distinct neurotransmitters found in humans is unknown, more than 100 have been discovered (Cuevas 2019).

1.8.1 Dopamine

Dopamine is an organic molecule that belongs to the phenethylamine family and is also known as DA (3,4-dihydroxyphenethylamine). Over 80% of the catecholamines in the brain are made up of dopamine. Dopamine is also produced by most mammals and plants. Dopamine is a neurotransmitter, which is a chemical that neurons (nerve cells) emit to communicate with other neurons. The brain has several distinct dopamine pathways, one of which is crucial for the motivational aspect of behaviour that is driven by rewards. When a reward is expected, the majority of incentives increase dopamine levels in the brain (Berridge 2007). A catechol structure (a benzene ring with two hydroxyl side groups) is connected to one amine group via an ethyl chain in a dopamine molecule (Fig 1-6). As a result, dopamine is the most basic

catecholamine, a class of neurotransmitters that also includes norepinephrine and epinephrine (Macit *et al.*,2018).

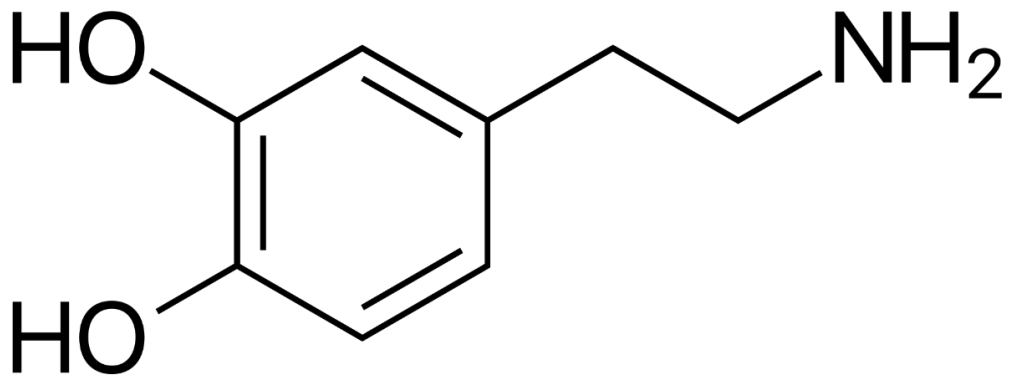


Figure (1-6) Dopamine structure (Macit *et al.*,2018)

Dopamine is produced by a small number of cell types, mostly neurons and cells in the adrenal glands' medulla (Seeman 2009). L-DOPA, dopamine's direct precursor, can be made either indirectly from phenylalanine, an essential amino acid, or directly from tyrosine, a non-essential amino acid (Musacchio 2013). The substantia nigra and ventral tegmental region are the largest and most important sources of dopamine in the vertebrate brain. In many ways, these structures are functionally similar and closely related. Both of these structures are found in the midbrain (Björklund and Dunnett 2007).

Dopamine represents the perceived importance of a motivational result (i.e., its desirability or aversiveness), which influences an organism's behaviour in favour of or against achieving that outcome (Wenzel *et al.*, 2015), (Puglisi-Allegra and Ventura 2012).

Dopamine has a lot of critical functions in the brain, including motor behaviour regulation, motivational rewards, and emotional arousal. It is

important in the reward system; low levels of dopamine have been connected to Parkinson's disease, and high amounts of dopamine have been linked to schizophrenia (Schacter *et al*., 2009).

Dopamine does not pass the blood-brain barrier outside of the nervous system, so its production and actions in the peripheral nervous system are largely independent of those in the brain (The National Collaborating Centre for Chronic Conditions, 2006). Dopamine circulates in the bloodstream in large amounts, although its exact functions are unknown (Eisenhofer *et al*., 2004).

1.9.2 Adrenaline

Adrenaline is a hormone and medicine that is also known as epinephrine (Lieberman *et al*., 2013). It plays a role in visceral function regulation (e.g. respiration) (Malenka *et al*., 2009). The adrenal glands and a small number of neurons in the medulla oblongata create adrenaline in the normal state. It aids in the fight-or-flight response by boosting blood flow to muscles and cardiac output via acting on the SA node (Brown *et al*.,1979), blood sugar level and pupil dilatation response (Bell, 2009; Khurana, 2008).

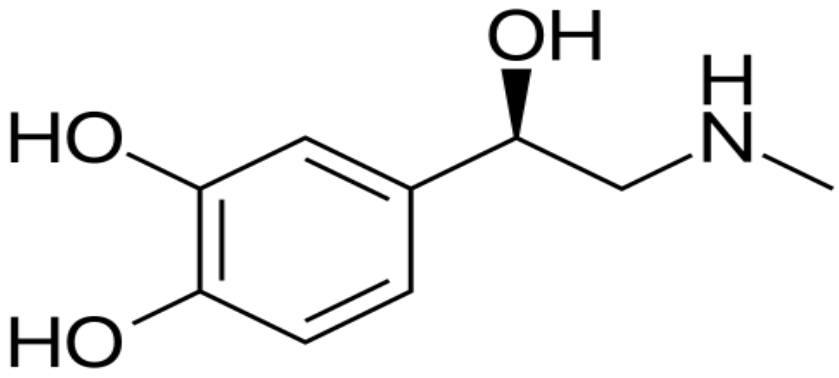


Figure (1-7) Adrenaline structure (Siddiqui *et al.*, 2015)

Adrenaline is produced via a metabolic pathway that transforms the amino acids phenylalanine and tyrosine into a sequence of metabolic intermediates and, finally, adrenaline in the chromaffin cells of the adrenal medulla and a limited number of neurons in the medulla oblongata of the brain (Lieberman *et al*., 2013; Malenka *et al.,* 2009). The rate-limiting phase is when tyrosine is oxidized to L-DOPA by tyrosine hydroxylase. DOPA decarboxylase then decarboxylates it to produce dopamine (aromatic L-amino acid decarboxylase).

Dopamine is then transformed into noradrenaline by an enzyme called dopamine beta-hydroxylase, which uses copper and ascorbic acid (vitamin C) to do so, the methylation of noradrenaline's main amine is the last step in adrenaline production, the enzyme phenylethanolamine N methyltransferase (PNMT), which uses S-adenosyl methionine (SAM) as the methyl donor, catalyzes this reaction (Verberne *et al*., 2016). While PNMT is mostly located in the cytosol of the adrenal medulla's endocrine cells, it has also been found in the heart and brain in low quantities (Verberne *et al*., 2016).

Adrenaline binds to all adrenergic receptors, including the main subtypes α1, α2, β1, β2, and β3 (Shen, 2008). The binding of adrenaline to these receptors causes a multitude of metabolic changes. Binding to -adrenergic receptors reduce pancreatic insulin output and increase glycogenolysis in the liver and muscle (Arnall *et al*.,1986), in muscle, it increases glycolysis while inhibiting insulin-mediated glycogenesis (Raz *et al.*,1991; Sircar 2007).

The binding of β adrenergic receptors causes the pancreas to secrete glucagon, the pituitary gland to secrete more adrenocorticotropic hormone (ACTH), and adipose tissue to secrete more lipolysis. These

interactions result in a rise in blood glucose and fatty acids, which provide energy-producing substrates for cells all across the body (Sircar 2007).

1.9.3 Neurotransmitter secretion disturbances due to *T. gondii* infection

According to the behavioural manipulation theory, parasites can manipulate host behaviour to benefit the parasites rather than the host (Adamo2013; Hughes 2013).

The parasitic protozoan a common example is *T. gondii*. *Toxoplasma* infection diminishes rodents' aversion to cat scents, potentially boosting predation by the felid host (Berdoy *et al.*,2000). *T. gondii* can make an infected mouse more aggressive and less afraid of the cat (the ultimate host) (Boillat *et al*., 2020).

There have been claims that *T. gondii* infection causes schizophrenia and a desire to commit crimes in people (Lindová *et al*., 2012). Although the parasite's acute infection is asymptomatic, the chronic stage of infection causes behavioural changes (Webster *et al*., 2013). The type and degree of the illness may be linked to the location of the brain damage, as well as the host's and parasite strain's genetic preparedness (Lindová *et al*., 2012). Type I, for example, cannot generate a cyst in the tissues or latent infection in the laboratory, but it can cause miscarriage, inflammatory effects, and possibly schizophrenia (Suzuki, 2012).

Parasites always attack the central nervous system, either by attacking neurons directly (Cabral *et al*., 2016) or by stimulating the immune

system to produce unique chemicals that alter behaviour. In any event, immunological manipulation, modification of neurotransmitters, or changes in the amounts of some essential hormones are all options for altering behaviour (Madlaina *et al*., 2020).

Infectious agents may play a role in some cases of schizophrenia, according to recent epidemiological studies. *T. gondii* infection can change behaviour and neurotransmitter activity in animals. acute *T. gondii* infection in humans can cause psychotic symptoms similar to those seen in people with schizophrenia (Torrey and Yolken 2003).

The parasite must finish its complex life cycle in the intestines of felines (cats) in order to reproduce (Montoya and Liesenfeld, 2004). Contrarily, non-feline mammals show cognitive and behavioural modifications that increase their vulnerability to catching by felines (Berdoy *et al.,* 2000).

They are believed to be survival-related evolutionary adaptations of the parasite. There is proof that the parasite alters the brain's neurotransmitters to bring about this behavioural shift (Parlog *et al.,* 2015). Infected neurons produce more dopamine, according to studies (Prandovszky *et al.,* 2011).

The parasite may have an impact on the glutamate signaling system through the kynurenine pathway (Notarangelo *et al*., 2014). Since the release of this data, scientists have been paying more attention to how a latent *T. gondii* infection in humans affects their cognition and behaviour. Uncertainty exists regarding the effects of the cognitive changes in latent *T. gondii* infection patients, including increased impulsivity (Peng *et al.*,2018).

However, meta-analyses revealed a significant association between *T. gondii* infection and a number of psychiatric disorders, including schizophrenia, suggesting that the infection may also have an impact on human behaviour (Sutterland *et al.*, 2015).

CHAPTER TWO

MATERIALS

AND

METHODS

2 Materials and Methods

2.1 Materials

2.1.1 Instruments

Instruments that have been used in this study were shown in Table (2-1).

Table (2-1) Instruments and equipment used in the present study

Instruments	Company	Origin
Autoclave	Biobase	China
Blood Cells Counter	Biobase	China
Centrifuge	Kokusan	Japan
Deep freeze	Biobase	China
ELISA Reader	Paramedical	Italy
ELISA washer	Paramedical	Italy
Incubator	Jrad	Syria

2.1.2 Tools

Tools that have been used in this study were shown in Table (2-2).

Table (2-2) Tools used in this study

Tools	Company	Origin
Cool box	Cool Ice Box Co.	United Kingdom
EDTA tubes	Beijing Hanbaihan Medical Co.	China
Microtiter plate Reader(450nm)	Bio Tek	USA
Eppendorf tubes	Beijing Hanbaihan Medical Co.	China
Jell tubes	Beijing Hanbaihan Medical Co.	China
Microtiter plate washer	Bio Tek	USA
Rack		England
Gloves	Jiangsu Yanfang Medical Technology	China
Micro Pipette tips	Bio Tek	USA
5 µL-, 100 µL- and 500 µL micro- and multichannel pipettes	Beijing Hanbaihan Medical Co.	China
Micropipettes and disposable Pipettes	Bio Tek	USA
Disposable tubes	Beijing Hanbaihan Medical Co.	China
Absorbent paper		China

2.1.3 Chemical Materials and Solutions

Chemical Materials and solutions that have been used in this study were shown in Table (2-3).

Table (2-3) Chemical Materials and Solution used in the present study

Chemical Materials and Solution	Company	Origin
Distilled Water	Saudi Pharmaceutical Industries	Kingdom of Saudi Arabia
Ethanol 75%	GRS	Iraq
Human Toxo IgM ELISA kit	Demeditec	Germany
Human Toxo IgG ELISA kit	Demeditec	Germany
Human Dopamine ELISA kit	Shanghai yehua biological technology	China
Human Epinephrine ELISA kit	Shanghai yehua biological technology	China

2.2 Methods

2.2.1 Subject Collection

This study was performed in Al- Amarah city South of Iraq among different groups of men and women (15-54 years), the samples were collected from Al-Shaheed Al-Sadder teaching hospital and the main blood bank in Al- Amarah city. The sample collection started from January 2020 till October 2021. The study was conducted on 174 venous blood samples collected from 21 females and 153 males. Detection of parasite antibodies and levels of dopamine and adrenaline was achieved by using Enzyme Linked Immunosorbent Assay (ELISA - IgM, ELISA- IgG, ELISA- Dopamine and ELISA- Adrenaline). A questionnaire sheet regarding the information about the patient was filled out for each one.

2.2.2 Study population

The study population consists of 174 participants, of whom 21 are females and 153 are males (Fig.2-1).

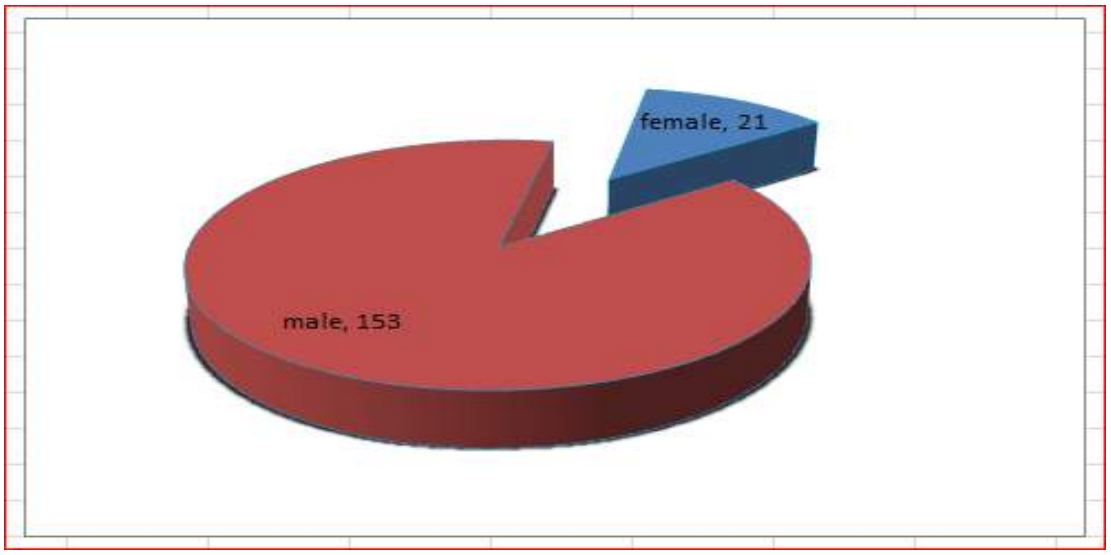
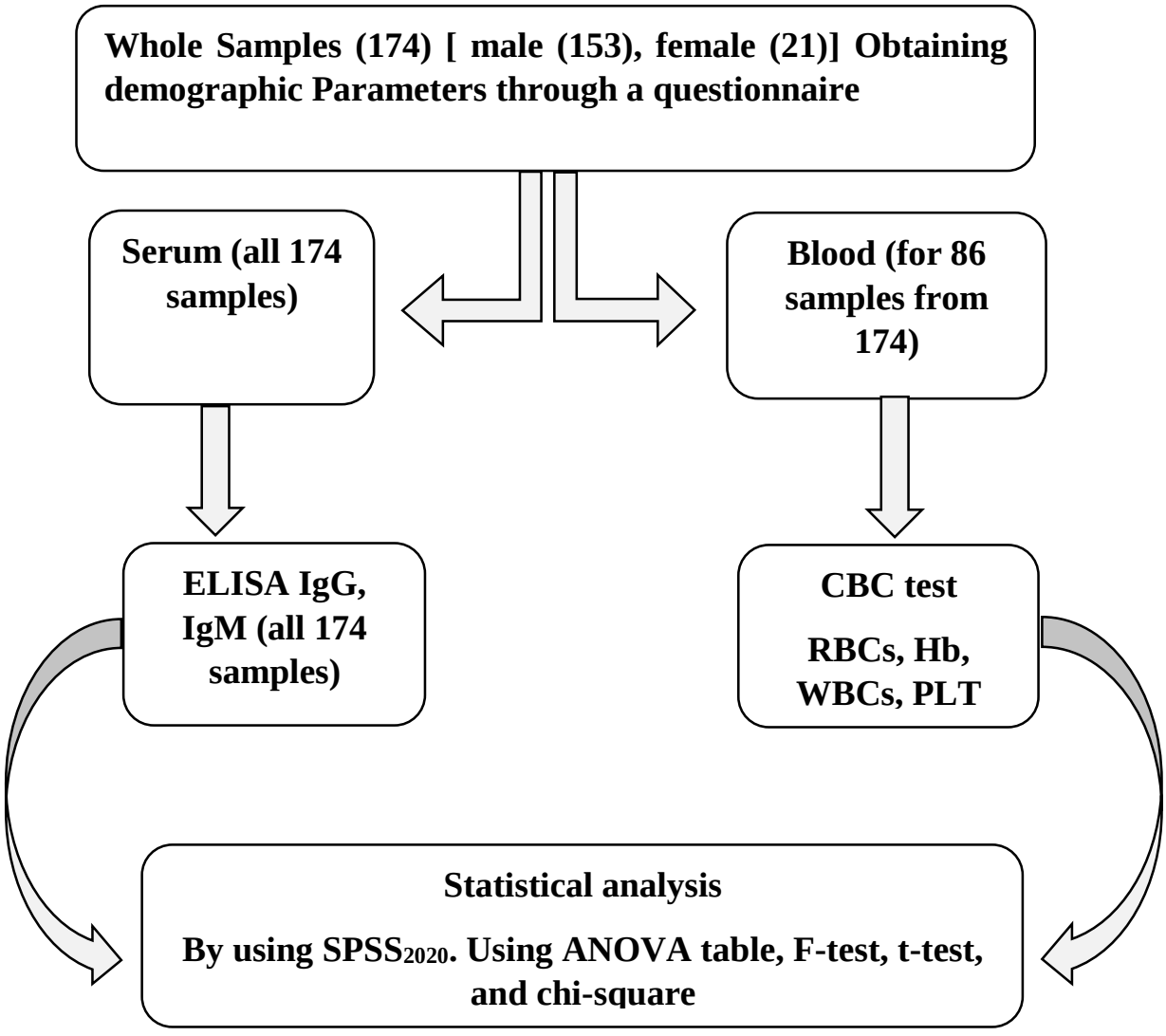


Fig. 2-1: The distribution of the study population for the gender.

2.2.3 Blood Sample Collection

Five ml of venous blood was collected from each patient. 2.5 ml of blood was placed in EDTA tubes for performing a Complete Blood Count (CBC) (only 86 of the total samples were CBC tested). while the remaining 2.5 ml of blood was placed in gel tubes and left to stand for 20 minutes at room temperature to clot, then placed in a centrifuge at 3000rpm for 10 minutes to collect the serum. Then the serum was placed in the Eppendorf tubes and kept in deep freeze at (-20℃) until the ELISA assay was performed.



2.3 Laboratory Methods

2.3.1 ELISA-IgM

The enzyme immunoassay (EIA) is the foundation of the Demeditec *Toxoplasma* IgM antibody test kit. The surface of the microtiter plates is coated with the *T. gondii* antigen. The wells of the microtiter plate are diluted with patient serum by micropipette. Serum IgM antibodies bind to the *Toxoplasma gondii* antigen that has been immobilized. To get rid of unbound material, the plate is rinsed with diluted wash solution after an hour-long incubation at room temperature. After that, a ready-to-use anti-human IgM peroxidase conjugate is added, and the reaction is allowed to sit for 30 minutes. Following a second washing procedure, the substrate tetramethylbenzidine (TMB) solutions are put into the wells where it is incubated for 20 minutes to cause the produce of a blue dye. The addition of a stop solution, which transforms the color from blue to yellow, stops the color development process. At a wavelength of 450 nm, the resultant dye is spectrophotometrically measured. The relationship between the IgM antibody concentration and color intensity is direct (Montoya, 2002), (Remington *et al*., 2004).

2.3.1.1 Reagents

The Demeditec *Toxoplasma* IgM antibody test set contains the materials listed in Table (2-4).

Table (2-4) The reagents of ELISA IgM

Chemical Materials and Solution	Ingredients	Volume/ number
Microtiter Strips	It was coated with a *T. gondii* antigen (strain RH, isolated from infected mice)	12
Calibrator A	Human serum diluted with PBS with 1 IU/mL of IgM antibodies against *T. gondii* Addition of 0.01 % methylisothiazolone and 0.01 % bromonitrodioxane	2mL
Calibrator B	Human serum diluted with PBS with 10 IU/mL of IgM antibodies against *T. gondii*. Addition of 0.01 % methylisothiazolone and 0.01 % bromonitrodioxane	2mL
Calibrator C	Human serum diluted with PBS with 30 IU/mL of IgM antibodies against *T. gondii* Addition of 0.01 % methylisothiazolone and 0.01 % bromonitrodioxane	2mL
Calibrator D	Human serum diluted with PBS with 120 IU/mL of IgM antibodies against *T. gondii* Addition of 0.01 % methylisothiazolone and 0.01 % bromonitrodioxane	2mL
Enzyme Conjugate	Anti-human -IgM-HRP (rabbit), in a protein-containing buffer solution. Addition of 0.01 % methylisothiazolone, 0.01 % bromonitrodioxane and 5 mg/L Proclin	15mL
Substrate	TMB (tetramethylbenzidine)	15mL
Stop Solution	0.5 M sulfuric acid	15mL
Sample Diluent	PBS/BSA buffer. Addition of 0.095 % sodium azide	60 mL
Washing Buffer	PBS + Tween 20, 10x concentrate	60 mL

2.3.1.2 Assay Procedure

All reagents and sera were left at (18-25 C^0) for 30 minutes before carrying out the assay, for which the manufacturer's instructions were followed. After that, conducted the following steps:

Assay Steps:

1- In order to prepare enough microtiter wells for the calibrators, samples, and substrate blank, take about 100µLof each of the diluted (1:101) samples and the ready-to-use calibrators into the wells. For the substrate blank, one well was left empty. For 60 minutes, the plate was incubated at room temperature with the reusable plate cover on, 300 µL of diluted washing solutions were added after had emptied the wells on the plate. three times in a row Remove the washing buffer, use tissue paper as a surface, and gently hit the microtiter plate.
2- 100 L of each ready-to-use conjugate was poured into the wells. For the substrate blank, one well was left unfilled. After covering the plate, allow it to incubate at room temperature for 30 minutes. Then poured 300µL of diluted washing solution after emptying the plate's wells three times. used tissue paper as a surface. To remove the washing buffer well, tap the microtiter plate.
3- The ready-to-use substrate was pipetted into each well in a quantity of 100µL. This time, the substrate blank is pipetted as well. For 20 minutes, keep the plate covered and incubate it at room temperature in the dark. pipetted 100µLof each of the ready-to-use stop solutions into the wells in order to stop the substrate reaction. Additionally, conducted a reading of the

absorbance at 450 nm after thoroughly mixing and wiping the plate's bottom, (Montoya, 2002), (Remington *et al*., 2004).

2.3.1.3 Qualitative Evaluation

The computed serum absorptions for the patients are contrasted with the calibrator's cutoff value (10 IU/mL). Positive results are obtained if the sample value is higher. There is a negative outcome for values below the cut-off calibrator. It has been determined as a "grey zone" as being within a +/-20% range around the cut-off number.

2.3.2.4 Quantitative Evaluation

The calibrators' absorptions are compared graphically to their concentrations for a quantitative evaluation. The concentration values for each patient sample can then be recovered in relation to their absorptions from the resulting calibration curve, and the serum level IgM was estimated using the standard curve, as shown in Fig (2-2).

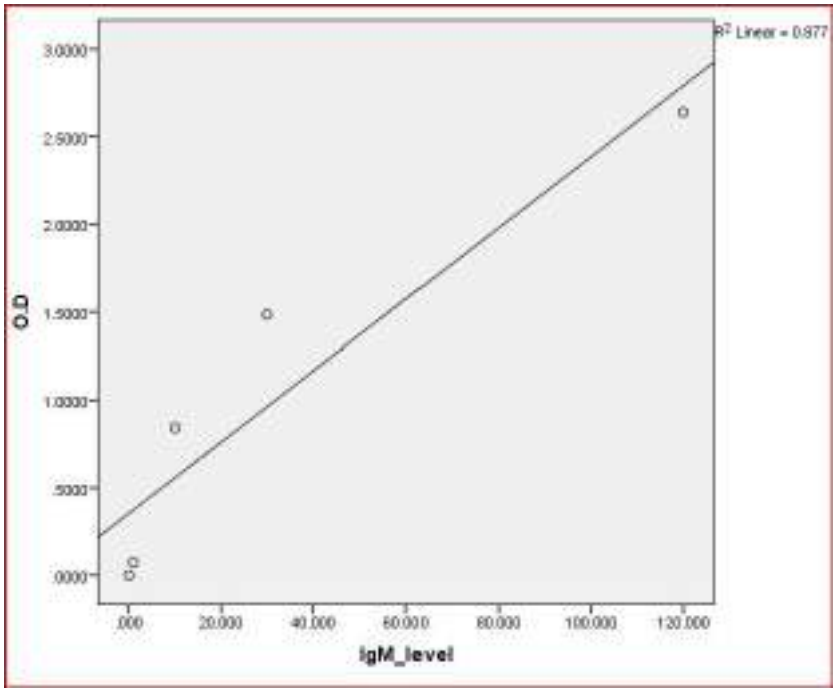


Figure (2-2) The standard curve of IgM

2.3.2 ELISA-IgG

This kit is based on the same principle as that on which ELISA IgM is based (see Page35).

2.3.2.1 Reagents

The Demeditec *Toxoplasma* IgG antibody test set contains the materials listed in Table (2-5).

2.3.2.2 Assay Procedure

We followed the same steps that we followed when conducting the ELISA IgM (see Page 37).

2.3.2.3 Qualitative Evaluation

The results are calculated as in Qualitative Evaluation of the ELISA IgM (see page 38).

2.3.2.4 Quantitative Evaluation

The calibrators' absorptions are compared graphically to their concentrations for a quantitative evaluation. The concentration values for each patient sample can then be recovered in relation to their absorptions from the resulting calibration curve, and the serum level of IgG was estimated using the standard curve, as shown in Fig (2-3).

Table (2-5) The reagents of ELISA IgG

Chemical Materials and Solutions	**Ingredients**	**Volume/ number**
Microtiter Strips	It was coated with a *T. gondii* antigen (strain RH, isolated from infected mice)	12
Calibrator A	Human serum diluted with PBS with 0IU/mL of IgG antibodies against *T. gondii* Addition of 0.01 % methylisothiazolone and 0.01 % bromonitrodioxane	2mL
Calibrator B	Human serum diluted with PBS with 10 IU/mL of IgG antibodies against *Toxoplasma gondii*. Addition of 0.01 % methylisothiazolone and 0.01 % bromonitrodioxane	2mL
Calibrator C	Human serum diluted with PBS with 40 IU/mL of IgG antibodies against *T. gondii* Addition of 0.01 % methylisothiazolone and 0.01 % bromonitrodioxane	2mL
Calibrator D	Human serum diluted with PBS with 100 IU/mL of IgG antibodies against *T. gondii* Addition of 0.01 % methylisothiazolone and 0.01 % bromonitrodioxane	2mL
Calibrator E	Human serum was diluted with PBS with 250 IU/mL of IgG antibodies against *T. gondii.* Addition of 0.01 % methylisothiazolone and 0.01 % bromonitrodioxane	2mL
Enzyme Conjugate	Anti-human -IgG-HRP (rabbit), in a protein-containing buffer solution. Addition of 0.01 % methylisothiazolone, 0.01 % bromonitrodioxane and 5 mg/L Proclin	15mL
Substrate	TMB (tetramethylbenzidine)	15mL
Stop Solution	0.5 M sulfuric acid	15mL
Sample Diluent	PBS/BSA buffer. Addition of 0.095 % sodium azide	60 mL
Washing Buffer	PBS + Tween 20, 10x concentrate	60 mL

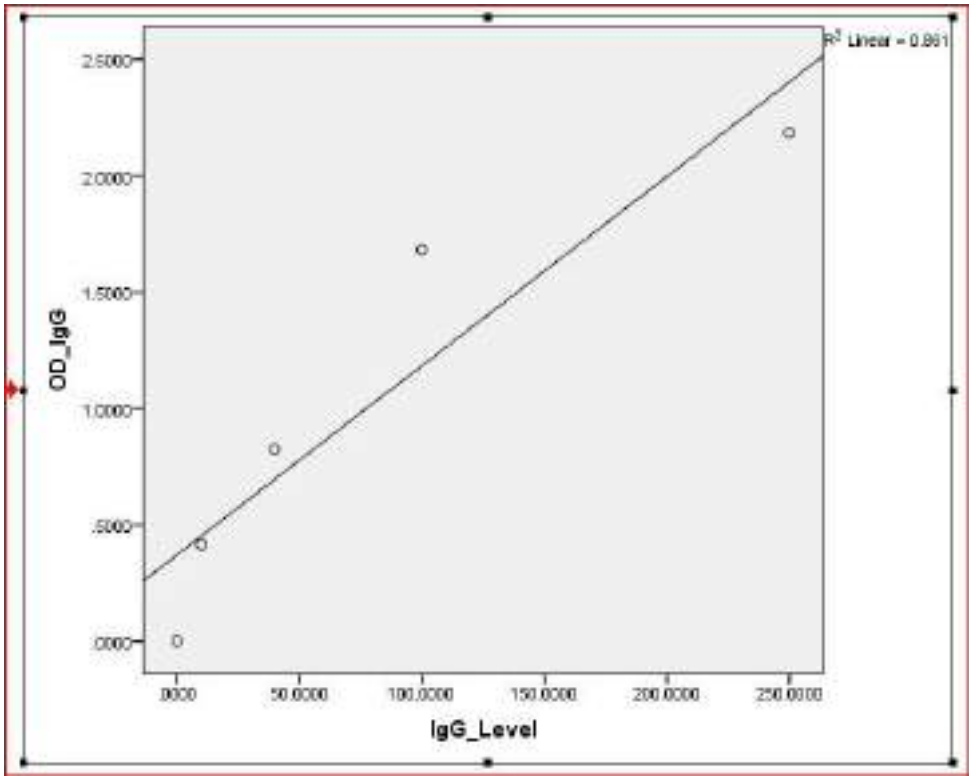


Figure (2-3) The standard curve of IgG Level Estimation against optical density (OD).

2.3.3 ELISA-Dopamine

This kit uses enzyme-linked immune sorbent assay (ELISA) based on biotin double antibody sandwich technology to assay Human Dopamine(DA).

2.3.3.1 Reagents

- Coated ELISA plate 12-well * 8 tubes
- Standard solution(960ng/L): 0.5ml×1
- Streptavidin-HRP: 6ml×1
- Stop Solution: 6ml×1
- chromogenic reagent A: 6ml×1

- chromogenic reagent B: 6ml×1
- Anti DA antibodies labelled with biotin: 1ml×1
- Standard dilution: 3ml
- Washing concentrate: (20ml×30) ×1

2.3.3.2 Assay procedure

Conducted the following steps

1- Diluted the standard solutions using small tubes according to Table (2-6).
2- Adding chromogen reagents, A & B and stop solution to the blank well and adding 50µl standard and streptomycin-HRP 50µl to the standard solution well, while in the sample well that is to be tested, adding 40µl sample and then 10µl DA antibodies, 50µl streptavidin-HRP. Then cover it with a sealed plate membrane, shake gently to mix, and incubate it at 37°C for 60 minutes.
3- Diluting the washing concentration (30X) with distilled water After that, carefully remove the seal plate membrane, drain the liquid, and shake off the remainder. filling each well with washing solution, leaving it for 30 seconds, and then draining it. repeating this procedure five times and then blotting the plate.
4- Each well first received 50µl of chromogen reagent A, and subsequently, each well received 50µl of chromogen reagent B. to blend, lightly shaking. for the formation of color, incubated for 10 minutes at 37°C without light.
5- To stop the reaction, add 50µl of the Stop Solution, and the color change from blue to yellow.

6- After adding the stop solution (within 10 minutes), take the blank well as zero, and measure the absorbance (DA) by putting the microtiter plates in the ELISA reader apparatus at 450 nm wavelength (Montoya, 2002), (Remington *et al*., 2004).

2.3.3.3 Estimated serum dopamine (DA) levels

The standard curve was plotted by the DA value versus the known level of samples of sera DA (included with the kit) Then OD values that were read by the ELISA reader were converted to a DA value by the standard curve Fig (2-4).

Table (2-6) Dilution of standard solutions (ELISA DA)

480ng/L	Standard No.5	120µl Original Standard + 120µl Standard diluents
240ng/L	Standard No.4	120µl Standard No.5 + 120µl Standard diluents
120ng/L	Standard No.3	120µl Standard No.4 + 120µl Standard diluent
60ng/L	Standard No.2	120µl Standard No.3 + 120µl Standard diluent
30ng/L	Standard No.1	120µl Standard No.2 + 120µl Standard diluent

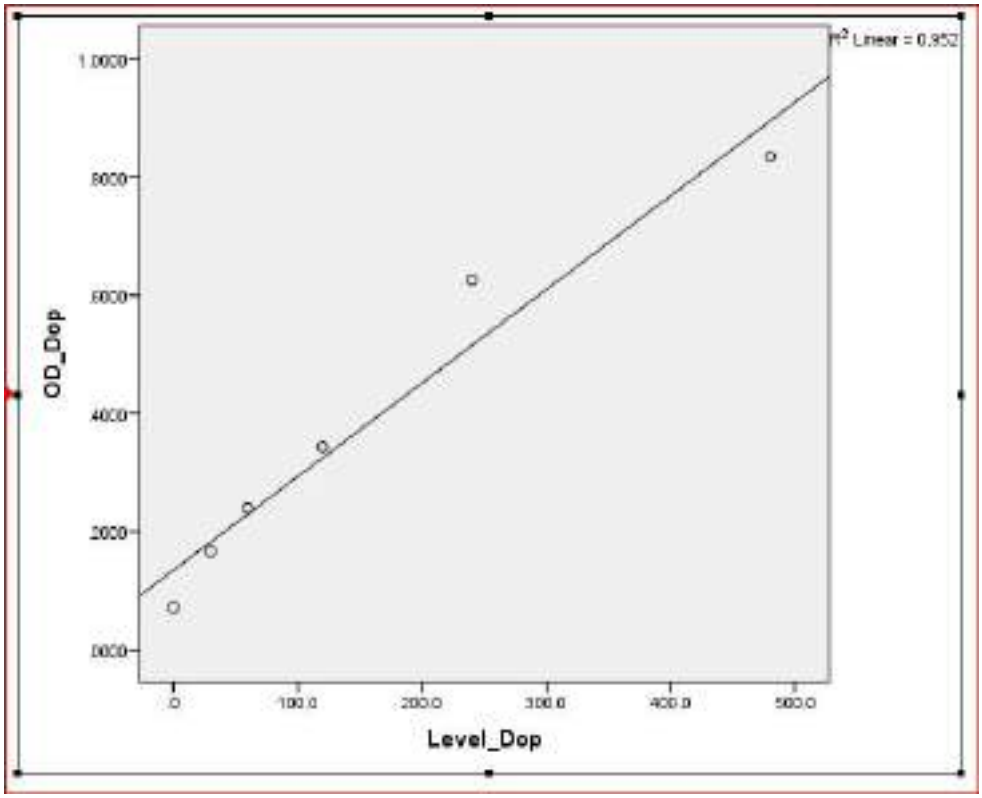


Figure (2-4) The standard curve of dopamine

2.3.4 ELISA-Adrenaline

This kit uses enzyme-linked immune sorbent assay (ELISA) based on biotin double antibody sandwich technology to assay Human Epinephrine(EPI).

2.3.4.1 Reagents

- Coated ELISA plate: 12-well * 8 tubes
- Standard solution: (1200ng/L) 0.5ml×1
- Streptavidin-HRP: 6ml×1
- Stop Solution: 6ml×1
- chromogenic reagent A: 6ml×1
- chromogenic reagent B: 6ml×1

- Anti EPI antibodies labeled with biotin: 1ml×1
- Standard dilution: 3ml×1
- Washing concentrate: (20ml×30) ×1

2.3.4.2 Assay procedure

1- Diluted the standard solutions using small tubes according to Table (2-7).
2- Adding chromogen reagents, A & B and stop solution to the blank well, and adding 50µl standard and streptomycin-HRP 50µl to the standard solution well, while in the sample well that is to be tested, adding 40µl sample and then 10µl EPI antibodies, 50µl streptavidin-HRP. Then cover it with a sealed plate membrane, shake gently to mix, and incubate it at 37°C for 60 minutes.
3- Diluting the washing concentration (30X) with distilled water After that, carefully remove the seal plate membrane, drain the liquid, and shake off the remainder. filling each well with washing solution, leaving it for 30 seconds, and then draining it. repeating this procedure five times and then blotting the plate.
4- Each well first received 50µl of chromogen reagent A, and subsequently, each well received 50µl of chromogen reagent B. to blend, lightly shaking. for the formation of color, incubated for 10 minutes at 37°C without light.
5- To stop the reaction, add 50µl of the Stop Solution, and the color change from blue to yellow.
6- After adding the stop solution (within 10 minutes), take the blank well as zero, and measure the absorbance (DA) by putting the

microtiter plates in the ELISA reader apparatus at 450 nm wavelength (Montoya, 2002), (Remington *et al*., 2004).

Table (2-7) Dilution of standard solutions (ELISA AD)

600ng/L	Standard No.5	120µl Original Standard + 120µl Standard diluents
300ng/L	Standard No.4	120µl Standard No.5 + 120µl Standard diluent
150ng/L	Standard No.3	120µl Standard No.5 + 120µl Standard diluent
75ng/L	Standard No.2	120µl Standard No.3 + 120µl Standard diluent
37.5ng/L	Standard No.1	120µl Standard No.2 + 120µl Standard diluent

2.3.4.3 Estimated serum adrenaline (AD) levels

The standard curve was plotted by the AD value versus the known level of samples of sera AD (included with the kit) Then OD values that were read by the ELISA reader were converted to an AD value by the standard curve Fig (2-5).

2.4. Statistical analysis

The results were statistically analyzed after extracting their mean and standard deviation using $SPSS_{2020}$ and using the analysis of variance table, ANOVA table, F-test and t-test, and significant differences were determined at ($p<0.05$) (Al-Baldawi, 2009).

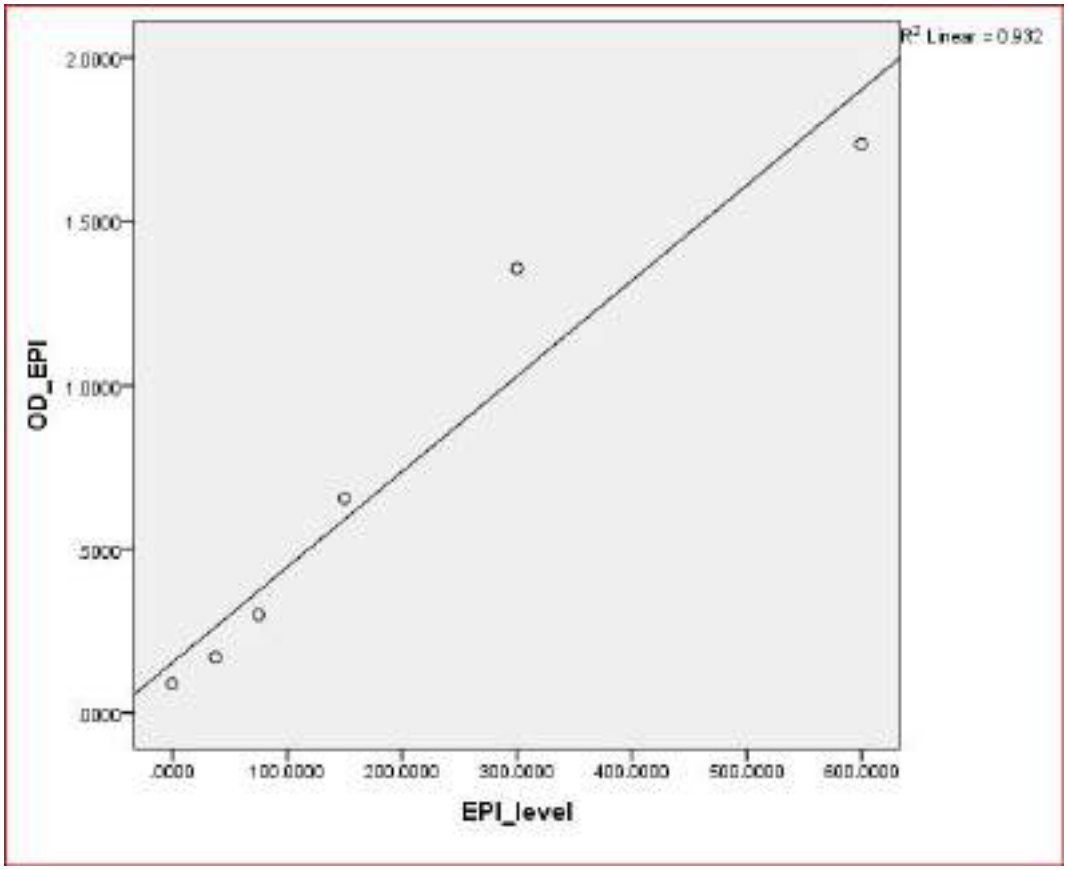


Figure (2-5) The standard curve of adrenaline level estimation against optical density (OD)

CHAPTER THREE

RESULTS

AND

DISCUSSION

3 Results and discussion:

3.1 The infection rates of participants' blood samples examination by ELISA

3.1.1 Total infection rates

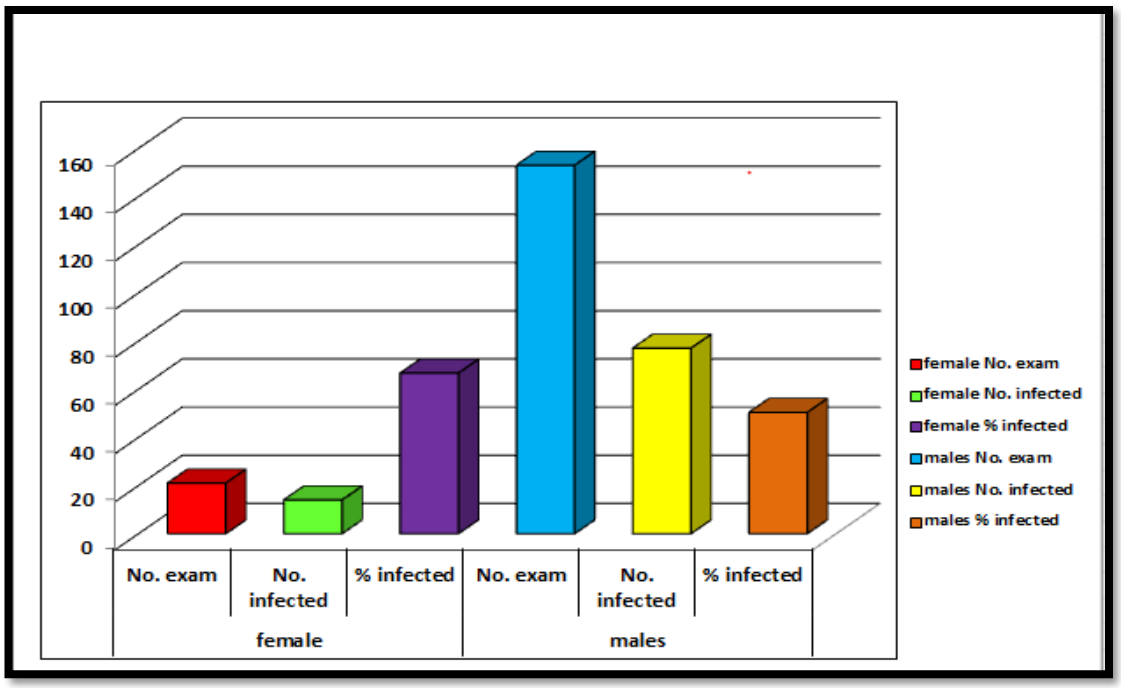
The total infection rate was 52.3% (91/174). These results are in line with the global infection rate of toxoplasmosis, which ranges from 30% to 50% (Flegr *et al.,* 2014). It is less than in Egypt at 59.6% (Elsheikha *et al.,* 2009), and 83.1% in Iran as recorded by Almasian *et al.* (2014). While it was higher than what was recorded in some Province like Basrah, 41.1-52.1% (Yacoub *et al.,* 2006) while 38.7% in Qadisiyah (Hadi *et al*., 2016), 12.4% recorded by Al-Sadoon *et al.* (2018), 22.98% in Dohuk (Husein and Balatay, 2019), 17.80% in Wasit (Al-Sray *et al.*, 2019), and Misan 20% (Alsaady *et al*., 2021).

In other countries like 0.8% in South Korea (Song *et al*., 2005), 9.1% in England (Nash *et al.,* 2005), United States of America 3.7% (Han *et al.,* 2008) and 0.2% in Canada by Bigna *et al.* (2020).

It is obvious that diverse results might be obtained within the same country when it comes to toxoplasmosis seroprevalence. This could be a result of the influence of a number of factors, including the person's health, age, nutrition habits, consumption of raw or undercooked meat, fruits and vegetables, hygiene practices, home ownership of cats, contact with cats, type of occupation, characteristics of the immediate environment, and geographical and climatic conditions. Wilking *et al.*, 2016; Alsaady *et al*., 2021), and showed that the hot-wet regions have a greater oocyst sporulation rate (Flegr and Kaňková, 2020).

3.1.2 The relationship between gender and Toxoplasmosis

The ELISA examination of 21 female sera detected the infection rate is 66.67% (14/21) and among males is about 50.33% (77/153). There are statistically significant differences between the two sexes in infection of toxoplasmosis (t=4.571, p=0.000017) (Fig.3-2).



(t=4.571, p=0.000017)

Fig. 3-1: The relationship between gender and toxoplasmosis.

This finding agreed with the study of Alsaady *et al*. (2021), who found in other regions of the same Province of Misan that women had a greater infection rate (24.083%) than men (4.545%), with statistically significant differences between them. And it agreed with the findings of the study by Coelho *et a*l. (2003) in Brazil, and it was not consistent with a study conducted in southern Iran (Sarkari *et al*., 2014).

3.1.3: The distribution of immunoglobulin among infected individuals

Figure3-3, Table3-1 show that IgM is found alone in about 32.97% (30/91) of all toxoplasmosis-positive sera, whereas the total of IgM was observed to be diagnosed in about 61.54% (56/91) of them. In contrast, IgG was found alone in about 38.46% (35/91) of all toxoplasmosis-positive sera, and the total IgG formed in about 67.03% (61/91) of them. On the other hand, it was found that 28.57% (26/91) of all positive sera are positive for both IgM and IgG.

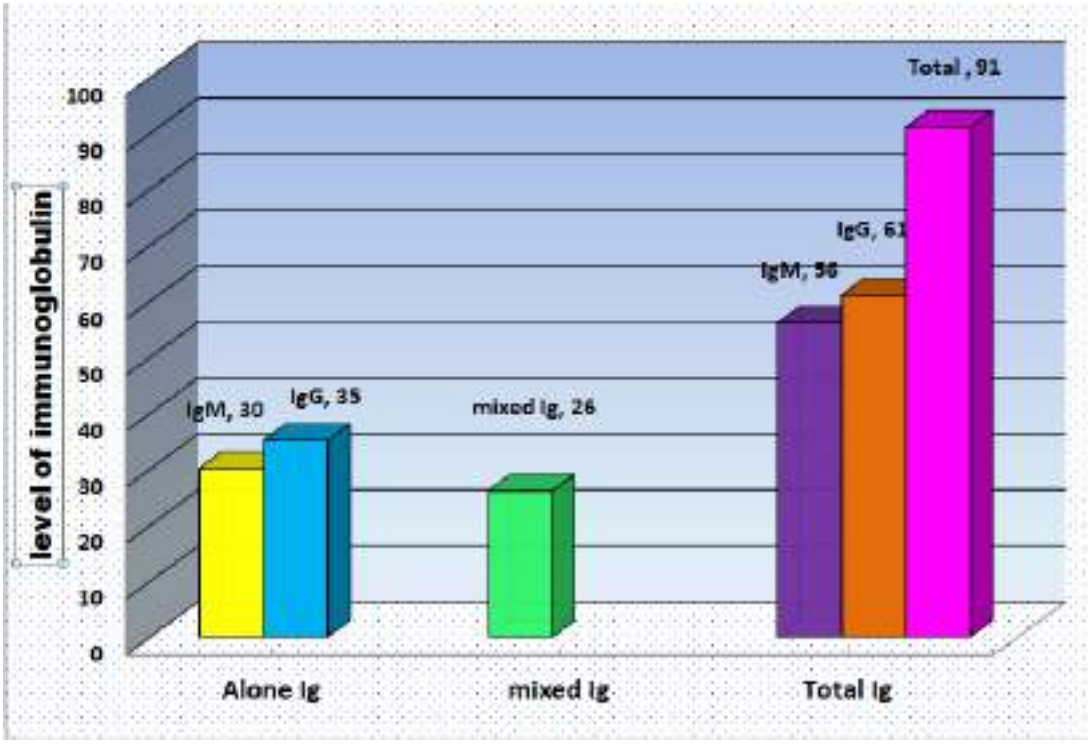


Figure 3-2: The distribution of immunoglobulins among toxoplasmosis-positive sera

Table3-1: The percentage of IgM and IgG distribution among Toxoplasmosis infected individuals

Immunoglobulin	Alone (%)	Mixed (%)	Total (%)
IgM	30 (32.97)	26 (28.57)	56 (61.54)
IgG	35 (38.46)		61 (67.03)
Total sera	65 (71.43)	26 (28.57)	91 (100)

These results are higher than the results of the study in Thi-Qar Province, where they reached 35.3% and 2.7% for IgG and IgM respectively (Al-Aboody, 2015), in Diyala IgM+ 4%, IgG+38% IgG+ and 2% both IgM+ and IgG+ (Darweesh *et al.*,2018).

3.1.4 The relationship between Toxoplasmosis and the levels of the immunoglobulins

The results recorded that significant difference ($p< 0.05$) between the levels of anti-*Toxoplasma* IgM of infected and uninfected individuals (Table 3-2).

Table 3-2: The relationship between the *T. gondii* infection and the level of the IgM.

	IgM level IU/mL ± SD	t- test	*p*<
Infected	29.667 ±16.591	10.869	0.000001
Uninfected	6.429 ± 1.838		

The mean IgG levels for infected individuals are 75.64 IU/mL and 31.18 IU/mL for uninfected individuals, with statistically significant differences in IgG levels between infected and uninfected individuals (Table 3-3).

Table 3-3: The relationship between the *T. gondii* infection and the level of the IgG.

	IgG level IU/mL	**t- test**	***p***
Infected	75.644 ± 94.614	2.561	0.012
Uninfected	31.187 ± 62.442		

± SD

In the acute stage of T. gondii infection, IgM antibodies estimate appear early in the course of infection while IgG antibodies appear about three weeks after appearance IgM and reach peak level within six months to one year and remain in a high level for long periods of time (Gras *et al*., 2004). IgG antibodies against T. gondii, on the other hand, show that T. gondii cysts were already present in the tissue when they were found in the blood (Dupon *et al*., 1995).

3.1.5 The relation between gender and immunoglobulins level and toxoplasmosis

This study shows the means of levels of IgM 97.486 IU/mL and IgG 33.976 IU/mL of infected males are higher than those of infected females (IgM, 26.282 IU/mL and IgG, 58.482 IU/mL), and these levels of IgM and IgG for each gender are higher than that of uninfected males and females (see Table 3-4), but there are no significant differences

between the interaction of genders, result of immunoassay on the levels of each IgM (F= 3.223, p= .076) and IgG (F= 0.954, p= 0.332).

Table 3-4: The levels of IgM and IgG of infected and uninfected males and females

	Infection	Examined No.	IgM level IU/mL	IgG level IU/mL
Male	Infected	11	33.976 ± 19.657	97.486 ± 101.013
	Uninfected	53	6.284 ± 1.898	31.113 ± 60.010
	Total	64	11.043 ± 13.236	42.521 ± 72.312
Female	Infected	14	26.282 ± 13.524	58.482 ± 89.200
	Uninfected	8	7.392 ± 0.979	31.674 ± 81.659
	Total	22	19.413 ± 14.144	48.734 ± 85.572
Total	Infected	25	29.667 ± 16.591	75.644 ± 94.614
	Uninfected	61	6.429 ± 1.838	31.187 ± 62.442
	Total	86	13.185 ± 13.884	44.110 ± 75.446

IgM (F= 3.223, *p*= 0.076) and IgG (F= 0.954, *p*= 0.332), ± SD

Gender does not seem to affect seroprevalence in the areas studied and both sexes thus have an equal chance of exposure to infection.

3.1.6 Association between toxoplasmosis and the levels of dopamine:

The present study (Table 3-5) finds that the mean level of dopamine in toxoplasmosis seropositive individuals is 29.543 ng/L and in seronegative individuals is 18.086 ng/L. It shows the serum level of

dopamine in toxoplasmosis-infected individuals is higher by about 163.347% (29.543/18.086) than that of uninfected individuals. There are significant differences in the level of dopamine between toxoplasmosis seropositive and seronegative individuals (Table 3-5).

Table 3-5: The relation between dopamine level (DA) and toxoplasmosis with respect to gender.

Gender	Result of immunoassay (R1)	Examined No.	DA Mean ng/L
Male	Infected	7	23.920
	Uninfected	47	18.998
	Total	54	19.636
Female	Infected	10	33.479
	Uninfected	8	12.725
	Total	18	24.255
Total(1)	Infected	17	29.543
	Uninfected	55	18.086
	Total	72	20.791

DA x IR (F= 5.942, p=0.017); (RI * gender) on DA (F=2.260, p= 0. 137) (P ≤ 0.05)

This percentage is higher than what was mentioned in some previous studies in mice, where the percentage of dopamine increased to about 114% in infected mice with toxoplasmosis (Prandovszky *et al.*, 2011). This increase in the level of dopamine in toxoplasmosis-infected cases has been shown in more than one study such as Stibbs (1985), McConkey *et al*. (2013), AL-Hadad *et al*. (2019), Ibrahim *et al*. (2020),

Mirzaeipour *et al*. (2021), Omidian *et al.* (2022). Dopamine performs many large functions in the brain (Best *et al*., 2009). Dopamine is formed in DA-ergic nerve cells from L-tyrosine with help of two enzymes, tyrosine hydroxylase and aromatic L-amino acid decarboxylase. The effect of *T. gondii* on dopamine levels may be coming by its promoting the synthesis of a neurotransmitter in the brain, it may be happing by altering the transmission of neurotransmitter signals through overexpressing an important gene due to its ability to synthesize the signaling molecule L-DOPA (3,4-dihydroxy-L-phenylalanine) (Gaskell *et al*., 2009), which is considered the root of dopamine (Wang *et al*., 2014), which leads to neurodegenerative diseases, and this explains by use of dopamine antagonists in psychiatric patients with toxoplasmosis reduces behavioral changes in patients (Prandovszky *et al*., 2011).

3.1.7 Association between Toxoplasmosis and levels of adrenaline.

The results of this study (Table3-6) show that the mean level of blood adrenaline in toxoplasmosis infected individuals is 28.75 ng/L, while its level in uninfected is 13.65 ng/L. The increase in the level of adrenaline is also shown in the study of Al-Hadad *et al.* (2019) in the Al-Najaf province. Although the concentration of adrenaline in infected cases is more than twice that in uninfected, there are no significant differences between its level in the blood of infected and uninfected cases ($t = 1.919$, $p = 0.058$). On the other hand, it was shown that the level of adrenaline in the serum of toxoplasmosis infected males or females had more than twice that of uninfected (Table 3-6). And there

is no statistically significant in the effect of the interaction between toxoplasmosis and gender on the adrenaline level (F=0.003, p= 0.958).

Table 3-6: The Relationship between Toxoplasmosis and AD level according to gender.

	Infection	**Examined No.**	**Mean ng/L**
Male	Infected	11	28.114
	Uninfected	53	13.649
	Total	64	16.135
Female	Infected	14	29.253
	Uninfected	8	13.809
	Total	22	23.637
Total	Infected	25	28.752
	Uninfected	61	13.670
	Total	86	18.054

(AD x Gender, t=0.902, p>0.05) (AD x gender-toxoplasmosis, F=0.003, p= 0.958).

The increase in the levels of adrenaline may be due to the effect of toxoplasmosis on neurons in the brain or on the cells of the adrenal glands (Kadhim and Al-Awadi, 2013; Al-Hadad *et al*., 2019), which are responsible for secreting the adrenaline in human body (Cosentino and Marino, 2012), or may be from converting dopamine to Norepinephrine and then adrenaline (Wang *et al*., 2014).

The statistical analysis showed a strong significant relationship between the level of dopamine and the levels of IgG ($p<0.000001$) and IgG ($p<0.000001$), as well as between the level of adrenalin and levels of IgG ($p<0.000001$) and IgM ($p<0.00005$), as shown in Table (3-7).

Table 3-7: The parameters between levels of IgG and IgM with levels of dopamine and adrenaline

The relation	**F**	***p***
DA level* IgG level	258.921	p<0.000001
DA level* IgM level	258.921	p<0.000001
AD level* IgG level	436.435	p<0.000001
AD level* IgM level	108.080	p<0.00005

Also found that there is a strong statistically significant effect of toxoplasmosis on the levels of the IgG-AD, IgM-AD, IgG-DA, and IgM-AD, as in Table (3-8).

Table 3-8: The effect of Toxoplasmosis on the levels of the IgG-AD, IgM-AD, IgG-DA, and IgM-AD

	Immunoglobulin	Hormone	X^2	*p*
Infected	**IgG**	**DA**	426.622	0.00006
		AD	495.900	0.000056
	IgM	**DA**	464.000	0.000055
		AD	526.833	0.000231
Uninfected	**IgG**	**DA**	1015.000	0.254
		AD	910.000	0.265
	IgM	**DA**	980.000	0.296
		AD	892.500	0.201

3.3 Complete Blood Count (CBC) test

3.3.1 The effect of Toxoplasmosis on the Hemoglobin level

The results (Table 3-9) showed that 44% (11/25) of toxoplasmosis-infected individuals had hemoglobin ranging between 11 and 13 g/dl compared with only 8.19% of the uninfected. In contrast, the hemoglobin in about 81.97% (50/61) of uninfected individuals ranged between 13-17g/dl compared with 40% (10/25) in the infected. There is a significant correlation between toxoplasmosis and the levels of hemoglobin in infected and uninfected individuals (χ^2=22.55, p = 0.000476).

Table 3-9 Relationship between toxoplasmosis and hemoglobin level

		hemoglobin levels (g/dl.)						
		≤9.0	9.1-11.0	11.1-13.0	13.1-15.0	15.1-17.0	17.1-19.0	total
No.	Infected	1	3	11	6	4	0	25
	Uninfected	0	3	5	20	30	3	61
Total		1	6	16	26	34	3	86

X^2=22.55, *p*=0.000476

It is noted that toxoplasmosis has a clear effect on hemoglobin levels in humans. These results are in agreement with the results of Abdul Abbas *et al*. (2015) in Al-Najaf, Mhamood (2016) in Tikrit city, Iraq; and Mahdi *et al.* (2020) in Thi-Qar, but are not in agreement with Hassen *et al*. (2019) in El-Beida, Libya, Mohamed (2020) in Makkah, Saudi Arabia; and Agordzo *et al*. (2020) in Ashanti, Ghana, who found no significant association between hemoglobin levels and toxoplasmosis. This decrease in hemoglobin may occur as a result of the decrease in the level of blood iron due to the infection with this parasite, and this leads to anemia in toxoplasmosis-infected individuals (Javadi *et al*., 2010).

3.3.2. Effect of *Toxoplasma gondii* infection on Red Blood Corpuscles (RBC) count.

The CBC test (Table 3-10) shows, that the mean count of RBC in infected individuals is 4.830 x 10^6/mm^3 compared with 5.144 x 10^6/mm^3 in the blood of uninfected individuals.

Table 3-10: The relationship between toxoplasmosis infection and count of red blood corpuscles (x10^6/mm^3).

	Mean of RBC x10^6/mm^3	**t-test**	***p***
Infected	4.830 ± 0.608	-2.504	0.014
Uninfected	5.144 ± 0.509		

$P \leq 0.05$, ±SD

There are statistically significant differences between the count of RBCs of toxoplasmosis infected and uninfected individuals (t= -2.504, p = 0.014). This finding corresponds with (Advincula *et al*., 2010) in Philippine, Al-Obaidi (2011) in Al-Mosul city, Iraq, and Wang *et al*. (2015) in Chain, and it disagrees with Hassen *et al*. (2019) in Libya, Mohamed (2020) in Suadi Arabia, and Agordzo *et al*. (2020) in Ghana. This decrease may be due to the occurrence of anemia as a result of the effect of *Toxoplasma* in both erythropoiesis and the period of erythrocyte survival in the circulation (Wang, *et al*., 2015). This study showed that the decrease in erythrocyte production during toxoplasmosis represents 9.40% (4.830/5.144), which consists of a small percentage of the total number of erythrocytes. This reduction of erythrocytes may be returned to other factors, as occur with malaria, when the uninfected RBCs that are bounded around the merozoites are exposed to destruct by phagocytosis (Jakeman *et al*., 1999). In many cases, the parasite does not directly attack the red blood cells and cause their lyses in blood vessels but may indirectly and rapidly cause the destruction of RBCs and hemolysis in the liver and spleen (McCullough, 2014). There are many mechanisms that have been

suggested such as the adsorption of immune complexes and complement on RBCs surfaces, production of cross-active immunoglobulins, and development of true autoimmunity with loss of tolerance secondary to parasites, for this, RBCs are affected by this pathogen (McCullough, 2014). on the other hand, it shows no significant effect of interaction between gender and toxoplasmosis on the count of RBCs (F=1.250, p= 0.267) (see Table 3-11).

Table 3-11: The relationship between toxoplasmosis-human gender on the count of RBCs (x10^6/mm^3).

The gender	**Infection**	**Examined No.**	**RBCs count mean x10^6/mm^3**
Male	infected	11	5.052
	uninfected	53	5.235
	Total	64	5.203
Female	infected	14	4.656
	uninfected	8	4.541
	Total	22	4.61455
Total	infected	25	4.83040
	uninfected	61	5.14377
	Total	86	5.05267

F= 1.250, p= 0.267

3.3.3. Effect of Toxoplasmosis on the percentage of hematocrit (HCT%)

The mean percentage of HCT in toxoplasmosis-infected individuals is 41.033%, compared to 46.806% in uninfected individuals. There are high statistically significant differences between *T. gondii* infected individuals and uninfected individuals in the percentage of HCT (t = -4.881, p = 0.000005).

Table (3-12): Effect of toxoplasmosis on the percentage of hematocrit (HCT%) levels.

Infection	Examined No.	Mean of HCT%	t-test	*p*
Infected	25	41.030 ± 5.374	-4.236	0.000005
Uninfected	61	46.806 ± 4.436		

± SD

This study finds that toxoplasmosis caused a significant reduction in the HCT% in infected individuals. This result was also shown in infected camels by Shehzad *et al*. (2022) in Pakistan. The reduction of HCT% in the toxoplasmosis seropositive individuals in this study may be due to the reduction of RBCs, where we observed a highly significant correlation between the number of erythrocytes and HCT%. (r= 0.809, p< 0.000001). But it disagrees with Hassen *et al.* (2019) in Libya and Mohamed (2020) in Saudi Arabia.

The results of the current study (Table 3-13) show the mean of HCT% of toxoplasmosis-seropositive among each male and female is higher

than that of uninfected, but there are no significant differences between the infected and uninfected males and females (F= 3.718, p> 0.05).

Table 3-13: Effect of toxoplasmosis on the percentage of HCT according to gender.

The gender		Examined No.	Mean of HCT%
Male	Infected	11	43.582 ± 5.285
	Uninfected	53	39.564 ± 3.242
	Total	64	41.332 ± 3.974
Female	Infected	14	47.891 ± 4.918
	Uninfected	8	39.625 ± 4.770
	Total	22	46.806 ± 4.750
Total	Infected	25	47.150 ± 5.374
	Uninfected	61	39.586 ± 4.436
	Total	86	45.21512 ± 5.319

F= 3.718, p> 0.05, ± SD

3.3.4 The effect of Toxoplasmosis on the Mean of Corpuscular Volume (MCV)

The results of the CBC test show that the mean MCV of erythrocytes of toxoplasmosis-infected individuals is 85.88 μm^3, while that of uninfected individuals is 91.17 μm^3 (Table 3-14). There are statistically significant differences between infected and uninfected in MCV (t = -3.27, p = 0.00035).

The mean normal volume of human erythrocytes ranges between 80-98μm^3. This result is in line with what has been shown by previous

studies such as Hassen *et al.* (2019) in Libya, Mohamed (2020) in Suadi Arabia and Agordzo *et al*. (2020) in Ghana. The MCV is one of the variables in a comprehensive blood count. It is used to differentiate between types of anemia (Tønnesen *et al*., 1986).

Table 3-14: The effect of toxoplasmosis on the mean of corpuscular volume (MCV)

	Mean µm^3	**t- test**	***p***
Infected	85.88 ± 8.779	-3.725	0.00035
Uninfected	91.15 ± 4.848		

± SD

3.3.5 The effect of Toxoplasmosis on the mean corpuscular hemoglobin concentration (MCHC)

The results of the current study show (Table 3-15) that the mean of MCHC for the toxoplasmosis infected individuals is 26.28g/dl and for uninfected is 28.93g/dl. There are statistically significant differences between the infected and uninfected individuals in the MCHC.

Table 3-15 The effect of Toxoplasmosis on the mean corpuscular hemoglobin concentration (MCHC)

	Mean g/dl	**t-test**	***P***
Infected	26.284 ± 3.736	-4.200	0.000066
Uninfected	28.933 ± 2.071		

± SD

These results are less than what was recorded in Saudi Arabia, where the MCHC of the blood of infected individuals is 31.8 and uninfected 32.1, where there were no statistically significant differences (Mohamed, 2020). The decline in MCHC may be due to the decrease in the hemoglobin level or/and RBCs count (see Table 3-9, 3-10).

3.3.6. The effect of Toxoplasmosis on the Red corpuscles distribution width (RDW)

The current study (Table 3-16) shows the mean RDW of toxoplasmosis-infected individuals is 12.11%, and that of the uninfected is 11.36%. There are no statistically significant differences.

Table 3-16The effect of Toxoplasmosis on the Red corpuscles distribution width (RDW%)

	RDW% ± SD	**t-test**	***p***
Infected	12.110 ± 1.272	2.636	0.093
Uninfected	11.368 ± 0.931		

These results are less than what was recorded in Saudi Arabia, where it was infected (14.4-14.9) and uninfected (14.1-13.4), where there were no statistically significant differences (Mohamed, 2020). The RDW (Red Blood Corpuscle Distribution Width) is a basic measure of the broadness of the erythrocyte size distribution, often known as anisocytosis (Salvagno *et al*., 2015).

3.3.7 The effect of toxoplasmosis on the count of white blood Cells (WBCs) (x10^3/mm^3)

The current results (Table 3-17) showed the mean of WBCs of toxoplasmosis-infected individuals was 6.654 x 10^3/mm^3, which is lower than the uninfected 6.992 x 10^3/mm^3. There are no significant differences between the counts of WBCs of infected and uninfected people ($t = -0.715$, $p = 0.477$).

This result did not agree with the findings of a study in Najaf, as there was a statistically significant increase at $p \leq 0.05$ (Al-Obaidi 2011; Abdul Abbas 2015). Also, it disagrees with Hassen *et al*. (2019) in Libya and Mohamed (2020) in Saudi Arabia. The reduction of the count of leukocytes may be returned to the destruction of many monocytes such as macrophages by the parasite or due to the destruction of blood-generating cells in the bone marrow, or it may be due to the effect of the parasite on macrophages that reduce the immune response as they secrete mediating factors to reduce the response, as it is interpreted that the spleen macrophages that were infected with *T. gondii* secreted some mediators that act as soluble factors that mediate a transient immune-suppressive, which helped the *T. gondii* to establish (Channon and Kasper, 1996).

Table 3-17: The effect of *Toxoplasma* on the number of the count of WBCs ($x10^3/mm^3$).

	Mean $10^3/mm^3$	**t-test**	***p***
Infected	6.654 ± 2.210	-0.715	0.477
Uninfected	6.992 ± 1.844		

± SD

It was also noted (Table 3-18) that the count of white blood cells in infected females is much higher than the count in uninfected females, in contrast to the uninfected ones. While it was noted that the count of blood cells in males is higher than in females, this is within the normal range.

There were highly statistically significant differences between infected and uninfected males and females (F= 14.741, p= 0.000245).

Table 3-18: The relation between toxoplasmosis-human gender on the count of WBCs (x10^3/mm^3).

	The gender	Examined No.	Mean x10^3/mm^3
Infected	**Male**	11	5.00909 ± 1.177671
	Female	13	8.04615 ± 1.905962
	Total	24	6.65417 ± 2.210839
Uninfected	**Male**	52	7.09615 ± 1.843267
	Female	8	6.31250 ± 1.817720
	Total	60	6.99167 ± 1.844211
Total	**Male**	63	6.73175 ± 1.912170
	Female	21	7.38571 ± 2.019972
	Total	84	6.89524 ± 1.948240

F= 14.741 p= 0.00024, ± SD

Table 3-19: The distribution of lymphocytes among Toxoplasmosis infected and uninfected individuals

	No. of lymphocytes **10^3/mm^3**																										total
	0.9	1.2	1.3	1.4	1.5	1.6	1.7	1.8	1.9	2.0	2.1	2.2	2.3	2.4	2.5	2.6	2.7	2.8	2.9	3.0	3.3	3.5	3.6	4.0	4.1	4.3	
Infected	0	0	0	3	0	0	0	3	0	5	0	2	1	0	0	2	0	1	0	0	0	0	1	1	0	1	20
Uninfected	1	1	1	2	2	6	2	4	8	1	1	1	8	3	3	4	1	2	2	2	1	2	0	0	1	0	59

X^2= 40.37, p= 0.027

The present results (Table 3-19) show there are statistically significant differences between infected and uninfected individuals in the count of lymphocytes (X^2=40.37, p=0.027). This result agrees with the findings of a study in Najaf, as there was a statistically significant increase at p≤0.05 (Abdul Abbas 2015).

It also shows in Table 3-20 that the mean of lymphocytes counting for infected individuals 2.52 x10^3/mm^3 is higher than that of uninfected 2.16 x10^3/mm^3. There is no statistically significant difference between the counts of lymphocytes of Toxoplasmosis infected and uninfected at the number of (P≤0.05).

Table 3-20: The effect of Toxoplasmosis on the number of lymphocytes ($x10^3/mm^3$)

	Mean $10^3/mm^3$	**t-test**	***p***
Infected	2.520 ± 0.833	1.762	0.269
Uninfected	2.160 ± 0.621		

± SD

This result agrees with the findings of a study by Abdul Abbas (2015) in the Najaf Province.

Also, the results of this study (Table 3-21) show no statistically significant differences in the distribution of monocytes among Toxoplasmosis-infected and uninfected individuals (X^2= 11.679, p = 0.112).

Table 3-21: Distribution of monocytes among Toxoplasmosis infected and uninfected individuals

	Count of monocytes $10^3/mm^3$								**Total**
	0.1	0.2	0.3	0.4	0.5	0.6	0.7	1.1	
Infected	3	6	6	2	1	1	1	0	20
Uninfected	1	13	19	17	6	2	0	1	59

X^2= 11.679, p= 0.112

The statistical analysis of the results shows no statistically significant

differences between the counts of monocytes of toxoplasmosis-infected and uninfected individuals (Table 3-22).

Table 3-22: The effect of Toxoplasmosis on the mean of the count of monocytes (x10^3/mm^3)

	Mean 10^3/mm^3 of monocytes	**t-test**	***p***
Infected	0.333 ± 0.163	-0.226	0.612
Uninfected	0.344 ± 0.159		

± SD

This result did not agree with the findings of a study in Najaf, but the increase was statistically significant($P \leq 0.05$) (Abdul Abbas 2015).

The CBC results revealed that the mean of granulocytes in Toxoplasmosis infected individuals is 4.74 x 10^3/mm^3, while that of uninfected individuals is 4.505 x 10^3/mm^3. There are no statistically significant differences between Toxoplasmosis infected and uninfected individuals in the count of granulocytes ($t = 0.419$, $p = 0.286$).

Table 3-23 The effect of Toxoplasmosis on the Mean of the number of granulocytes (x10^3/mm^3)

	Mean 10^3/mm^3	**t-test**	***p***
Infected	4.740 ± 2.282	0.419	0.286
Uninfected	4.505 ± 1.703		

± SD

Although the results of this study are close to the results found by Abdul Abbas (2015) in Najaf Province, they differ from them in that the differences between the groups of individuals infected with and not infected with *Toxoplasma* are not statistically significant (see Table 3-23).

3.3.8 The effect of Toxoplasmosis on the count of Platelets

The results of the current study show the mean (± S.D) of the platelet count is 256.6 (±90.02) in the infected and the uninfected is 197 (±62.8). There are no statistically significant differences in platelet counts between Toxoplasmosis-infected and uninfected people (see Table 3-24).

Table 3-24: The effect of Toxoplasmosis on the platelets count($x10^3/mm^3$)

	Mean $10^3/mm^3$	**t-test**	***p***
Infected	256.600 ± 90.021	3.053	0.076
Uninfected	197 ± 62.838		

± SD

This result did not agree with the findings of Al -Obaidi (2011) in the Nineveh Province. This increase in the count of platelets in the present study may be due to the effect of *Toxoplasma gondii* on the multiplication of animalcules quickly inside the muscle cell, which leads to the occurrence of essential and direct harm in the lining cells,

which leads to the gathering of platelets and the formation of the blood thrombosis (Jonies *et al*.,1997).

On the other hand, the results of the current study show (Table3-25) that the mean (± S.D) of the mean of the platelet volume is 8.915 μm^3 (±0.7) in the infected and the uninfected 8.627 μm^3 (±0.9), but there are no statistically significant differences between the mean of MPV and the Toxoplasmosis infected and uninfected individuals (see Table 3-25).

Table 3-25: **The effect of Toxoplasmosis on the platelet volume (MPV)**

	MPV in μm^3	**t-test**	***p***
Infected	8.915 ± 0.7322	1.212	0.166
Uninfected	8.627 ± 0.943		

± SD

The current study's results (Table3-26) show that the mean (± S.D) of procalcitonin is 0.230% (±0.08) in the infected and 0.168% (±0.05) in the uninfected, with no statistically significant differences between the mean of PCT and the Toxoplasmosis infected and uninfected individuals (see Table 3-26).

Table 3-26: **The effect of Toxoplasmosis on the procalcitonin (PCT%)**

	Mean of PCT %	**t-test**	***p***
Infected	0.230 ± 0.085	3.526	0.07
Uninfected	0.168 ± 0.053		

± SD

The results of the current study (Table3-27) show that the mean (± S.D) of the mean platelet distribution width is 13.850% (±2.04) in the infected and the uninfected, 13.235% (±1.22), and there is no statistically significant in the effect of *T. gondii* on PDW (t=1.501, p>0.05).

Table 3-27: The effect of Toxoplasmosis on the mean of the platelet distribution width (PDW%)

	Mean of PDW	**t-test**	***p***
Infected	13.850 ± 2.046	1.501	0.078
Uninfected	13.235 ± 1.229		

± SD

The statistical analysis of the results (Table 3-28) reveals a statistically significant association between toxoplasmosis infection and PDW (p = 0.034).

Table 3-28: The effect of Toxoplasmosis on the Platelet distribution width (PDW%)

	Platelet distribution width %								Total
	≤11.0	11.1-12.0	12.1-13.0	13.1-14.0	14.1-15.0	15.1-16.0	16.1-17.0	20.1 +	
Infected	0	3	10	2	8	1	0	1	25
Uninfected	1	6	22	20	5	5	2	0	61

X2=15.179, p=0.034

3.4 Relationship between Toxoplasmosis and some sociodemographic factors

In this study, the relationship between toxoplasmosis and some sociodemographic factors is investigated by questionnaire according to the information of the direct question to each participant whose blood samples were taken.

3.4.1 The relationship between Toxoplasmosis and the age

The current study (Table 3-29) exhibited about three quarters of the study population ranged between 25-44 years, and the age of 71% of infected cases ranged between 25-44 years, no statistically significant association between the age and *T. gondii* infection (X^2=4.613, p>0.005).

Table 3-29: The relationship between Toxoplasmosis and the age

Result of ELISA	Years age groups				Total
	15-24	25-34	35-44	45-54	
Infected	14	30	31	10	85
Uninfected	5	20	34	7	66
Total	19	50	65	17	151

X^2=4.613, p>0.05

These results agreed with the results of a study conducted in Egypt (Elsheikha *et al*., 2009) which found that toxoplasmosis was not related to age, and not compatible with (Pinlaor *et al*., 2000; Coelho *et al.*, 2003 and Sarkari *et al*., 2014) who found there is a significant association between age and toxoplasmosis.

3.4.2 The Relationship between Toxoplasmosis and the academic level

Table3-30 showed a very strong relationship between education level and *T. gondii* infection (X^2=50.094, P<0.000005). It showed that low education (illiterate and primary) and high education (Bachelor's and high education) had the highest infection rate (Table 3-30).

Table 3-30: Relationship between education level and toxoplasmosis

		Infected	Uninfected	Total
Education level	Unknown	4	0	4
	% of overall ↓	4.4%	0.0%	2.3
	% of row →	100%	0.0%	100%
	Illiterate	8	0	8
	% of overall ↓	8.8%	0.0%	4.6%
	% of row →	100%	0.0%	100%
	Primary	30	8	38
	% of overall ↓	33%	9.6%	21.8%
	% of row →	78.9%	21.1%	100%
	Middle	14	26	40
	% of overall ↓	15.4%	31.3%	23%
	% of row →	35%	65	100%
	Secondary	10	27	37
	% of overall ↓	11%	32.5%	21.3%
	% of row →	27%	73%	100%
	Diploma	7	18	25
	% of overall ↓	7.7%	21.7%	14.4%
	% of row →	28%	72%	100%
	Bachelor's	15	4	19
	% of overall ↓	16.5%	4.8%	10.9%
	% of row →	78.9%	21.1%	100%
	High education	3	0	3
	% of overall ↓	3.3%	0.0%	1.7%
	% of row →	100%	0.0%	100%
Total		91	83	174
% of overall		100%	100%	100%
% of row		52.3%	47.7%	100%

X^2=50.094, p<0.000005

These results agreed with the results of a study conducted in Egypt by Elsheikha *et al*. (2009), in Durango, Mexico (Alvarado-Esquivel *et al*., 2007) and in Iran (Sarkari *et al.,* 2014).

It was noted that the infection rate increased among people with a low educational level (Daryani *et al*., 2014), such as illiterates and primary, and decreased at the intermediate educational level such as middle school secondary school, and diploma, and this may be due to their lack of culture and they are ignoring to follow health instructions in order to avoid factors that may make him a victim of the disease(Carmo and Guizardi 2018), but it is striking that the rate of infection recurred when people with higher education such as Bachelor's and Master's, there is no clear explanation for this situation, only one reason, that may be associated with the economic level and the culture of these categories of society, the fact the educated people may have higher positions and more salaries, which may make them eat meat more than lower categories or/and they may have cats in their homes (Wilking *et al*. 2016).

3.4.3 Relationship between the level of income and Toxoplasmosis

The present study (Table 3-31) shows that the highest infection of 100% (3/3) was recorded in low income level category, the statistical analysis of the results reveals a statistically significant association between toxoplasmosis infection and income level (X2= 97.814, p= 0.000011).

Table 3-31: The relationship between the income level and the infection in Toxoplasmosis

		Low	Medium	High	Very high	Total
Infected	No.	3	13	9	0	25
	% of row→	12	52	36	0	
	% of total↓	100	24.07	34.62	0	
Uninfected	No.	0	41	17	3	61
	% of row→	0	67.2	27.9	4.9	
	% of total↓		75.93			
Total		3	54	26	3	86

X^2= 97.814, P< 0.0000001

This finding is in line with the finding of Mareze *et al*. (2019) who found about 93.29% of low-income people are infected with *T. gondii*. Also, some studies reported that the low income individuals had a higher infection rate of toxoplasmosis (Mareze *et al*., 2019, Lachkhem *et al*., 2020, Singh *et al*., 2021) and disagreed with Alvarado-Esquivel *et al*. (2011). For this, show the economic level is one of the affected factors in *Toxoplasma* prevalence (Al-Malki, 2021). Some believe that

getting eradication of toxoplasma cannot be done unless the economic situation of the population is improved (Al-Malki, 2021). The high infection rate among the low income individuals may be returned to more than one socio-economic factors like, poor nutrition, education, jobs, house style, water supply, sanitation, contact with cats, feeding habitats and region (Daryani *et al*., 2014; Mareze *et al*., 2019, Carvalho *et al*., 2021).

3.4.4 The relationship between Toxoplasmosis and Diabetes

Table 3-32 shows that the seroprevalence of *T. gondii* in diabetic cases is 68% and in healthy-diabetic individuals is 20.51%. The seroprevalence of toxoplasmosis in diabetic cases is more than three folds than that of healthy-diabetic groups (331.54%). There is a significantly relationship between diabetes and toxoplasmosis (X^2= 14.432 p=0.000145).

Table 3-32: The relationship between Toxoplasmosis and the diabetes

	Diabetes		**Total**
	No. had diabetes (%)	No. non had diabetes (%)	
Infected	17(68)	8(20.51)	25
Uninfected	8(32)	31(79.49)	39
Total	25	39	64

X^2=14.432, p=0.000145

This finding agreed with the finding of Saheb (2017) in Baghdad, Iraq, that the prevalence of toxoplasmosis in diabetic cases and healthy controls were 55.81% and 38.78% respectively, and in Iran with Shirbazou *et al.* (2013), in that the seroprevalence of IgG Anti-*T. gondii* in diabetic cases and healthy-individuals were 60.43% and 38% respectively and Khalili *et al*. (2018), found the anti-*T. gondii* IgG seropositivity in type I and type II diabetes and healthy individuals were 69%, 63% and 59% respectively.

This also agreed with Alvarado-Esquivel *et al*. (2011) in Mexico. From this, can conclude that people who are infected with Toxoplasmosis are more receptive to diabetes than healthy-individuals, this may be due to the fact that *Toxoplasma gondii* causes the destruction of pancreatic cells such as beta cells, which reduced its effectiveness for producing insulin, or the parasite may work to destroy nerve (Shirbazou *et al*., 2013).

3.4.5 The relationship between Surgery and Toxoplasmosis

This study **(**Table 3–33), finds that 61.5% of individuals who history of surgery is infected with *T. gondii* compared with 33.3% without. There is a statistically significant relationship between the history of surgeries and infection with toxoplasmosis (X^2= 3.462, $p<0.05$).

Table 3-33: The relationship between and surgeries Toxoplasmosis

		Total	History of surgery	
			No	Yes
Infected	No.	17	8	25
	% of total infected	68.0%	32.0%	100.0%
	% of total column	33.3%	61.5%	39.1%
Uninfected	No.	34	5	39
	% of total infected	87.2%	12.8%	100.0%
	% of total column	66.7%	38.5%	60.9%
Total	No.	51	13	64
	% of total infected	79.7%	20.3%	100.0%
	% of total column	100.0%	100.0%	100.0%

X^2= 3.462, p<0.05

This finding agrees with the finding of Alvarado-Esquivel *et al*. (2006), and with Alvarado-Esquivel *et al.* (2015) in Mexico and with a history of caesarean sections (Alvarado-Esquivel *et al.,* 2016), but disagrees with Alvarado-Esquivel *et al*. (2018a), in Mexico. For this, it is considered that the history of surgery is an important risk factor for the transmission of infection with *T. gondii* (Alvarado-Esquivel *et al.*, 2018a).

3.4.6 The relationship between blood transfusion and infection of recipients with the Toxoplasmosis

The current result (Table 3–34) showed all individuals (100%) who had histories of blood transfusion are infected with toxoplasmosis compared

with 30.4% among individuals who no had histories. The infected individuals who had histories of blood transfusion count for 32% of total infections, and they consist of 12.5% of total samples. There is a statistically significant relationship between the infection with toxoplasmosis and blood transfusion (X^2= 14.263, p= 0.000159).

Table 3-34: The relationship between blood transfusions and infection with *T. gondii*

		Blood transfusions		Total
		No	Yes	
Infected	No.	17	8	25
	% of total infected	68.0%	32.0%	100.0%
	% of total column	30.4%	100.0%	39.1%
Uninfected	No.	39	0	39
	% of total infected	100.0%	0.0%	100.0%
	% of total column	69.6%	0.0%	60.9%
Total	No.	56	8	64
	% total	87.5%	12.5%	100.0%
	% within blood-trans.	100.0%	100.0%	100.0%

X^2 = 14.263, p = 0.000159

The results of this study are closely identical to those in Serbia, where it was found that 98.2% of seropositive blood donors' samples contain IgG antibodies (Stopić *et a.*, 2022). And these results agreed with more than one study which reported that blood transfusion is active in the transmission of toxoplasmosis (Karimi *et al*., 2014, Mahmoudvand *et al*., 2015, Foroutan-Rad *et al*., 2016, Alvarado-Esquivel *et a*l 2018b, Wangetal., 2018).

The presence of the organism in blood during the course of infection ensures its transmission through transfusion (Perkins and Busch 2010), for this, blood transfusions are considered an important risk factor for the blood transmission of infection with *T. gondii.*

3.4.7 The relationship between Toxoplasmosis and visual impairment:

Table (3-35) shows there are only 6 patients suffering from visual impairment, about 66.7% of them had toxoplasmosis, compared to 33.3% of individuals without infection. In another hand, it shows only 16% of infected individuals had visual impairment.

There is no statistically significant relationship between toxoplasmosis and visual impairment (X^2= 2.119, p>0.05).

This finding agrees with the finding of Alvarado-Esquivel *et al*., (2016), who found no statistically significant relationship between toxoplasmosis and visual impairment (p=0.27), and disagrees

Table 3-35: The relationship between Toxoplasmosis and visual impairment

		Visual impairment defect		Total
		No	Yes	
infected	No.	21	4	25
	% of infected	84.0%	16.0%	100.0%
	% within visual	36.2%	66.7%	39.1%
uninfected	No.	37	2	39
	% of uninfected	94.9%	5.1%	100.0%
	% within visual	63.8%	33.3%	60.9%
Total	No.	58	6	64
	% of infected	90.6%	9.4%	100.0%
	% within visual	100.0%	100.0%	100.0%

X^2= 2.119, p>0.05

with previous studies of Kadarisman *et al*. (1991), Tan *et al*. (2007), and Sheets Aleixo *et al*. (2016), who found a significant relationship between toxoplasmosis and visual impairment.

3.4.8 The relationship between Toxoplasmosis and Gland dysfunction:

Table 3-36 shows no one of the patients with gland dysfunction had been infected with toxoplasmosis, and there was no significant relationship between infection and gland dysfunction (X^2=2.018, *p*= 0.155). In the world, there are very few studies on the relationship between *T. gondii* and gland dysfunctions (Alvarado-Esquivel *et al*. 2019). In Iraq, did not find studies in this field except for the study of

Molan and Rasheed (2016), who only discussed the relationship between thyroid cancer toxoplasmosis. It believes that the current study confirmed what to Alvarado-Esquivel *et al.* (2019) found that there was no significant relationship between thyroid dysfunction and toxoplasmosis. Remarkably, it was observed in this study that although there was no significant relationship (P > 0.05), subjects with glandular dysfunction were free of toxoplasmosis infection.

Table 3-36: The relationship between Toxoplasmosis and gland dysfunction

		Gland dysfunction		Total
		No	Yes	
infected	No.	25	0	25
	% of infected	100.0%	0.0%	100.0%
	% within gland dysfunction	41.0%	0.0%	39.1%
uninfected	No.	36	3	39
	% of uninfected	92.3%	7.7%	100.0%
	% within gland dysfunction	59.0%	100.0%	60.9%
Total	No.	61	3	64
	% within the result of ELISA	95.3%	4.7%	100.0%
	% within gland dysfunction	100.0%	100.0%	100.0%

X^2=2.018, p= 0.155

3.4.9 The relationship between Toxoplasmosis and suffering from an intermittent headache

Table 3–37 shows that all individuals (100%) suffering from intermittent headaches are infected with toxoplasmosis, while the healthy free from toxoplasmosis.

There is a significant relationship between toxoplasmosis and intermittent headaches (X^2=10.328, p=0.001).

This finding is in line with the finding of Koseoglu *et al*. (2019) who reported that migraine headaches are significantly related to toxoplasmosis.

Prandota (2009) reported that toxoplasmosis patients who suffer from different types of headaches and other neurological signs appear to be produced by a Jarisch-Herxheimer reaction caused by the apoptosis of *Toxoplasma gondii* tachyzoites. But it disagrees with the finding of Alvarado-Esquivel *et al*., (2018c), who showed no relationship between toxoplasmosis infection and some headache types. In contrast, it shows only 24% (6/25) of *Toxoplasma gondii*-seropositive cases are suffering from intermittent headaches, this closely resembles findings by Nayeri *et al.* (2021) who found that 17.67% of toxoplasmosis cases are suffering from headaches. For this, it can be concluded that headache is an important sign of toxoplasmosis (Cox and John-Alde, 2005).

Table 3-37: The relationship between Toxoplasmosis and intermittent headache.

		headache		Total
		No	Yes	
Infected	No.	19	6	25
	% of infected	76.0%	24.0%	100%
	% within headache	32.8%	100%	39.1%
Uninfected	No.	39	0	39
	% of uninfected	100%	0.0%	100%
	% within headache	67.2%	0.0%	60.9%
Total	No.	58	6	64
	% of row	90.6%	9.4%	100%
	% of column	100%	100%	100%

X^2= 10.328, p= 0.001

3.4.10 The relationship between Toxoplasmosis and fever:

Table 3–38, Shows that 71.4% of those suffering from fever are toxoplasmosis seropositive, compared with 28.6% of seronegative. In contrast, the cases suffering from fever comprise 40% of the toxoplasmosis seropositive compared with 6.6% of uninfected individuals. There is a significant relationship between toxoplasmosis and suffering from fever (X^2= 14.552, p=0.000136). Fever is one of the important signs or symptoms that a person is exposed to when infected with the *Toxoplasma* parasite (Dubey, 1996; Hadfield and Guy, 2017).

Table 3-38: The relationship between Toxoplasmosis and the fever sign.

		Suffering from a fever		Total
		No	Yes	
Infected	No.	15	10	25
	% of infected	60.0%	40.0%	100.0%
	% of fever	20.8%	71.4%	29.1%
Uninfected	No.	57	4	61
	% of uninfected	93.4%	6.6%	100.0%
	% of fever	79.2%	28.6%	70.9%
Total	No.	72	14	86
	% row	83.7%	16.3%	100.0%
	% % of fever	100.0%	100.0%	100.0%

3.4.11 The relationship between Toxoplasmosis and Suffering from muscular pain

Table 3-39 shows that 69.2% of toxoplasmosis seropositive cases are suffering from muscular pain compared with 31.8% of seronegative. The percentage of individuals who suffering from muscle pain in toxoplasmosis infected cases is more than twice (217.61%) than that of non-infected. There is significant relationship between toxoplasmosis and Muscular pain (X^2= 11.980, p=0.01). These findings confirm what Dubey mentioned that muscular pains are one of toxoplasmosis signs (Dubey, 1996). The muscle damage caused by Toxoplasmosis may be due to a limited immune response in these tissues (Wiendl *et a*l., 2005).

Table 3-39: Relationship between *T. gondii* and muscular pain.

		Suffering from muscular pain		Total
		NO	YES	
Infected	No.	16	9	25
	% infected	64.0%	36.0%	100.0 %
	% within Suffering from muscular pain	21.9%	69.2%	29.1%
Uninfected	No.	57	4	61
	% infected	93.4%	6.6%	100.0 %
	% within Suffering from muscular pain	78.1%	30.8%	70.9%
Total	No.	73	13	86
	% infected	84.9%	15.1%	100.0 %
	% within Suffering from muscular pain	100.0%	100.0%	100.0 %

3.4.12 The relationship between Toxoplasmosis and Suffering from tiredness and fatigue

The current results (Table 3-40) shows that 60% of individuals who suffering from tiredness and fatigue are from toxoplasmosis cases. There is significant relationship between toxoplasmosis and tiredness and fatigue sign (X^2=8.43, p=0.04). It was shown that the tiredness and fatigue sign and then high mortality rates of COVID-19 patients in cases in their latent *T. gondii* infection (Harrison *et al*. 2020; Hamer *et al.* 2020). This results don't agree with Roe who found only 4% of individuals who could meet the criteria for chronic fatigue syndrome

were infected with toxoplasmosis. Therefore, toxoplasmosis can be considered an uncommon cause of chronic fatigue syndrome.

Table 3-40: The relationship between Toxoplasmosis and the tiredness and fatigue

		Suffering from tiredness and fatigue		Total
		NO	YES	
Infected	No.	16	9	25
	% infected	64.0%	36.0%	100.0 %
	% tiredness and fatigue	22.5%	60.0%	29.1%
Uninfected	No.	55	6	61
	5 infected	90.2%	9.8%	100.0 %
	% tiredness and fatigue	77.5%	40.0%	70.9%
Total	No.	71	15	86
	% infected	82.6%	17.4%	100.0 %
	% tiredness and fatigue	100.0%	100.0%	100.0 %

X^2=8.43, p=0.04

3.4.13 The relationship between Toxoplasmosis and Suffering from constant headache

Table 3–41 shows that 71.4% of suffering from constant headaches are infected with toxoplasmosis, compared with 28.6% of uninfected. These results are less than what was mentioned by Hill *et al*. (2007) who reported that 88% of cases suffering from headaches. There is a significant relationship between toxoplasmosis and constant headaches (X^2=6.631, p=0.02).

Table 3-41 The relationship between Toxoplasmosis and the suffering from constant headache

		Suffering from chronic headache		Total
		NO	YES	
Infected	Count	20	5	25
	% infected	80.0%	20.0%	100.0%
	% chronic headache	25.3%	71.4%	29.1%
Uninfected	Count	59	2	61
	% infected	96.7%	3.3%	100.0%
	% chronic headache	74.7%	28.6%	70.9%
Total	Count	79	7	86
	% infected	91.9%	8.1%	100.0%
	% chronic headache	100.0%	100.0%	100.0%
		91.9%	8.1%	100.0%

X^2= 6.631, p= 0.02

This finding is in line with the finding of Dubey (1996) and Koseoglu *et al*. (2019) who reported that the types of headaches are significantly related to toxoplasmosis. This sign appears to be produced by a Jarisch-Herxheimer reaction caused by the apoptosis of *Toxoplasma gondii* tachyzoites (Prandota, 2009). But it disagrees with the finding of Alvarado-Esquivel *et al*. (2018c), who showed no relationship between toxoplasmosis infection and some headache types. Table 41 shows that 20%(5/25) of toxoplasmosis-seropositive cases are suffering from constant headaches, this finding is closely to that the findings by Nayeri and colleagues (Nayeri *et al.*, 2021) who found that 17.67% of toxoplasmosis cases are suffering from headaches. For this, it can be concluded that the headache is an important sign of toxoplasmosis (Cox and John, 2005).

3.4.14 The relationship between Toxoplasmosis and Eating dairy products from street vendors and Drink unpasteurized milk

Table 3-42 shows that 37.5% of toxoplasmosis infected individuals are drinking unpasteurized milk compare with 22.20% of non-drinking unpasteurized-milk. There is no statistically significant-relationship between toxoplasmosis and drinking unpasteurized milk (X^2= 0.304, p>0.05).

On the other hand, Table 3-42 shows that 42.42% of infected individuals are eating dairy products from street vendors compared with 20.75% among non-eating these products. There is statistically significant-relationship between toxoplasmosis and eating dairy products from street vendors (X^2= 4.631, p=0.031).

Table 3-42 The relationship between toxoplasmosis drink unpasteurized milk and eating dairy products from street vendors.

Taking dairy product		**Toxoplasmosis**			**X^2**	***p***
		Infected %	**Uninfected %**	**Total**		
Unpasteurized Milk	**Yes**	3 (37.5)	5 (62.5)	8	0.304	0.05
	NO	22 (28.20)	56 (71.80)	78		
	Total	25	61	86		
Other Dairy products from street vendors.	**Yes**	14 (42.42)	19 (57.58)	33	4.631	0.031
	NO	11 (20.75)	42 (79.25)	53		
	Total	25	61	86		

X^2= 4.631, p=0.031

These results have not agreed with the results of some studies such as, Egypt (Elsheikha *et al*., 2009), USA (Jones *et al*., 2009), (Lopez *et al*., 2000), (Cook *et al*., 2000) and (Dubey and Jones 2008). And agree with them about eating other dairy products, for which statistical analysis has proven a significant relationship between infection with *Toxoplasma* gondii and eating cheese products from street vendors. It was shown that the consumption of unpasteurized goat's milk has been linked to acquired clinical toxoplasmosis in humans on rare occasions (Tenter *et al*., 2000).

3.4.15 The relationship between Toxoplasmosis and contact with cats:

Table (3-43) shows that 63.6% of individuals who contact with cats are infected with toxoplasmosis, compared with 24.0% among non-contact with cat. There are statistically significant differences between Toxoplasmosis and contact with cats (X^2=7.309, p= 0.007).

Table 3-43 The relationship between Toxoplasmosis and contact with cats

		Contact with cat		Total
		NO	YES	
Infected	No.	18	7	25
	%infected	72.0%	28.0%	100.0%
	%contact with cat	24.0%	63.6%	29.1%
Uninfected	No.	57	4	61
	%infected	93.4%	6.6%	100.0%
	%contact with cat	76.0%	36.4%	70.9%
Total	No.	75	11	86
	%infected	87.2%	12.8%	100.0%
	%contact with cat	100.0%	100.0%	100.0%

(X^2=7.309, p= 0.007).

These findings agreed with a study by Salih *et al*. (2020), in Dohuk, Iraq (Salih *et al*., 2020), and with Elsheikha *et al*. (2009) in Egypt. Some studies have revealed a significant relationship between the contact with cats and the prevalence of *T. gondii* from (Ayi *et al*., 2009; Zemene *et al*. 2012; Mizuri and Mero 2020). The present study falls in with what was stated by Alvarado-Esquivel *et al*. (2007), that contact with cats as a potential risk factor for acquiring toxoplasmosis (Ayi *et al*., 2009). In contrast, Nissapatorn *et al*. (2002) did not detect a link between contact with cats and *T. gondii* seropositivity in blood donors. It reported that contact with cat fur may not be effective in transmitting the infection, because the immature unsporulated oocytes shed by cats need a period to complete their maturation and transform into mature sporocysts (Alvarado-Esquivel *et al.*, 2007) contact. with cats may be and another study in the city of Durango, Mexico (Alvarado-Esquivel *et al*., 2007). As a result, *Toxoplasma* infections are rare when people come into direct touch with cats (Tenter *et al*., 2000).

3.4.16 The relationship between Toxoplasmosis and Clean the house garden:

The current results (Table, 3-44), shows that only 28.0% of infected individuals had a history of garden cleaning compared with 72.0% are non-cleaning garden. There is no significant relationship between cleaning garden and toxoplasmosis (X^2=0.245, p>0.05).

Table 3-44 The relationship between Toxoplasmosis and the mean number of people who Clean the house garden

		Cleaning the house garden		Total
		NO	YES	
Infected	No.	18	7	25
	%infected	72.0%	28.0%	100.0%
	% cleaning the house garden	27.7%	33.3%	29.1%
Uninfected	No.	47	14	61
	%infected	77.0%	23.0%	100.0%
	% cleaning the house garden	72.3%	66.7%	70.9%
Total	No.	65	21	86
	%infected	75.6%	24.4%	100.0%
	% cleaning the house garden	100.0%	100.0%	100.0%

X^2=0.245, p>0.05

This finding doesn't agree with Alsaady and his and colleagues who reported that human achieved infection with toxoplasma oocyst may be from feral/ stray cats that roam and defecate in the soil of home gardens or around homes in alleys and the infection may occur via swallowing the oocysts in the contaminated dust, soil, food, or water during garden cleaning (Alsaady *et al*., 2021) and the infection may achieved via cleaning the house garden by inhaling the dust that contaminated with the oocyst coming from the cat feces, for this, the cleaning the house or garden had a high risk factor for T. gondii infection (Al-Sadoon *et al.* 2018).

3.5: Relationship between late pregnancy, abortion, and the birthing of a deformed child and toxoplasmosis

3.5.1 The relationship between late pregnancy and Toxoplasmosis

Table (3-45) showed that only 7.69% (1/13) of infected cases had late pregnancy compared without late pregnancy (92.31) %. There is no statistically significant relationship late pregnancy and toxoplasmosis (X^2=1.556, p>0.05).

Table 3-45 The relationship between late pregnancy and Toxoplasmosis

		Late pregnancy		Total
		NO	YES	
Infected	No.	12	1	13
	% infected	92.3%	7.7%	100.0 %
	% late pregnancy	70.6%	33.3%	65.0%
Uninfected	No	5	2	7
	% infected	71.4%	28.6%	100.0 %
	% late pregnancy	29.4%	66.7%	35.0%
Total	No.	17	3	20
	% % infected	85.0%	15.0%	100.0 %
	% late pregnancy	100.0%	100.0%	100.0 %

X^2=1.556, p>0.05

This results also showed by Alvarado-Esquivel *et al.* (2007) that toxoplasmosis didn't related with late pregnancy, and disagreed with Paquet *et al*. (2013) who found the toxoplasmosis effect on pregnancy.

3.5.2 The relationship between abortion and the infection with toxoplasmosis:

It is clear from Table 3–46 that there is a statistically significant difference between abortion and infection (p=0.0001).

Table 3-46 The relationship between abortion and the infection with the toxoplasmosis

	Having an abortion		**Total**
	yes	No	
Infected	7	6	13
Uninfected	2	5	7

X^2=23.292, P=0.0001

This conclusion was consistent with research from Kut City by Al- AL-Mayahi (2011), a study from Mosul by Al-Ubaydi (2004), which revealed that women with a single prior abortion were more likely to be seropositive for *T. gondii* antibodies, and also with findings from Karem (2007). This explains the effect of the parasite in the pregnant women. The sample selection in the current study may have contributed to the relatively high frequency of toxoplasmosis in women who had abortions; the sample was taken from the Al-Shaheed Al-Sadder teaching hospital. People can get toxoplasmosis by eating unwashed, raw vegetables or fruits, or by eating meat that has parasite tissue cysts, especially after 2003, during which time meat consumption increased. In addition, inadequate knowledge of the dangers of

toxoplasmosis among women may contribute significantly to the high likelihood of infection (Nash *et al*., 2005).

3.5.3 The relationship between birthing a deformed child and the infection with toxoplasmosis:

It is clear from Table3-47 that there is a statistically significant difference between birthing a deformed child and infection with Toxoplasmosis (p=0.002).

Table 3-47 The relationship between birthing a deformed child and the infection with the toxoplasmosis

	Having given birth to a child with a congenital malformation		**Total**
	yes	**No**	
Infected	3	10	13
Uninfected	0	7	7

X^2= 17.293, P=0.002

This finding was in agreement with that recorded in Kut city by Al- AL-Mayahi (2011), in Saudi Arabia (Aqeely *et al.*, 2014) and with (Giannoulis *et al.* ,2008). The outcome demonstrated how the parasite affected pregnant women. If the parasite is transmitted to the fetus during the final stages of pregnancy, it will result in pathogenic variation in the newborn in the first few months. Because of the parasite's direct impact on the fetus, when it passes through the placenta and infects the fetus, the infection is severe, especially in the first trimester (Mayahi, 2011).

CONCLUSIONS

AND

RECOMMENDATIONS

Conclusions and Recommendations

Conclusions:

1- The majority of infection cases are chronic, and they are associated with an increase in IgG.
2- A slight increase in acute cases accompanied by an increase in IgM levels or both.
3- The high incidence of infection among individuals participating in the study, especially among females.
4- There is a statistically significant difference in the levels of dopamine between sera with and without toxoplasmosis.
5- There is a statistically significant difference between the levels of adrenaline in the infected and the uninfected.
6- The hemoglobin level and RBCs count in the blood of toxoplasmosis-infected individuals are lower than those of uninfected individuals with a statistically significant difference.
7- WBC and platelet counts in the blood of individuals with toxoplasmosis are lower than those of individuals without the disease, but there are no statistically significant differences between them.
8- No significant differences between the infection ratio and the age.
9- A statistically significant relationship between toxoplasmosis and education level as well as income rate.
10- A statistically significant relationship between diabetes and toxoplasmosis.

11- A statistically significant relationship between toxoplasmosis and the history of surgery, blood transfusion, fever, headache, muscle pain, and fatigue.

12- A statistically significant relationship between toxoplasmosis and contact with cats.

13- No significant differences between the infection ratio and vision problems, and gland dysfunction.

14- No significant differences between the infection ratio and clean the home garden, consuming dairy from street sellers, or drink unpasteurized milk.

15- A statistically significant relationship between toxoplasmosis and late pregnancy, abortion, and mothers of children with birth defects.

Recommendations

1- The Toxoplasmosis antibodies test must be included with other pre-blood donor tests.
2- Increase education and medical care levels between individuals and informed them by the danger of contact with cats.
3- Meat should be cooked above (66 ℃) before eating or freezing under (-20℃).
4- Introducing veterinary hospitals and the necessity of conducting serial examinations for domestic cats and eliminating stray cats, as they are a major source of disease transmission.
5- Conduct survey studies for drinking water to detect oocytes or parasites.
6- Conduct more studies on all factors of the epidemiology of this parasite to know the route of transmission.
7- Conduct more studies to detect the relationship between *T. gondii* and human's behavior.

REFERENCES

References

- Abdel Abbas, H., Abdel-Abbas, M., Abdul-Abbas, S., Mutaib, A. (2015) Measurement of (IgG, IgM) level and some blood parameters in women infected with *Toxoplasma gondii* parasite in Najaf Province. *Journal of Babylon University / Pure and Applied Sciences* 23(1): 454-460 (In Arabic).
- Adamo SA. (2013). Parasites: evolution's neurobiologists. *J Exp Biol*. 2013 Jan 1; 216(Pt 1):3–10.
- Adou-Bryn, K. D., Ouhon, J., Nemer, J., Yapo, C. G., and Assoumou, A. (2004). Serological survey of acquired toxoplasmosis in women of childbearing age in Yopougon (Abidjan, Cote d'Ivoire). *Bull Soc Pathol Exot*, 97(5): 345-348.
- Advincula J.K.C., Iewida, S.Y. and Cabanacan-Salibay, C. (2010) Serologic detection of *Toxoplasma gondii* infection in stray and household cats and its hematologic evaluation. *Sci. Med*., 20(1): 76-82.
- Ageel, N.F. (2003). *Serological and biochemical study of Toxoplasmosis in Tikrit teaching hospital*. M.Sc. Thesis college of Medicine, Tikrit University, Iraq.
- Agordzo, S.K., Badu, K., Addo, M.G., Owusu, C.K., Mutala, A.H., Tweneboah, A., Abbas, D.A. and Ayisi-Boateng, N.K. (2020). Seroprevalence, risk factors and impact of *Toxoplasma gondii* infection on haematological parameters in the Ashanti region of Ghana: a cross-sectional study. *AAS Open Res*. 2020 Jun 17; 2:166.

- Ajioka, J. W. and Soldati, D. (2007). Preface. In: Ajioka, J. W. and Soldati, D. (ed.), *Toxoplasma* Molecular and Cellular Biology. Horizon Bioscience Norfolk, UK. p. xiii–xviii.
- AL-Aboody B A. (2015*).* Prevalence Study of Toxoplasmosis Among Males Blood Donors in Thi-Qar Province –Iraq. M.Sc. Thesis. College of Science University of Thi Qar, Iraq.
- Al-Baldawi, A. (2009) Statistics methods for economic sciences and business administration with the use of the SPSS program, Wael Publishing House, Jordan, first edition. (in Arabic).
- AL-Hadad M. T. S., Kadhim R. A., and Al-Rubaye A.F. (2019). Effect of Chronic Toxoplasmosis on Levels of Some Neurotransmitters (Dopamine, Adrenaline, and Noradrenaline) in Human Serum. *Journal of Pharmaceutical Sciences and Research*. Vol. 11(2), 402-405.
- Al-Jebouri, M., Al-Janabi, M. and Ismail, H. (2013). The prevalence of Toxoplasmosis among female patients in Al-Hawija and Al-Baiji Districts in Iraq. *Open. Epidem*. 3(2):85-88.
- AL-Juburi, G., Muhsin, M., Al-Saeed, M. (2008) Study the role of Toxoplasmosis Cytomegalovirus ant phospholipids in Cases of abortion among women in Hilla city. *Al-Qadisiah Medical Journal*, 4(2):27-34.
- Al-Kalaby, R. F. (2008). Sero-epidemiological study of Toxoplasmosis among different groups of population in Najaf city, M.Sc. thesis College Medicine. Kufa University. Iraq.
- Al-Malki, E. S. (2021). Toxoplasmosis: stages of the protozoan life cycle and risk assessment in humans and animals for an

enhanced awareness and an improved socio-economic status. *Saudi Journal of Biological Sciences*, 28(1): 962-969.

- Almasian, R., Almasian, M. and Zibaei, M. (2014) Sero-Epidemiology of Toxoplasmosis among the People of Khorram Abad, Iran. *J Infect Dis Ther* 2(5): 159.
- AL-Mayahi J. R. Gzar (2011) Epidemiological Study on *Toxoplasma gondii* in aborted women in Kut city, Master thesis, University of Baghdad, Iraq. pp:125.
- Al-Obaidi, G. F. (2011) Effect of *Toxoplasma gondii* on some serum biochemical levels and blood contents of infected pregnant women in Mosul. *Journal of Education and Science* 24 (4): 50-55 (In Arabic).
- Al-Qassab, S., Reichel, M. P., Su, C., Jenkins, D., Hall, C., Windsor, P.A., Dubey, J. P., Ellis, J. (2009). Isolation of *Toxoplasma gondii* from the brain of a dog in Australia and its biological and molecular characterization. Vet. Parasitol. 164(2-4):335–339.
- Alsaady, A.M., Al-Abboodi, A. and Abood, E. (2021) Seroepidemiology of *Toxoplasma gondii* among men and pregnant women in Maysan Province, south of Iraq. *Iranian Journal of Ichthyology* 8: 27-37.
- AL-Sadoon, M.A., Nasir, M.A., Yasir, E.T., Khalaf, A.O. and Kadim, S.J. (2018). Toxoplasmosis and risk factors among female students of medical colleges at Basra University, Iraq. *Biomedical and Pharmacology Journal* 11(4): 2117-2122.
- Al-Sray, A.H., Sarhan, S.R. and Mohammed, H.A. (2019). Molecular and serological characterization of *Toxoplasma gondii*

in women in Wasit Province. *Advances in Animal and Veterinary Sciences* 7(8): 657-663.

- AL-Ubaydi, G. T. (2004). Toxoplasmosis in pregnant women and its relation with some parameters. M.Sc. Thesis. College of Science. Mosul University, Iraq. pp 127 (In Arabic).
- Alvarado-Esquivel C., Liesenfeld O., Torres-Castorena A., Estrada-Martínez S., Urbina-Alvarez J.D., Ramos-de la Rocha M., Márquez-Conde J.A., and Dubey J.P. (2010). Seroepidemiology of *Toxoplasma gondii* infection in patients with vision and hearing impairments, cancer, HIV, or undergoing hemodialysis in Durango, Mexico. *J Parasitol*. 96:505-508.
- Alvarado-Esquivel C., Mercado-Suarez M.F., Rodriguez-Briones A., Fallad-Torres L., Ayala-Ayala J.O., Nevarez-Piedra L.J., Duran-Morales E., Estrada-Martinez S., Liesenfeld O., Marquez-Conde J.A., and Martinez-Garcia S.A. (2007) Seroepidemiology of infection with *Toxoplasma gondii* in healthy blood donors of Durango, Mexico. *BMC Infect* Dis 7: 75.
- Alvarado-Esquivel C., Rascón-Careaga, A., Hernández-Tinoco, J., Guadalupe Corella-Madueño, M. A., Sánchez-Anguiano, L. F., Aldana-Madrid, M. L., Almada-Balderrama, G. J., Nuñez-Aguirre, A. D., and Liesenfeld, O. (2016). Seroprevalence and correlates of *Toxoplasma gondii* infection in Yoremes (Mayos) in Mexico: a cross-sectional study. *BMJ Open*, 6(5): e010218.
- Alvarado-Esquivel C., Sánchez-Anguiano, L. F., Hernández-Tinoco, J., Ramos-Nevarez, A., Estrada-Martínez, S., Cerrillo-Soto, S. M., Mijarez-Hernández, M. A., Guido-Arreola, C. A., Pérez-Álamos, A. R., Beristain-Garcia, I., and Rábago-Sánchez,

E. (2018a). *Toxoplasma gondii* Infection and a History of Surgery: A Case Control Seroprevalence Study. *European Journal of Microbiology and Immunology*, 8(4):155-158.

- Alvarado-Esquivel, C., Alanis-Quiñones, P., Arreola-Valenzuela, Á., Rodríguez-Briones, A., Piedra-Nevarez, J., Duran-Morales, E., Estrada-Martínez, S., Martínez-García, A., and Liesenfeld, O. (2006). Seroepidemiology of *Toxoplasma gondii* infection in psychiatric inpatients in a northern Mexican city. *BMC Infectious Diseases*, 6: 178.
- Alvarado-Esquivel, C., Estrada-Martínez, S., and Liesenfeld, O. (2011) *Toxoplasma gondii* infection in workers occupationally exposed to unwashed raw fruits and vegetables: a case control seroprevalence study. *Parasites & Vectors* 4:235
- Alvarado-Esquivel, C., Pacheco-Vega, S. J., Salcedo-Jaquez, M., Sánchez-Anguiano, L. F., Hernández-Tinoco, J., Rábago-Sánchez, E., Centeno-Tinoco, M. M., Flores-Garcia, I. D., Ramos-Nevarez, A., Cerrillo-Soto, S. M., Guido-Arreola, C. A., Beristain-García, I., Liesenfeld, O., Berumen-Segovia, L. O., Saenz-Soto, L., and Sifuentes-Álvarez, A. (2015). Stillbirth history and *Toxoplasma gondii* infection in women attending public health centers in a northern Mexican City. *European Journal of Microbiology & Immunology*, 5(2):164-171.
- Aqeely H., El-Gayar E K., Khan D P., Najmi A., Alvi A., Bani I., Mahfouz M S., Abdalla S E. and Elhassan I M. (2014) Seroepidemiology of *Toxoplasma gondii* amongst Pregnant Women in Jazan Province, *Saudi Arabia Journal of Tropical Medicine* Volume 2014, 6 pages

- Arnall D.A., Marker J.C., Conlee R.K., and Winder W.W. (1986). "Effect of infusing epinephrine on liver and muscle glycogenolysis during exercise in rats". *The American Journal of Physiology*. 250 (6 Pt 1): E641–E649.
- Attias M., Teixeira D. E., Benchimol M., Vommaro R. C., Crepaldi P. H. and De Souza W. (2020) The life-cycle of *Toxoplasma gondii* reviewed using animations. *Parasites & Vectors* (2020) 13:588
- Ayi, I., Edu, A., Apea-Kubi, K., Boamah, D., Bosompem, K., and Edoh, D. (2009) Sero-epidemiology of toxoplasmosis amongst pregnant women in the greater Accra region of Ghana. *Gh Med J*. 43: 107-114.
- Bak, J., Shim, S.H., Kwon, Y.J., Lee, H.Y., Kim, J.S., Yoon, H., Lee, Y.J., (2018). The association between suicide attempts and *Toxoplasma gondii* infection. *Clin Psychopharmacol Neurosci* 16 (1): 95–102.
- Baliki M.N., Mansour A., Baria A.T., Huang L., Berger S.E., Fields H.L., Apkarian A.V. (2013). Parceling human accumbens into putative core and shell dissociates encoding of values for reward and pain. *The Journal of Neuroscience*. 33 (41): 16383–16393.
- Barbosa C.J., Molina R.J., de Souza M.B., Silva A.C., Micheletti A.R., dos Reis M.A., de Paula Antunes Teixeira V., and Silva-Vergara M.L. (2007). Disseminated Toxoplasmosis presenting as sepsis in two AIDS patients. *Rev. Inst. Med. Trop. Sao-paulo*. 49(2): 113-116.

- Basu S. and Dasgupta P.S. (2000) Dopamine, a neurotransmitter, influences the immune system. *J Neuroimmunol* 102 (2): 113-124.
- Bay-Richter, C., Buttenschon, H.N., Mors, O., Eskelund, A., Budac, D., Kaerlev, L., and Wegener, G., (2019). Latent toxoplasmosis and psychiatric symptoms - a role of tryptophan metabolism? *J. Psychiatr. Res*. 110, 45–50.
- Behbehani, K. and Al-Karmi, T. (1980) Epidemiology of toxoplasmosis in Kuwait. I. Detection of antibodies to *Toxoplasma gondii* and percentage distribution among the inhabitants. *Transactions of the Royal Society of Tropical Medicine and Hygiene* 74(2): 209-212.
- Bell D.R. (2009). Medical physiology: principles for clinical medicine (3rd ed.). Philadelphia: Lippincott Williams and Wilkins. p. 312.
- Berdoy M., Webster J.P. and Macdonald D.W. (2000) Fatal attraction in rats infected with *Toxoplasma gondii*. *Proceedings Biological Sciences* 267(1452): 1591–1594.
- Berger, F., Goulet, V., Le Strat, Y. and Desenclos, J. (2009). Toxoplasmosis among pregnant women in France: risk factors and change of prevalence between 1995 and 2003. *Rev. Epidemiol. Sante. Publique*57 (4) :241–248.
- Berridge K.C. (2007). The debate over dopamine's role in reward: the case for incentive salience. *Psychopharmacology* 191 (3): 391–431.

- Best, J. A., Nijhout, H. F., and Reed, M. C. (2009). Homeostatic mechanisms in dopamine synthesis and release: a mathematical model. Theoretical Biology & Medical Modelling, 6, 21.
- Bhopale, G. M. (2003). Pathogenesis of toxoplasmosis. Company Immunol. *Microb. Infect. Dist*. 26 :213-222.
- Bigna J.J., Tochie J. N., Tounouga D. N., Bekolo A. O., Ymele N. S., Youda E. L., Sime P. S. and Nansseu J. R. (2020) Global, regional, and country seroprevalence of *Toxoplasma gondii* in pregnant women: a systematic review, modelling and met analysis. *Scientific Reports* (2020) 10:12102.
- Björklund, A., and Dunnett, S.B. (2007). Dopamine neuron systems in the brain: an update. *Trends in Neurosciences* 30 (5): 194–202.
- Black, M. W., and Boothroyd, J. C. (2000). Lytic Cycle of *Toxoplasma gondii*. *Microbiol. Mol. Bio. Rev*, 64(3): 607-623.
- Boillat, M., Hammoudi, P.M.; Dogga, S.K.; Pagès, S.; Goubran, M.; Rodriguez, I., and SoldatiFavre, D. (2020). Neuroinflammation associated a specific manipulation of mouse predator fear by *Toxoplasma gondii*. *Cell Rep*, 30(2), 320-334.
- Bonfioli, A. A., and Orefice, F. (2005). Toxoplasmosis. *Semin. Ophthalmol* 20 (3):129-141.
- Boothroyd, J.C., and Grigg, M.E. (2002) Population biology of *Toxoplasma gondii* and its relevance to human infection: Do different strains cause different disease? *Current Opinion. Microbiol*.5 (4):438–442.
- Brown H.F., DiFrancesco D., and Noble S.J. (1979). How does adrenaline accelerate the heart? *Nature*. 280 (5719): 235–236.

- Buffolano, W., Gilbert, R., Holland, F., Fratta, D., Palumbo, F., and Ades, A. (1996). Risk factors for recent *Toxoplasma* infection in pregnant women in Naples. *Epidemiology and infection.* 116(3): 347- 351.
- Cabral, C.M., Tuladhar, S., Dietrich, H.K., Nguyen, E., MacDonald, W.R., Trivedi, T., Devineni, A., and Koshy, A.A. (2016). Neurons are the primary target cell for the brain-tropic intracellular parasite *Toxoplasma gondii*. *PLoS. Pathog*., 12(2), e1005447.
- Carme, B., Demar, M., Ajzenberg, D., and Darde, M.L. (2009). Severe acquired toxoplasmosis caused by wild cycle of *Toxoplasma gondii*, French Guiana. *Emerg. Inf. Dis.* 15(4):656–658.
- Carmo M.E. do and Guizardi F.L. (2018) The concept of vulnerability and its meanings for public policies in health and social welfare. *Cadernos de Saúde Pública*. 34(3): e00101417.
- Carvalho M.C., Ribeiro-Andrade M., Melo R.P.B., Guedes D.M., Pinheiro Junior J.W., Cavalcanti EFTSF., Magalhães F.J.R.3, and Mota R.A. (2021). Cross-sectional survey for *Toxoplasma gondii* infection in humans in Fernando de Noronha island, Brazil. *Braz J Vet Parasitol*; 30(3): e005121.
- Çelik, T., Kartalci, S., Aytaş, Ö., Akarsu, G. A., Gözükara, H. and Ünal, S. (2015) Association between latent toxoplasmosis and clinical course of schizophrenia – continuous course of the disease is characteristic for *Toxoplasma gondii*-infected patients. *Folia Parasitological* 62: 015:1-6.

- Cenci-Goga, B. T., Rossitto, P. V., Sechi, P., McCrindle, C. M., and Cullor, J. S. (2011). *Toxoplasma* in animals, food, and humans: an old parasite of new concern. *Foodborne. Path. Dis*. 8(7): 751-762.
- Channon J.Y., and Kasper L.H. (1996) *Toxoplasma gondii*-induced immune suppression by human peripheral blood monocytes: role of gamma interferon. *Infect Immun*. 64: 1181–1189.
- Charles, E. (2009). Coccidial infections in cats and dogs. *Briti. Med. J*; 321(7254):142–147.
- Coccaro, E.F., Lee R., Groer, M.W., Can, A., Coussons-Read, M., and Postolache, T.T., (2016). *Toxoplasma gondii* infection: relationship with aggression in psychiatric subjects. *J. Clin. Psychiatr*. 77 (3): 334–341.
- Coelho R.A., Kobayashi M., and Carvalho Jr., L.B. (2003) Prevalence of IgG antibodies specific to *Toxoplasma gondii* among blood donors in Recife, Northeast Brazil. *Rev Inst Med Trop Sao Paulo* 45 (4): 229-231.
- Cook, A.J., Gilbert, R.E., Buffolano, W., Zufferey, J., Petersen, E., Jenum, P. A.; Foulon, W., Semprini, A.E., and Dunn, D.T. (2000). Sources of *Toxoplasma* infection in pregnant women: European multicentre case-control study. European Research Network on Congenital Toxoplasmosis. *Brit. Med.J.* 321(7254):142–147.
- Correa, D., Canedo-Solares, I., Ortiz-Alegrĺ, L. B., Caballero-Ortega, H., and Rico-Torres, C.P. (2007). Congenital and acquired Toxoplasmosis: diversity and role of antibodies in

different compartments of the host. *Parasit. Immunol* 29(12):651-660.

- Cosentino, M. and Marino, F. (2012). Nerve Driven Immunity: Noradrenaline and Adrenaline. In: Levite, M. (eds) Nerve-Driven Immunity. Springer, Vienna. pp 47–96.
- Costa, A. J. (2007). Isolation of *Toxoplasma gondii* from goat semen. Abstract book WAAVP Congress, Ghent, 2007.
- Cox, R. M. and John-Alde, H.B. (2005). Testosterone has opposite effects on male growth in lizard (Sceloporus spp.) with opposite patterns of sexual size dimorphism. *J. Exp. Biol*.208 (24): 4679-4687.
- Cuevas J. (2019). Neurotransmitters and Their Life Cycle. *Reference Module in Biomedical Sciences*. 2007:1-7.
- Dabritz, H. A.; Gardner, I. A.; Miller, M. A.; Lappin, M. R.; Atwill, E. R.; P ackham, A. E.; Melli, A. C.; and Conrad, P. A. (2007). Evaluation of two *Toxoplasma gondii* serotilogic tests used in a serosurvey of domestic cats in California. *J. Parasitol*. 93(4):806-816.
- Đaković-Rode O., Židovec-Lepej S., Vodnica Martucci M., Lasica Polanda V., and Begovac J. (2010). Prevalence of antibodies against *Toxoplasma gondii* in patients infected with human immunodeficiency virus in Croatia. *Croat. J. Infect*. 30:5–10.
- Darde´, M. L., Bouteille, B. and Pestre-Alexandre, M. (1987). Differenciation iso-enzymatique de 7 souches de *Toxoplasma gondii* par iso-electrofocalisation en gel de polyacrylamide. *Bull. Soc. Fr. Parasitol*. 5:33–39.

- Darweesh, N.H., Hussein, R.A., Salman, S.T. and Shaker, M.J. (2018) Immunological and Molecular study of *Toxoplasma gondii* from aborted women in Diyala / Iraq. *J. Ilam Uni Med Sci*.2(6):75-82.
- Daryani A., Sarvia S., Aarabib M., Mizanic A., Ahmadpourc E., Shokric A., Rahimi M.–T. and Sharifa M. (2014). Seroprevalence of *Toxoplasma gondii* in the Iranian general population: A systematic review and meta-analysis; *Acta Tropica*, 137:185-194.
- David, F.L., Jason, S. S., Andew, E.G., Adeeb, H. R., Devon, K.T., Christopher, A.H., and Laurence, A.T. (2008). T cell expression of my D88 is required for resistance to *Toxoplasma gondii. Proc. Na tl. Acad. USA*. 105(10): 3855-3860.
- De Bles, N., Van Der Does, J., Kortbeek, L., Hofhuis, A., Van Grootheest, G., *et al*. (2021) *Toxoplasma gondii* seropositivity in patients with depressive and anxiety disorders. *Brain, Behavior, & Immunity - Health* 11(34):100197
- De Paschale, M., Agrappi, C., Manco, M. T., Cerulli, T., and Clerici, P. (2010). Implementation of Screening for *Toxoplasma gondii* Infection in Pregnancy. *J. Clin Med Res*. 2(3), 112-116.
- Delgado Betancourt E., Hamid B., Fabian B.T., Klotz C., Hartmann S. and Seeber F. (2019) From Entry to Early Dissemination—*Toxoplasma gondii*'s Initial Encounter with Its Host. *Front. Cell. Infect. Microbiol*. 9:46.
- Desmonts, G., Naot, Y., and Remington, J.S. (1981). Immunoglobulin M-immunosorbent agglutination assay for diagnosis of infectious diseases: diagnosis of acute congenital and

acquired *Toxoplasma* infections. *J. Clin. Microbiol*. 14(5):486–491.

- Di Genova B. M., Wilson, S.K., Dubey, J.P., and Knoll, L.J. (2019) Intestinal delta-6-desaturase activity determines host range for *Toxoplasma* sexual reproduction. *PLoS Boil* 17(8): e3000364.
- Dickerson F., Stallings C., Origoni A., Katsafanas E., Schweinfurth L., Savage C., Khushalani S. and Yolken R. (2014) Antibodies to *Toxoplasma gondii* and cognitive functioning in schizophrenia, bipolar disorder, and nonpsychiatric controls. *Journal of Nervous and Mental Disease* 202(8): 589–593.
- Djurkovic-Djakovic O., Dupouy-Camet J., Giessen J. V., and Dubey J. P. (2019) Toxoplasmosis Overview from a One Health Perspective. *Food and Water borne Parasitology*; 15, e0054.
- Dubey J. P. (1996). *Toxoplasma Gondii*. Chapter 84, In: Baron S, (ed.) Medical Microbiology. 4th edition. Galveston (TX): University of Texas Medical Branch at Galveston.
- Dubey J.P. and Jones J.L. (2008) *Toxoplasma gondii* infection in humans and animals in the United States. *Int J Parasitol* 38(11):1257–1278.
- Dubey, J. (2004) Toxoplasmosis-a waterborne zoonosis. *Vet. Parasitol*. 126(1-2): 57-72.
- Dubey, J. P. (1998). *Toxoplasma gondii* oocyst survival under defined temperatures. *J. Parasitol*. 84 (4) 862-865.
- Dubey, J. P. (2000). Sources of *Toxoplasma gondii* infection in pregnancy. Until rates of congenital toxoplasmosis fall, control measures are essential. *Brit.Med.J*.321(7254):127-128.

- Dubey, J. P. (2002). Tachyzoite-induced life cycle of *Toxoplasma gondii* in cats. *J. Parasitol*. 88(4):713–717.
- Dubey, J. P. (2007). The history and life cycle of *Toxoplasma gondii*. In: Weiss, L. M. & Kim, K. (ed.), *Toxoplasma gondii*. The Model Apicomplexan: Perspectives and Methods. Academic Press, New York. p. 1–17.
- Dubey, J. P., Graham, D. H., Blackston, C. R., Lehmann, T., Gennari, S. M., Ragozo, A. M. A., Nishi, S. M., Shen, S. K., Kwok, O. C. H., Hill, D. E., and Thulliez, P. (2002). Biological and genetic characterization of *Toxoplasma gondii* isolates from chickens (Gallus domesticus) from Sa˜o Paulo, Brazil: unexpected findings. *Int. J. Parasitol*., 32(1):99–105.
- Dubey, J. P., Lunney, J. K., Shen, S. K., Kwok, O. C. H., Ashford, D. A., and Thulliez, P. (1996). Infectivity of low numbers of *Toxoplasma gondii* oocysts to pigs. *J. Parasitol*., 82(3):438–443.
- Dubey, J.; Beattie, C. (1988). General Biology. In toxoplasmosis of Animals and Man, CRC Press, Inc., Boca Raton, Florida, pp. 1-40.
- Dubey, J.P. (2010). Toxoplasmosis of Animals and Humans, vol. 313. Boca Raton: CRC Press; 2010.
- Dubey, J.P. and Carpenter, J. L. (1993). Histologically confirmed clinical Toxoplasmosis in cats: 100 cases (1952–1990). *J. Am. Vet. Med Assoc*. 203(11):1556–1566.
- Dubey, J.P., (2009). History of the discovery of the life cycle of *Toxoplasma gondii*. *Int. J. Parasitol*. 39(8): 877–882.
- Dubey, J.P., and Jones, J.L. (2014). Comments on “detection of *Toxoplasma gondii* in raw caprine, ovine, buffalo, bovine, and

camel milk using cell cultivation, cat bioassay, capture ELISA, and PCR methods in Iran." *Foodborne Pathogens and Disease* 11: 500-501.

- Dubey, J.P., Darrington, C., Tiao, N.; Ferreira, L. R., Choudhary, S., Molla, B., *et al*. (2013). Isolation of viable *Toxoplasma gondii* from tissues and feces of cats from Addis Ababa, Ethiopia. *J. Parasitol*. 99(1): 56–58.
- Dubey, J.P., Thayer, D.W., Speer, C.A., and Shen, S.K., (1998). Effect of gamma irradiation on Unsporulated and sporulated *Toxoplasma gondii* oocysts. *Int. J. Parasitol*. 28(3): 369–375.
- Dubremetz, J.F. and Lebrun, M. (2012). Virulence factors of *Toxoplasma gondii*. *Microbes Inf*. 14(15):1403–1410.
- Duffy, A.R., Beckie, T.M., Brenner, L.A., Beckstead, J.W., Seyfang, A., Postolache, T.T., and Groer, M.W. (2015). Relationship between *Toxoplasma gondii* and mood disturbance in women veterans. *Mil. Med*. 180 (6), 621–625.
- Dumetre, A., Aubert, D., Puech, P.H., Hohweyer, J., Azas, N., and Villena, I. (2012). Interaction forces drive the environmental transmission of pathogenic protozoa. *Appl. Environ. Microbiol*. 78(4): 905–912.
- Dupon, M., Cazenave, J. Pellegrin, J., Ragnaud, J., A. Cheyron, A., Fischer, I., Leng, B., and Lact, J. (1995). Detection of *T. gondii* by PCR and tissue culture in cerebrospinal fluid and blood of human immunodeficiency virus-seropositive patient. J. Clin. Microbiol. 33(9): 2421-2426.

- Dzitko, K., Malicki, S. and Komorowski, J. (2008) Effect of hyperprolactinaemia on *Toxoplasma gondii* prevalence in humans. *Parasitol. Res*. 102(4):723-729.
- Eisenhofer G., Kopin I.J., and Goldstein D.S. (2004). Catecholamine metabolism: a contemporary view with implications for physiology and medicine. *Pharmacological Reviews.* 56 (3): 331–349.
- El Mansouri, B., Rhajaoui, M., Sebti, F., Amarir, F., Laboudi, M., Bchitou, R., Lyagoubi, M. (2007). Seroprevalence of toxoplasmosis in pregnant women in Rabat, Morocco. *Bull. Soc. Pathol. Exot* 100(4), 289-290.
- Elsheikha H. M. and Jarroll E. L. (2017) Illustrated Dictionary of Parasitology in the Post- Genomic Era. Caister Academic Press UK.
- Elsheikha H.M. (2008) Congenital toxoplasmosis: priorities for further health promotion action. *Public Health* 122(4):335–353.
- Elsheikha H.M., Azab M.S., Abousamra N.K., Rahbar M.H., Elghannam D.M., and Raafat D. (2009) Seroprevalence of and risk factors for *Toxoplasma gondii* antibodies among asymptomatic blood donors in Egypt. *Parasitol Res* 104(6): 1471-1476.
- Ferreira, M. S. and Borges, A. S. (2002). Some aspects of protozoan infections in immunocompromised patients- a review. *Mem. Inst. Oswaldo. Cruz* 97(4): 443-457.
- Filisetti, D. and Candolfli, E. (2004). Immune response to *Toxoplasma gondii*. *Ann. Ist. Supr. Sanita*., 40(1): 71 – 80.

- Flegr J. (2007). Effects of *Toxoplasma* on Human Behaviour. *Schizophrenia Bulletin* 33 (3): 757–760.
- Flegr J. (2013) How and why *Toxoplasma* makes us crazy. *Trends in Parasitology* 29(4): 156–163.
- Flegr J. (2015) Host Manipulation by *Toxoplasma gondii*. pp. 91-99. In: Mehlhorn H, (ed.) Host Manipulations by Parasites and Viruses. Springer, London UK.
- Flegr J., Prandota J. h., Sovickova M. and Israili Z. H. (2014) Toxoplasmosis – A Global Threat. Correlation of Latent Toxoplasmosis with Specific Disease Burden in a Set of 88 Countries. *Plos One* 9 (3): e 90203.
- Flegr, J. and Kaňková, Š. (2020). The effects of toxoplasmosis on sex ratio at birth. *Early Human Development* 141: 104874.
- Flori, P., Bellete, B., Crampe, C., Maudry, A., Patural, H., Chauleur, C., Hafid, J., Raberin , H., and Tran ManhSung, R. (2008). A technique for dating Toxoplasmosis in pregnancy and comparison with the Vidas anti *Toxoplasma* IgG avidity test. *Clin. Microbial. Infect.*, 14(3):242-249.
- Foroutan-Rad, M., Majidiani, H., Dalvand, S., Daryani A., Kooti W., Saki J., Hedayati-Rad F. and Ahmadpour E. (2016). Toxoplasmosis in Blood Donors: A Systematic Review and Meta-Analysis. *Transfus Med Rev*.;30(3):116-22.
- Frenkel, J. K. (2000). Biology of *Toxoplasma gondii*. pp. 9-25. In: P. Ambroise-Thomas (eds.). Congenital Toxoplasmosis: Scientific Background, Clinical Management and Control. Paris: Springer-Verlag France.

- Frenkel, J. K. (1967). Adoptive immunity to intracellular infection. *J. Immunol*., 98(6):1309–1319.
- Frenkel, J. K. (1973). *Toxoplasma* in and around us. *BioScience*. 23(6):343–352.
- Freppel, W., Ferguson, J.P., Shapiro, K., Dubey, J.P., Puech, P.H., and Dumètre, A., (2019). Structure, composition, and roles of the *Toxoplasma gondii* oocyst and sporocyst walls. *Cell Surf*. 5: 100016.
- Gale, S.D., Brown B.L., Erickson L.D., Berrett A. and Hedges D.W. (2015) Association between latent toxoplasmosis and cognition in adults: a cross sectional study. *Parasitology* 142(4): 557–565.
- Garcia L. S. (2007) Diagnostic medical parasitology. 5th edition. American Society for Microbiology, Washington, USA.
- Gascon, J., Torres-Rodriguez, J. M., Soldevila, M., and Merlo, A. M. (1989). Seroepidemiology of Toxoplasmosis in 2 communities of Rwanda (Central Africa). Rev. *Inst. Med. Trop. Sao Paulo*. 31(6): 399-402.
- Gaskell E.A., Smith J.E., Pinney J.W., Westhead D.R., and McConkey G.A. (2009) A Unique Dual Activity Amino Acid Hydroxylase in *Toxoplasma gondii*. *PLoS ONE* 4(3): e4801.
- Gazzinelli, R. T., Hakim, F. T., Hieny, S., Shearer, G. M., and Sher, A. (1991). Synergistic role of CD41 and CD81 T lymphocytes in IFN-gamma production and protective immunity induced by an attenuated *Toxoplasma gondii* vaccine. *J. Immunol*., 146(1):286–292.

- Giannoulis, C.B., Zournatzi, B., Giomisi, A., Diza, E., and Tzafettas, I. (2008). Toxoplasmosis during pregnancy: a case report and review of the literature. *Hippokratia* 12(3):139-143.
- Gilbert, R. E. and Stanford, M. R. (2000). Is ocular Toxoplasmosis caused by prenatal or postnatal infection. *Br. J. Ophthalmo*.84:224-226.
- Gilbert, R.E., Dunn, D.T., Lightman, S., Murray, P.I., Pavesio, C.E., Gormley, P.D., Masters, J., Parker, S.P., and Stanford, M.R. (1999) Incidence of symptomatic *Toxoplasma* eye disease: aetiology and public health implications. *Epidemiol. Infect*. 123(2): 283–289.
- Gras, L., Gilbert, R. E., Wallon, M., Peyron, F., and Cortina-Borjia, M. (2004). Duration of the IgM response in women acquiring *T. gondii* during pregnancy: implication for clinical and cross- sectional incidence studies. *Epidemiol. Infect*. 132:541-548.
- Gross, U. and Bohne, W. (1994). *Toxoplasma gondii*: strain- and host cell-dependent induction of stage differentiation. *J. Eukaryote. Microbiol*. 41(5): 10-11.
- Grossklaus, D. and H. J. Baumgarten (1968). Die Uberlebensdauer von Toxoplasma-Cysten. 48: 930-932. In: I. Schweinefl- leisch (ed.). Mitteilung: Ergebnisse von Lagerungs-versuchen beiverschied- enen, Temperaturen. Fleischwirtschaft.
- Hadi, H.S.; Kadhim, R.A. and Al-Mammori, R.T. (2016). Seroepidemiological aspects for *Toxoplasma gondii* infection in women of Qadisiyah province, Iraq. *International Journal of PharmTech Research* 9(11): 252-259.

- Hakko, E., Ozkan, H. A., Karaman, K. and Gulbas, Z. (2013) Analysis of cerebral toxoplasmosis in a series of 170 allogeneic hematopoietic stem cell transplant patients. *Transpolar. Infect. Dis*. 15(6):575–580.
- Hamer M., Gale C.R., Kivimäki M., and Batty G. D. (2020). Overweight, obesity, and risk of hospitalization for COVID-19: a community-based cohort study of adults in the United Kingdom. *Proc Natl Acad Sci USA*.;117(35):21011–21013.
- Han, K., Shin, D.W., Lee, T.Y. and Lee, Y.H. (2008). Seroprevalence of *Toxoplasma gondii* infection and risk factors associated with seropositivity of pregnant women in Korea. *Journal of Parasitology* 94(4): 963-965.
- Harrison S. L., Fazio-Eynullayeva E., Lane D.A., Underhill P., and Lip G. Y. H. (2020). Comorbidities associated with mortality in 31,461 adults with COVID-19 in the United States: a federated electronic medical record analysis. *PLoS Med*.;17(9): e1003321.
- Hassen, A.H., Ali, M.S., and Ekhnafer, A.M. (2019). Effect of *Toxoplasma gondii* Infection on Haematological and liver function parameters among abortive women in El-Beida City. *Saudi J. Biomed. Res*., 4:295-303.
- Hill, D.E., Sreekumar, C., Jones J., and Dubey J.P. (2007) *Toxoplasma gondii*. Ch 12 In: Simjee S (ed.) Foodborne diseases. Humana Press, Totowa, p. 337–353
- Howe, D.K. and Sibley, L. D. (1995) *Toxoplasma gondii* comprises three clonal lineages: Correlation of parasite genotype with human disease. *J. Infect. Dis*. 172(6):1561–1566.

- Ho-Yen, D. O. and Joss A. W.L. (1992). Human Toxoplasmosis. Oxford Univ., Press.pp:1-25.
- Hughes D. (2013) Pathways to understanding the extended phenotype of parasites in their hosts. *J Exp Biol*. 216(1):142–147.
- Hussein, N. and Balatay, A.A. (2019). The Seroprevalence of *Toxoplasma*, Cytomegalovirus and Rubella Infections in Women with Abortion in Kurdistan Region of Iraq: A Brief Report. *International Journal of Infection* 6(1): e86734.
- Ibrahim A. M., Abdel Gawad M. I. M., Abd-Elftah A.-A. G., Abdel-Latif M., Mohamed S. R., Salah H., Sayed A. G. S, and Abu-Sarea E. Y. (2020) Toxoplasmosis in schizophrenic patients: Immun-diagnosis and serum dopamine level. *Pak J Biol Sci*.; 23(9):1131-1137.
- Jakeman G.N., Saul A., Hogarth W.L., and Collins W.E. (1999). Anaemia of acute malaria infections in non-immune patients primarily results from destruction of uninfected erythrocytes. *Parasitology*;119 (2):127-133.
- Javadi S., Rezaei, S.A., Tajik, T., Hadian, M., and Shokouni, F. (2010). Hematological changes of cats with *Toxoplasma gondii*-Specific antibodies. *Comparative Clinical Pathology* 19:307–310.
- Johnson, A. M. (1997). Speculation on possible life cycles for the clonal lineages in the genus *Toxoplasma*. *Parasitol. Today*. 13(10): 393-397.
- Jones J. L., Dargelas V., Roberts J., Press C., Remington J.S. and Montoya J. G. (2009) Risk Factors for *Toxoplasma gondii*

Infection in the United States. *Clinical Infectious Diseases* 49(6):878–884.

- Jones, J. L., (2006). Recently acquired *Toxoplasma gondii* Infection, Brazil. *Emerg. Infect. Dis*., 12(4):582-582.
- Jones, J. L., Kruszon-Moran, D., Wilson, M., McQuillan, G., Navin, T., and McAuley, J. B. (2001). *Toxoplasma gondii* infection in the United States: seroprevalence and risk factors. *Am. J. Epidemiol*. 154(4): 357-365.
- Jonies, T. C., Hunt, R. D., and King, N. W. (1997). Veterinary Pathology. 6th ed. Lippin Cott Williams & Wilkins, Awolters Kluwer Company, USA, pp. 555- 560.
- Juanah, L. Y., Jalaludin, J., Osman, M. and Osman, Z. J. (2013) Seroprevalence of *Toxoplasma gondii* Among Schizophrenics at Hospital Kajang. *American Journal of Infectious Diseases* 9 (1): 11-16.
- Kadhim R.A. Al-Rubaye Abeer F. and Mahdi T. S. (2020). Etiology of Parkinson's disease: *Toxoplasma* parasite as a model. *AIPConference Proceedings* 2290(1): 020012-1–020012-6.
- Kadhim R.A. and Al-awadi H.M., (2013). Seroprevalence of *Toxoplasma gondii* Antibodies Among Pregnant Women in Babylon Province, Iraq. *Kufa Journal for Nursing Sciences* 3(3): 153-159.
- Kaňková, Š. And Flegr, J. (2007). Longer pregnancy and slower fetal development in women with latent" asymptomatic" toxoplasmosis. *BMC Infectious Diseases* 7(1): 1-7.
- Kapperud, G., Jenum, P. A., Stray-Pedersen, B., Melby, K.K., Eskild, A., and Eng, J. (1996). Risk factors for *Toxoplasma*

gondii infection in pregnancy: results of a prospective case-control study in Norway. *Am. J. Epidemiol.*144(4): 405-412.

- Karem, L. O. M. (2007). *Serological study of Toxoplasma gondii antibody from aborted women in Sulaimania city by using ELISA and Minividas test*. M.Sc. Thesis. College of Science University of Baghdad. pp 126 (In Arabic).
- Karimi C., Mardani A., and Zadsar M. (2014). *Toxoplasma* and Blood Transfusion. *Iranian Journal of Parasitology*, 9(1):597-598.
- Kasper, L. H. (2005). *Toxoplasma* infection. 243-1248. In: D. L. Kasper (ed.). Harrison principles of internal medicine. 16 th ed. McGraw-Hill Companies, U.S.A.
- Khalili, M., Mahami-Oskouei, M., Shahbazi, A., Safaiyan, A., Mohammadzadeh-Gheshlaghi, N., and Mahami-Oskouei, L. (2018). The correlation between serum levels of anti-*Toxoplasma gondii* antibodies and the risk of diabetes. *Iranian Journal of Parasitology*, 13(4): 637-642.
- Khurana, I. (2008). Essentials of Medical Physiology. Elsevier India. p. 460.
- Kotula, A. W., Dubey, J. P., Sharar, A. K., Andrew, C. D., Shen, S. K. and Lindsay, D. S. (1991). Effect of freezing on infectivity of *Toxoplasma gondii* tissue cysts in pork. *J. Food Protection* 54(9):687–690.
- Kuticic, V. and T. Wikerhauser (1996). Studies of the effect of various treatments on the viability of *Toxoplasma gondii* tissue cysts and oocysts. pp. 261-265. In: U. Gross (ed.). *Toxoplasma gondii*. Springer-Verlag. Berlin.

- Lachkhem A., Lahmar I., Galal L., Babba O., Mezhoud H. Hassine M. Ahmed Lachkhem A., Dardé M-L., Mercier A. and Babba H. (2020). Seroprevalence of *Toxoplasma gondii* among healthy blood donors in two locations in Tunisia and associated risk factors. *Parasite* 27 (51):9.
- Lieberman M., Marks A., and Peet A. (2013). Marks' Basic Medical Biochemistry: A Clinical Approach (4th ed.). Philadelphia: Wolters Kluwer Health/Lippincott Williams & Wilkins. p. 175.
- Liesenfield, O., Press, C., Montoya, J. G., Gill, R., Isaac-Renton, J. L., Hedman, K., and Remington, J.S. (1997). False positive results in immunoglobulin M(IgM) *Toxoplasma* antibody tests and importance of confirmatory testing the related *Toxoplasma* IgM test. *J. Clin. Microbiol*.; 135(1): 174-178.
- Lindová, J., Příplatová, L., and Flegr, J. (2012). Higher extraversion and lower conscientiousness in humans infected with *Toxoplasma*. *Eur. J. Personal*. 26 (3), 285–291.
- Ling, V.J., Lester, D., Mortensen, P.B., Langenberg, P.W., and Postolache, T.T. (2011). *Toxoplasma gondii* seropositivity and suicide rates in women. *J. Nerv. Ment. Dis*. 199: 440–444.
- Lodish H., Berk A., and Zipursky S.L. (2000). "Section 21.4 Neurotransmitters, Synapses, and Impulse Transmission". Molecular Cell Biology (4th ed.). New York: W. H. Freeman.
- Lopez, A., Dietz, V.J., Wilson, M., Navin, T.R., and Jones, J.L. (2000) Preventing congenital toxoplasmosis. *MMWR Recomm Rep*. 49(RR-2):59–68.

- Lopez, W.D., Santos, T.R., Silva, R. S., Rossanese, W. M., Souza, F.A., Rodrigues, J.D., Mendonca, R. P., Vando-Edesio, V. E., and Costa, A. J (2010). Seroprevalence of and risk factors for *Toxoplasma gondii* in sheep raised in the Jaboticabal micro region, Sao Paulo State, Brazil. *Veterinary. Science*. 88:104-106.
- Lopez, W.D.Z. (2007). Aspects of toxoplasmatic infection in reproductive organs of artificially infection male ovines (Ovis aries). In: Abstract Book Waavp Congress, Gent. 49:69-68.
- Lori, C., Kenneth, H. E., Martha, E. W., and Imtiaz, A. (2002).CD8 + T-cell immunity against *Toxoplasma gondii* can be induced but not maintained in mice lacking conventional CD4+ T cells. *Infect. Immunol*. 70(2): 434-443.
- Luft, B.J. and J.S. Remington. (1992). Toxoplasmic encephalitis in AIDS. *Clin. Infect. Dis*., 15(2): 211 – 222.
- Lunden, A.; U. Carlsson and K. Naslund (1992). Toxoplasmosis and border disease in 54 Swedish sheep flocks: seroprevalence and incidence during one gestation period. *Acta Vet. Scand*. 33(2): 175-184.
- Lyons, R.E., McLeod, R. and C. W. Roberts (2002). *Toxoplasma gondii* tachyzoite-bradyzoite interconversion. *Trends Parasitol*. 18(5):198-201.
- Macit, H. B., Macit, G. and Gungor, O. (2018) A Research on Social Media Addiction and Dopamine Driven Feedback. *Mehmet Akif Ersoy Üniversitesi İktisadi ve İdari Bilimler Fakültesi Dergisi*. 5(3):882-897.
- Maclean, J. D. (2005). Clinical Parasitology. 20 th Ed. WB Sannders Co. London.

- Madlaina, B., Pierre-Mehdi, H., Sunil Kumar, D., Stéphane, P., Maged, G., Ivan, R., Dominique, S. (2020). Neuroinflammation-Associated A Specific Manipulation of Mouse Predator Fear by *Toxoplasma gondii*. *Cell Rep*., 30(2): 320–334.
- Maenz, M., Schluter, D., Liesenfeld, O., Schares, G., Gross, U., and Pleyer, U. (2014) Ocular toxoplasmosis past, present and new aspects of an old disease. *Prog. Retin. Eye Res*. 39:77–106.
- Mahalakshmi, B., Therese, K. L., Madhavan, H.N., and Biswas, J. (2006). Diagnostic value of specific local antibody production and nucleic acid amplification technique; nested polymerase chain reaction (nPCR) in clinically suspected ocular Toxoplasmosis. *O. Immunol. Inflamm*.14(2):105-112.
- Mahdi D. S., Awad A.H. and Ali A. T. (2020). The effect of Toxoplasmosis on Hematological and Biochemical Parameters in Pregnant Women in Thi-Qar Province. *Indian Journal of Forensic Medicine & Toxicology*, 14(1):989-992.
- Mahmood O. I. (2016). Effect of Toxoplasmosis on hematological, biochemical and immunological parameters in pregnant women in Tikrit city, Iraq. *Tikrit Journal of Pure Science*; 21 (3):24-27.
- Mahmoudv and H., Saedi Dezaki E., Soleimani S., Baneshi M.R., Kheirandish F., Ezatpour B., and Zia-Ali N. (2015). Seroprevalence and risk factors of *Toxoplasma gondii* infection among healthy blood donors in south-east of Iran. *Parasite Immunol*.;37(7):362-7.

- Mahmud R., Lim Y.A. Lian and Amir A. (2017) *Medical Parasitology*. Springer International Publishing Switzerland p (53-58).
- Malenka, R.C., Nestler, E.J., and Hyman, S.E. (2009). " Widely Projecting Systems: Monoamines, Acetylcholine, and Orexin". In Sydor A, Brown RY (eds.). Molecular Neuropharmacology: A Foundation for Clinical Neuroscience (2nd ed.). New York: McGraw-Hill Medical. pp. 147–48, 154–57.
- Maner, B.S., and Moosavi, L. (2022). Mean Corpuscular Volume. Stat Pearls. Treasure Island (FL): Stat Pearls Publishing.
- Mareze, M., Nascimento Benitez, A. D., Drulla Brandão, A. P., Pinto-Ferreira, F., Miura, A. C., Cardoso Martins, F. D., Caldart, E. T., Biondo, A. W., Freire, R. L., Mitsuka-Breganó, R., and Navarro, I. T. (2019). Socioeconomic vulnerability associated to *Toxoplasma gondii* exposure in southern Brazil. *Plos One*, 14(2): e0212375.
- Marrelli, Mauro Toledo *et al*. (2020) Detrimental effects of malaria, toxoplasmosis, leishmaniosis and Chagas disease on cardiac and skeletal muscles. *Medical Research Archives*, [S.l.], v. 8(9): 2-17.
- McConkey, G. A., Martin, H. L., Bristow, G. C. and Webster, J. P. (2013). *Toxoplasma gondii* infection and behaviour-location, location, location? *J. Exp. Biol*. 216, 113-119.
- McCullough J. (2014). RBCs as targets of infection. *Hematology Am Soc Hematol. Educ. Program* 2014(1): 404–409.
- McLeod, R., Boyer, K.M., Karrison, T., Kasza, K., Lee, D., Mui, E., Wroblewski, K., Noble, A.G., Withers, S., Swisher, C.N.,

Roizen, N. *et al*. (2006). Outcome of treatment for congenital Toxoplasmosis,1981-2004: The National Collaborative Chicago–Based, Congenital Toxoplasmosis Study. *Clin. Infect. Dis.* 42(10): 1383-1394.

- Migaki, G.; Sawa, T. R. and Duby, J. P. (1990) Fatal disseminated toxoplasmosis in a spinner dolphin (Stenella longirostris). *Vet Pathol*; 27(6):463-464.
- Mirzaeipour M., Mikaeili F., Asgari Q., Nohtani M., Rashidi S., and Bahreini, M.S., (2021) "Evaluation of the Tyrosine and Dopamine Serum Levels in Experimental Infected BALB/c Mice with Chronic Toxoplasmosis", *Journal of Parasitology Research*. 2021(5):1-9.
- Mizuri, S. S. M., and Mero, W. M. (2020). Seroprevalence of anti-*Toxoplasma gondii* antibodies among women of childbearing age in Zakho City, Kurdistan Region/Iraq. *Zanco J. Pure AND Applied Sciences*, 31(3):75-84.
- Mohamed. K. (2020). Hematological Changes during Chronic *Toxoplasma gondii* Infection in Pregnant Women in Makkah, Saudi Arabia. *American Journal of Infectious Diseases*. 16 (2): 36.39.
- Montoya, J. G. and Liesenfeld, O. (2004) Toxoplasmosis. *The Lancet*. 363: 1965-1976.
- Montoya, J. and Rosso, F. (2005). Diagnosis and management of Toxoplasmosis. *Clin. Perinatol*. 32: 705-726.
- Montoya, J. G. (2002). Laboratory diagnosis of *Toxoplasma gondii* infection and toxoplasmosis. *J. Infect. Dis*. 185(1):73-82.

- Montoya, J. G. and Remington, L. S. (1996). Toxoplasmic chorior- etinitis in the setting of acute, acquired toxoplasmosis. *Cym. Infec. Dist*. 23(2): 277-282.
- Montoya, J.G. and Remington, J.S. (2000). *Toxoplasma gondii*. In: Principles and Practice of Infectious Diseases. G.L. Mandell, J. E. Bennett and R. Dolin, (eds). Curchill Livingston. Philadelphia.
- Mordue, D. G. and Sibley, L. D. (1997). Intracellular fate of vacuoles containing *Toxoplasma gondii* is determined at the time of formation and depends on the mechanism of entry. *J. Immunol*. 159(9): 4452–4459.
- Musacchio J.M. (2013). " Enzymes involved in the biosynthesis and degradation of catecholamines". In Iverson L (ed.). Biochemistry of Biogenic Amines. Springer. pp. 1–35.
- Naot, Y. and Remington, J. S. (1980). An enzyme-linked immunosorbent assay for detection of IgM antibodies to *Toxoplasma gondii*: use for diagnosis of acute acquired toxoplasmosis. *J. Infect. Dis*. 142(5):757–766.
- Nash, J., Chissl, S., Jones, J., Warburton, F., and Verlander, N. (2005). Risk factors for toxoplasmosis in pregnant women in Kint, United Kingdom. *Epidemiol. Infect*. 133(3):475-483.
- Nayeri, T., Sarvi, S., Moosazadeh, M., Hosseininejad, Z., Amouei, A., and Daryani, A. (2021) Association between *Toxoplasma gondii* Infection and Headache: A Systematic Review and Meta-Analysis. *Infect Disord Drug Targets*, 21(4):643-650.

- Nazan, M.D. (2008). Congenital *Toxoplasma gondii* infection. *J.Mar. Med*., 21(1):89-101.
- Ndiaye, D., Sene, P. D., Ndiaye, M., Faye, B., Ndiaye, J. L., and Ndir, O. (2011). Update on Toxoplasmosis Prevalence based on serological tests in pregnant women in Dakar, Senegal from 2002to 2006. *Med. Trop (Mars).* 71(1), 101-102.
- Nicolle, C. and Manceaux, L. (1909). Sur un protozoaire nouveau du gondi. *C. R. Seances Acad. Sci.* 148:369–372.
- Nissapatorn, V., Kamarulzaman. A., Init, I., Tan, L.H., Rohela, M., Norliza, A., Chan, L.L, Latt, H.M., Anuar, A.K., and Quek, K.F. (2002) Seroepidemiology of toxoplasmosis among HIV-infected patients and healthy blood donors. *Med J Malaysia* 57(3):304–310.
- Nissapatorn, V., Suwanrath, C., Sawangjaroen, N., Ling, L. Y., and Chandeying, V. (2011). Toxoplasmosis-serological evidence and associated risk factors among pregnant women in southern Thailand. *Am. J. Trop. Med. Hyg*. 85(2): 243-247.
- Notarangelo, F., Wilson, E., Horning, K., Thomas, M., Harris, T., Fang, Q., Hunter, C. and Schwarcz, R. (2014) Evaluation of kynurenine pathway metabolism in *Toxoplasma gondii*-infected mice: implications for schizophrenia. *Schizophrenia Research* 152(1): 261–267.
- Omidian M., Asgari Q., Bahreini M.S., Moshki S., Sedaghat B., and Adnani Sadati S.J. (2022) Acute toxoplasmosis can increase serum dopamine level. *J Parasit Dis*. 46(2):337-342.

- Ortega, Y.R. (2007). Protozoan parasites. Ch 31 In: Doyle MP, Beuchat LR (eds) Food microbiology: Fundamentals and frontiers. 3rd ed, ASM Press, Washington D.C., p. 663–681.
- Paniker, C. K. J. (2002). Text book of Medical Parasitology. 5 th ed. Chap. (6). Jaypee brothers medical Publishers, New Delhi. pp:89-96.
- Pappas, G., Roussos, N., and Falagas, M. E. (2009) Toxoplasmosis snapshots: Global status of *Toxoplasma gondii* seroprevalence and implications for pregnancy and congenital toxoplasmosis. *Int. J. Parasitol*. 39(12): 1385–1394.
- Paquet, C., Yudin, M. H., and Society of Obstetricians and Gynaecologists of Canada (2013). Toxoplasmosis in pregnancy: prevention, screening, and treatment. *Journal of obstetrics and gynaecology Canada.*35(1), 78–81.
- Parameswaran, N., Thompson, R.C., Sundar, N., Pan, S., Johnson, M., Smith, N.C., and Grigg, M.E. (2010) Non-archetypal Type II-like and atypical strains of *Toxoplasma gondii* infecting marsupials of Australia. *Inter. J. Parasitol*. 40(6):635–640.
- Parlog A., Schluter D., and Dunay I.R. (2015) *Toxoplasma gondii*-induced neuronal alterations. *Parasite Immunology* 37(3): 159–170.
- Paul, M. (1998). Potencjalne zrodla zarażenia *Toxoplasma gondii* przypadkach badanych krotkim czasie po zarażeniu. *Przegl. Epidemiol*. 52: 447-454.

- Pelloux, H., Bessieres, M.H. and Chemla, C. (2006). Detection of anti-Toxoplasma IgM in pregnant women. *Ann. Biol. Clin. (Paris)*.64(1): 95-98.
- Peng X., Brenner L.A., Mathai A.J., Cook T.B., Fuchs D., Postolache N., Groer M.W., Pandey J.P., Mohyuddin F., Giegling I., Wadhawan A., Hartmann A.M., Konte B., Brundin L., Friedl M., Stiller J.W., Lowry C.A., Rujescu D. and Postolache T.T. (2018) Moderation of the relationship between *Toxoplasma gondii* seropositivity and trait impulsivity in younger men by the phenylalanine-tyrosine ratio. *Psychiatry Research* 270: 992–1000.
- Perkins H.A., and Busch M.P. (2010) Transfusion-associated infections: 50 years of relentless challenges and remarkable progress. *Transfusion* 50: 2080-2099.
- Pfefferkorn, L. C. and Pfefferkorn, E. R. (1980). *Toxoplasma* gondii: genetic recombination between drug resistant mutants. *Exp. Parasitol*. 50(3):305– 316.
- Pinlaor S., Ieamviteevanich K., Pinlaor P., Maleewong W., and Pipitgool V. (2000) Seroprevalence of specific total immunoglobulin (Ig), IgG and IgM antibodies to *Toxoplasma gondii* in blood donors from Loei Province, Northeast Thailand. *Southeast Asian J Trop Med Public Health* 31(1): 123- 127.
- Prandovszky E., Gaskell E., Martin H., Dubey J.P., Webster J.P. and McConkey G.A. (2011) The neurotropic parasite *Toxoplasma gondii* increases dopamine metabolism. *Plos One* 6(9): e23866.

- Puglisi-Allegra S., and Ventura R. (2012). Prefrontal/accumbal catecholamine system processes high motivational salience. *Front. Behav. Neurosci*. 6: 31.
- Purves, W. K., Sadava, D., Orians, G.H., and Heller, H. C. (2004) Life: The Science of Biology. ‘Sunderland, Mass: Sinauer Associates, USA.
- Raz I., Katz A., and Spencer M.K. (1991). Epinephrine inhibits insulin-mediated glycogenesis but enhances glycolysis in human skeletal muscle. *The American Journal of Physiology*. 260 (3 Pt 1): E430–E435.
- Remington, J. S., P. Thulliez and J. G. Montoya (2004). Recent developments for Diagnosis of toxoplasmosis. *J. Clin. Microbiol.* 42(3):941-945.
- Remington, J. S. (1969). The present status of the IgM fluorescent antibody technique in the diagnosis of congenital toxoplasmosis. *J. Pediatr*., 75(6):1116–1124.
- Remington, J. S., Mcleod, R. and Desmonts, G. (1995). Toxoplasmosis in: J.S. Remington and J.O. klein. (eds): Infectious disease of the Fetus and Newborn Infant. 4PthP ed. W.B. Saunders Company Philadelphia.
- Remington, J. S., McLeod, R., Thulliez, P. and Desmonts, G. (2005). Infectious Diseases of the fetus and newborn infant, W.B. Saunders Comp. Philadelphia. pp.947-1091.
- Remington, J. S., McLeod, R., Thulliez, P. and Desmonts, G. (2006). Toxoplasmosis. In: Remington, J. S. and Klein, J. O. (ed.), Infectious Diseases of the Fetus and Newborn Infant. 5th ed. W. B. Saunders, Philadelphia, PA. p. 947–091.

- Remington, J. S., Miller, M. J. and Brownlee, I. E. (1968). IgM antibodies in acute toxoplasmosis. II. Prevalence and significance in acquired cases. *J. Lab. Clin. Med*., 71(5):855–866.
- Remington, J.S., Eimstad, W.M., and Araujo, F.G. (1983). Detection of immunoglobulin M antibodies with antigen-tagged latex particles in an immunosorbent assay. *J. Clin Microbiol.*,17(5):939–941.
- Robert-Gangneux, F. and Dardé, M. L. (2012) Epidemiology of and diagnostic strategies for toxoplasmosis. *Clin. Microbiol. Rev*. 25(2): 264–296.
- Robinson, C.M., O'Dee, D., Hamilton T., and Nau, G.J. (2009). Cytokines involved in interferon - gamma production by human macrophages. *J. Inn. Imm*. 2(1): 56-65.
- Roe K. (2021). The Symptoms and Clinical Manifestations Observed in COVID-19 Patients/Long COVID-19 Symptoms that Parallel *Toxoplasma gondii* Infections. *J Neuroimmune Pharmacol.* 16(3), 513–516.
- Roghmann, M. C., Faulkner, C. T., Lefkowitz, A., Patton, S., Zimmerman, J., and Morris, J. G. (1999). Decreased seroprevalence for *Toxoplasma gondii* in Seventh Day Adventists in Maryland. *Am. J. Trop. Med. Hyg*. 60: 790-792.
- Roitt, I.; Brostoff, G. and Male, D. (2001). Parasitology and Vector biology 2PndP ed. Academic Press.pp165-178.
- Ruelhmann, D. S. (2010). Myopathic disorders. Consultations in feline internal medicine. Elsevier Saunders, Philadelphia, 603.
- Sabin, A. B. and Olitsky, P. K. (1937). *Toxoplasma* and obligate intracellular parasitism. *Science*, 85(2205):336–338.

- Sabin, A.B. and, Feldman, H.A. (1948). Dyes as microchemical indicators of a new immunity phenomenon affecting a protozoon parasite (*Toxoplasma*). *Science.*108(2815):660–663.
- Sacks, J., Roberto, R. and Brooks, N. (1982). Toxoplasmosis infection associated with raw goat's milk. *JAMA* 248:1728-1732.
- Saheb E. J. (2017). Detection of toxoplasmosis infection in diabetic patients. *Diyala J. of Medicine* 12(1):70-74.
- Salih J. M., Mero W. S. and Eassa S. H. (2020) Seroprevalence and some demographic factors associated with *Toxoplasma gondii* infection among female population in Duhok province, Iraq. *International Journal of Research in Medical Sciences* 8(3):921-926.
- Salvagno G.L., Sanchis-Gomar F., Picanza A., *et al*. (2015) Red blood cell distribution width: A simple parameter with multiple clinical applications. *Crit Rev Clin Lab Sci* 52:86-105.
- Sarkari B., Shafiei R., Zare M., Sohrabpour S., and Kasraian L. (2014) Seroprevalence and molecular diagnosis of *Toxoplasma gondii* infection among blood donors in southern Iran. *J Infect Dev Ctries* 8(4):543-547.
- Schacter D., Gilbert D., Nock M. and Wegner D. (2009). Psychology. 5th Edition, Worth Publisher, Macmillan, New York, USA.
- Schmidt, G. D.; Roberts, L.S. and Janovy, J. (2005). Foundations of parasitology. 7Pth P ed., McGraw – Hill Companies, Inc. U.S.A., p 134 – 138.

- Schmidt, M. *et al*. (2013) Clinical features and outcomes in patients with disseminated toxoplasmosis admitted to intensive care: a multicenter study. Clin. Infect. Dis. 57, 1535–1541.
- Seeman P. (2009). " Historical overview: Introduction to the dopamine receptors". In Neve K (ed.). The Dopamine Receptors. Springer. pp. 1–22.
- Shapiro K., Bahia-Oliveira L., Dixon B., Dumètre A., Wit L., VanWormer E. and Villena I. (2019) Environmental transmission of *Toxoplasma gondii*: Oocysts in water, soil and food. *Food and Waterborne Parasitology* 15: e00049.
- Shehzad A., Masud A., Fatima T., Khan F.M., Rehman S., Effendi M.H., Suwanti L.T., Khan I., Tyasningsih W., Faisal S., Abadeen Z.U., and Bibi S. (2022) Seroprevalence of *Toxoplasma gondii* and associated alterations in hematology and serum biochemistry of one-humped camels (*Camelus dromedarius*) in Pakistan. *Veterinary World*, 15(1): 110-118.
- Shen H. (2008). Illustrated Pharmacology Memory Cards: PharMnemonics. Minireview. p. 4.
- Shirbazou, S., Delpisheh, A., Mokhetari, R., and Tavakoli, G. (2013). Serologic Detection of Anti *Toxoplasma gondii* Infection in Diabetic Patients. Iranian Red Crescent Medical Journal, 15(8), 701-703.
- Sibley, L.D. and Ajioka, J.W. (2008). Population structure of *Toxoplasma gondii*: Clonal expansion driven by infrequent recombination and selective sweeps. *Ann. Rev. Microbiol*. 62:329–351.

- Sibley, L.D. and Boothroyd, J. C. (1992). Virulent strains of *Toxoplasma gondii* Comprise Single Clonal lineage. *Nature* 359(6390):82–85.
- Siddiqui, M.R., Rafiquee, M. Z. A., Wabaidur, S. M., Alothman, Z.A., ALI, M.S. and Allohedan, H.A. (2015). Synthesis of Silver Nanoparticle: A New Analytical Approach for the Quantitative Assessment of Adrenaline. *Analytical Sciences*.31(5):437-443.
- Silveira, C.; Belfort, R. J. and Muccioli, C. (2002). The effect of long term intermittent trimethoprim / sulfamethoxazole treatment on recurrences of Toxoplasmic retinochoroiditis. *Am. J. Ophthalmol*. 134(1):41-6.
- Singh, B., Debrah L. B., Acheampong G. and Debrah A. Y. (2021). Seroprevalence and risk Factors of *Toxoplasma gondii* infection among pregnant women in Kumasi: A cross-sectional; Study at a District-Level Hospital, Ghana. *Infectious Diseases in Obstetrics and Gynecology*, 2021(2):1-9.
- Sircar S. (2007). Medical Physiology. Thieme Publishing Group. p. 536.
- Skallova, A., Kodym, P., Frynta, D. and Flegr, J. (2006) The role of dopamine in *Toxoplasma*-induced behavioural alterations in mice: an ethological and ethopharmacological study. *Parasitology*. 133(5): 525-535.
- Song, K.J., Shin, J.C., Shin, H.J. and Nam, H.W. 2005. Seroprevalence of toxoplasmosis in Korean pregnant women. *The Korean Journal of Parasitology* 43(2): 69.

- Stibbs H.H. (1985). Changes in brain concentrations of catecholamines and indole amines in *Toxoplasma gondii* infected mice. *Ann Trop Med Parasitol*. 79:153-157.
- Stopić M, Štajner T, Marković-Denić L, Nikolić V, Djilas I, Srzentić SJ, Djurković-Djaković O, and Bobić B. (2022) Epidemiology of Toxoplasmosis in SERBIA: A Cross-Sectional Study on Blood Donors. *Microorganisms*;10(3):492.
- Strabelli T. M., Siciliano R.F., Vidal Campos S., Bianchi Castelli J., Bacal F., Bocchi E. A. and Uip D. E. (2012). *Toxoplasma gondii* myocarditis after adult heart transplantation: successful prophylaxis with pyrimethamine. *J Trop Med*. 12:853562.
- Su, C., Khan, A., Zhou, P., Majumdar, D., Ajzenberg, D., Darde, M.L., Zhu, X.Q., Ajioka, J.W., Rosenthal, M., Dubey, J.P. and Sibley, L.D. (2012). Globally diverse *Toxoplasma gondii* isolates comprise six major clades originating from a small number of distinct ancestral lineages. *Proceedings of the National Academy of Science* 109(15):5844–5849.
- Sugden K., Moffitt T.E., Pinto L., Poulton R., Williams B.S. and Caspi A. (2016) Is *Toxoplasma gondii* infection related to brain and behavior impairments in humans? Evidence from a population-representative birth cohort. *Plos One* 11(2): e0148435.
- Sutterland A., Fond G., Kuin A., Koeter M., Lutter R., van Gool T., Yolken R., Szoke A., Leboyer M. and de Haan L. (2015) Beyond the association *Toxoplasma gondii* in schizophrenia,

bipolar disorder, and addiction: systematic review and meta-analysis. *Acta Psychiatrica Scandinavica* 132:161–179.

- Suzuki, Y. (2012). Host resistance in the brain against *Toxoplasma gondii*. *J. Infect. Dis*., 185(1): 58–65.
- Suzuki, Y., Orellana, M. A., Schreiber, R.D. and Remington, J.S. (1988). Interferon-gamma: the major mediator of resistance against *Toxoplasma gondii*. *Science* 240(4851):516-8.
- Tenter, A. M., Heckeroth, A. R. and Weiss, L.M. (2000). *Toxoplasma gondii*: From animals to humans. *Int. J. Parasitol*. 30(12-13):1217–1258.
- The National Collaborating Centre for Chronic Conditions, ed. (2006). "Symptomatic pharmacological therapy in Parkinson's disease". Parkinson's Disease. London: Royal College of Physicians. pp. 59–100.
- Tibayrene, M., Kjellberg, F., Arnaud, J., Oury, B., Brenie`re, S. F., Darde´, M. L. and Ayala, F. J. (1991). Are eukaryotic microorganisms clonal or sexual? A population genetics vantage. *Proc. Natl. Acad. Sci. USA*, 88:5129–5133.
- Tønnesen, H., Hejberg, L., Frobenius, S., and Andersen, J. (1986). Erythrocyte mean cell volume--correlation to drinking pattern in heavy alcoholics. *Acta Med Scand*. 219(5):515-8.
- Torrey E.F., and Yolken R.H. (2003) *Toxoplasma gondii* and Schizophrenia. *Emerg Infect Dis.* 9(11):1357-1380.
- Verberne, A., Korim, W., Sabetghadam, A., and Llewellyn-Smith, I. (2016) Adrenaline: insights into its metabolic roles in hypoglycaemia and diabetes. *British Journal of Pharmacology* 173: 1425–1437

- Wang T, Han Y, Pan Z, Wang H, Yuan M, and Lin H. (2018). Seroprevalence of *Toxoplasma gondii* infection in blood donors in mainland China: a systematic review and meta-analysis. *Parasite*.;25:36.
- Wang X., Li J., Dong G., and Yue J. (2014). The endogenous substrates of brain CYP2D. *European Journal of Pharmacology*. 724: 211–218.
- Wang Z., Zhang D.-X. and Zhao Q. (2015). Infection-stimulated Anemia Results Primarily from Interferon Gamma-dependent, Signal Transducer and Activator of Transcription 1-independent Red Cell Loss. *Chin Med J (Engl)*. 128(7): 948–955.
- Warren, M. D. (2006). Review of Medical Microbiology and Immunology. J. Clin. Microbiol. 35: pp360-362.
- Webster R. (2001) Neurotransmitters, Drugs and Brain Function. 1st ed. Wiley, Chi Chester, UK.
- Webster, J.P., Kaushik, M., Bristow, G.C., and McConkey, G.A. (2013). *Toxoplasma gondii* infection from predation to schizophrenia: can animal behaviour help us understand human behaviour? *J. Exp. Biol*., 216: 99–112.
- Wenzel J.M., Rauscher N.A., Cheer J.F., Oleson E.B. (2015). "A role for phasic dopamine release within the nucleus accumbens in encoding aversion: a review of the neurochemical literature". *ACS Chemical Neuroscience*. 6 (1): 16–26.
- Wiendl, H., Hohlfeld, R., and Kieseier, B. (2005). Immunobiology of muscle: advances in understanding an immunological microenvironment. *Trends in Immunol*., 26(7):373-80.

- Wilking, H., Thamm, M., Stark, K., Aebischer, T. and Seeber, F. (2016). Prevalence, incidence estimations and risk factors of *Toxoplasma gondii* infection in Germany: a representative, cross-sectional, serological study. *Scientific Reports* 6(1): 1-9.
- Willis F. (2001). Diseases and Disorders Toxoplasmosis. *Modren Drug Discovery*, 4(5):11-18.
- Willis, M. S., P. Southern and F. Latimer (2002). *Toxoplasma* infection: Making the Best Use of Laboratory Test. *Infect. Med*. 19:522-532.
- Woldemichael, T., Fontanet, A. L., Sahlu, T., Gilis, H., Messele, T., Rinke de Wit, T. F and Van Gool, T. (1998). Evaluation of the Eiken latex agglutination test for anti-*Toxoplasma* antibodies and seroprevalence of *Toxoplasma* infection among factory workers in Addis Ababa, Ethiopia. *Trans. R. Soc. Trop. Med. Hyg*. 92(4), 401-403.
- Wu, L. and Garcia R.A. (2005). Toxoplasmosis. Med.Com. Inc, Section 1-11.
- Yacoub, A. H., Bakr, S., Hameed, A.M., Al Thamery, A. and Fartoci, M.J. (2006). Seroepidemiology of selected zoonotic infections in Basra region of Iraq. *EMHJ-Eastern Mediterranean Health Journal* 12(1-2): 112-118.
- Yereli K., Balcioglu I.C., and Ozbilgin A. (2006). Is *Toxoplasma gondii* a potential risk for traffic accidents in Turkey. *Forensic. Sci. Int*. 163(1-2):34-37.
- Zemene, E., Yewhalaw, D., Abera, S., Belay, T., Samuel, A. and Zeynudin, A. (2012). Seroprevalence of *Toxoplasma gondii* and

associated risk factors among pregnant women in Jimma town, Southwestern Ethiopia. *BMC Infectious Diseases* 12(1): 1-6.

- Zumla, A., Savva, D., Wheeler, R., Hira, S. L., Kaleebu, P., Sempala, S., and Holliman, R. (1991). *Toxoplasma* Serology in Zambian and Ugandan Patients Infected with The Human Immunodeficiency Virus. *T. Royal. Soc. Trop. Med. H.* 85(2):227 -229.

APPENDIX

Questionnaire sheet used for each individual participant in this study

- The number ……
- The age …….
- Academic level……...
- Income level ……...
- Marital status…….
- Do you have diabetes? Yes……, No…….
- Do you have a fever? Yes……, No…….
- Do you have constant headaches? Yes……, No…….
- Do you suffer from intermittent headaches? Yes……, No…….
- Do you suffer from visual impairment? Yes……, No…….
- Do you have gland dysfunction? Yes……, No…….
- Do you suffer from muscle pain? Yes……, No…….
- Do you suffer from fatigue tiredness? Yes……, No…….
- Have you had surgery? Yes……, No…….
- Have you had a blood transfusion? Yes……, No…….
- Do you clean the garden of the house? Yes……, No…….
- Do you come into contact with cats? Yes……, No…….
- Do you drink unpasteurized milk? Yes……, No…….
- Do you eat dairy products from street vendors? Yes…, No….

For female

- Are you pregnant? Yes……, No…….
- Do you have a late pregnancy? Yes……, No…….
- Have you had abortion? Yes……, No…….
- Did you give birth to an abnormal child? Yes……, No……

الخلاصة

التوكسوبلازما جوندي هو طفيلي واسع الانتشار على مستوى العالم أصاب حوالي 30- 50٪ من سكان العالم وتسبب في الإصابة بداء المقوسات، يصيب أكثر من 300 نوع من الحيوانات ذوات الدم الحار، بما في ذلك البشر (وفقا لبيانات منظمة الصحة العالمية)، والتي تعتبر مضيفًا وسيطًا، والقطط، وهي العائل النهائي.

أجريت الدراسة الحالية في محافظة ميسان جنوب العراق للتحقيق في انتشار داء المقوسات وتحديد علاقته بمستويات الناقلات العصبية الدوبامين والأدرينالين. تم جمع 174 عينة دم وريدي (153 ذكر و21 أنثى) من الأفراد الذين زاروا مستشفى الشهيد الصدر التعليمي ومصرف الدم الرئيسي في ميسان من كانون الاول (2020) إلى تشرين الأول (2021).

تم استخدام مقايسة الممتز المناعي المرتبط بالإنزيم (ELISA)لتحديد مستويات الغلوبيولين المناعي مثل الغلوبيولين المناعي M (IgM) والغلوبيولين المناعي G (IgG)، وكذلك لتحديد مستويات الدوبامين والأدرينالين في دم الأفراد المشاركين. تم استخدام اختبار تعداد الدم الكامل (CBC) لتحديد تأثير التوكسوبلازما جوندي على محتويات هذا الاختبار.

تم تسجيل بعض العوامل الاجتماعية والديموغرافية للأفراد المشاركين في استبيان مصمم لتقييم ارتباطها بداء المقوسات.

تتلخص النتائج في النقاط التالية:

1-كانت نسبة الإصابة بداء المقوسات 52.3٪ منها 32.97٪ موجبة للـ IgM وحده، و38.46٪ لـ IgG، و28.57٪ لـ IgMو IgGمعًا.

2-نسبة الإصابة عند الإناث 66.67٪ وللذكور 50.33٪.

3-توجد فروق ذات دلالة إحصائية بين مستوى الدوبامين للمصل الموجب والسالب. وتبين أن مستوى الدوبامين لدى الأفراد المصابين بداء المقوسات أعلى بنحو 163.347٪ من الأفراد غير المصابين.

4-مستوى الأدرينالين في التوكسوبلازما الإيجابية المصلية أعلى إحصائيا من تلك الموجودة في التوكسوبلازما - السالبة المصلية مع وجود فرق معنوي بينهما.

5-يوجد انخفاض ذو دلالة إحصائية في مستوى الهيموجلوبين وعدد كرات الدم الحمراء في دم الأفراد المصابين بداء المقوسات مقارنة بالأفراد غير المصابين.

6-انخفض تعداد كرات الدم البيضاء والصفائح الدموية في الأفراد المصابين بداء المقوسات مقارنة بالأفراد غير المصابين ولكن لم يلاحظ وجود فروق ذات دلالة إحصائية بينهم.

7- توجد علاقة ذات دلالة إحصائية بين مستوى التعليم ومعدل الدخل وداء المقوسات.

8- توجد علاقة ذات دلالة إحصائية بين تاريخ الجراحة ونقل الدم والحمى والصداع وآلام العضلات والتعب وداء المقوسات.

9- توجد علاقة ذات دلالة إحصائية بين الاتصال بالقطط والإصابة بداء المقوسات.

10-لا توجد علاقة ذات دلالة إحصائية بين الإيجابية المصلية لداء المقوسات ومشاكل الرؤية واختلال وظائف الغدد.

11- لا توجد علاقة ذات دلالة إحصائية بين إيجابية التوكسوبلازما وبعض السلوكيات مثل شرب الحليب غير المبستر أو تناول مشتقات الألبان من الباعة الجائلين وتنظيف حديقة المنزل.

12- توجد علاقة ذات دلالة إحصائية بين إيجابية التوكسوبلازما المصلية وتأخر الحمل والإجهاض والنساء اللواتي ولد أطفالهن بعيوب خلقية.

وزارة التعليم العالي والبحث العلمي

جامعة ميسان

كلية العلوم

قسم علوم الحياة

دراسة مصلية للعلاقة بين مرضى داء المقوسات ومستويات الدوبامين والأدرينالين

رسالة مقدمة الى

مجلس كلية العلوم / جامعة ميسان

كجزء من متطلبات نيل شهادة الماجستير

في علوم الحياة

من قبل

زهراء خالد مجبل

بكالوريوس تربية علوم الحياة / جامعة البصرة (2004)

بأشراف

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