Risk factors of toxoplasmosis in Libya: a brief review
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Abstract Toxoplasmosis is caused by the protozoan parasite Toxoplasma gondii which infects both humans and animals as a zoonotic pathogen widespread in nature. Infection in humans occurs worldwide, but prevalence varies significantly between populations. In Libya, T. gondii is prevalent among humans as well as animals, however, the main reason yet unclear as no adequate studies regarding transmission modes were available. Thus, the present review aimed to collect and summarize data on the risk factors associated with T. gondii infection in Libya. Published data from national and international databases were reviewed. Prevalence of T. gondii infection among humans and animals were also discussed. This review would be a useful tool for proposing appropriate national toxoplasmosis control programs.

Keywords: toxoplasmosis, Toxoplasma gondii, risk factors, Libya.

Introduction
Toxoplasmosis is an important zoonotic disease caused by an obligate intracellular protozoan parasite called Toxoplasma gondii [1,2]. The parasite is able to infect humans as well as other warm blooded domestic and wild animals [3,4]. All mammals, including humans, and birds are intermediate hosts, whereas Felidae (cats) are definitive as well as intermediate hosts [1]. T. gondii was first described in Tunisia by Nicolle and Manceaux at the Pasteur Institute in 1908. Whilst conducting leishmaniasis research on the North African rodent, Ctenodactylus gondi, the investigators isolated T. gondii merozoites from its blood, liver and spleen. In 1923, Janku described the parasitic cysts in the retina of a child with congenital hydrocephalus which considers first infection in human [2]. In 1939, Wolf and colleagues successfully isolated the parasite from tissue from a neonate with encephalitis [5,6]. Toxoplasma has a number of genetic types but about 95% of them are categorized into three classes of strains simply named as Type I, II and III. These genotypes are considered to be clonal in their structural features [7]. Type I causes infection in rodents. Type II has been established as the infectious agent of toxoplasmosis in small ruminants while Type III has yet not been proved as infectious strain. All the three genetic types I, II and III can cause toxoplasmosis in human. Recently, a newly discovered genotype (Type IV) has also been reported mostly in wildlife [8]. Human infections are acquired through direct or indirect contact with cat feces. Transmission of the parasite can occur in several ways, by eating raw or undercooked meat containing T. gondii tissue cysts, ingesting oocysts via contact with infected cat feces, or by touching contaminated soil or consuming food or water contaminated with oocysts [9], or by acquiring congenital infection through the placenta [10]. In addition, T. gondii infection can also be acquired by blood transfusion and organ transplantation [4,11]. The overall life cycle of Toxoplasma contains two distinct cycles: a sexual that occurs in the small intestine of the feline family (definitive hosts), and an asexual phase in infected animals, including humans (intermediate hosts). [6,12]. The parasite has three infective forms during its life cycle: tachyzoite, bradyzoite, and oocysts. However, all hosts, including humans, can be infected by any one of the three forms of the parasite. The rapidly dividing Tachyzoite forms...
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found during the acute stage of infection which are capable to invade and replicate within cells and are responsible for congenital infection. The slowly dividing bradyzoite found during latent infections; this form of the parasite is present in tissue cysts. Oocysts are the zygotic stage of the life cycle, and are excreted unsporulated in cat faeces. Sporulation occurs outside of the body to form sporulated oocyst containing four sporozoites. The oocysts are environmentally robust, and can retain infectivity in a cool damp environment for months [3,13]. Primary infection of toxoplasmosis in healthy individuals is normally asymptomatic or associated with influenza-like illness, which includes limited symptoms such as fever, myalgia, malaise, and headache [14,15,16]. However, toxoplasmosis can cause serious implications in immuno-suppressed individuals including HIV patient, pregnant women as well as its severe consequences on fetuses in congenital transmission [17]. The disease in pregnancy has been associated with spontaneous abortion, miscarriage, stillbirth, hydrocephalus, intrauterine malformations in the fetus, cerebral calcification and chorioretinitis in the newborn [17,18].

Diagnosis of toxoplasmosis in humans is performed using different techniques including serological testing, histological identification, isolation in tissue culture as well as, molecular methods using the Polymerase Chain Reaction (PCR) or by a combination of these techniques. Acute and latent T. gondii infections are mostly diagnosed by serological tests including increased antibody levels such as IgG, IgM, IgA and IgE. In a primary T. gondii infection, IgM appears a few weeks after infection, followed by IgA and IgE. These acute phase immunoglobulins peak after about two months and are usually undetectable by serological tests by six to nine months but can persist for longer periods of time. IgG appears after IgM peaks after four months and persists at low levels throughout the duration of the host’s life [6,16], the most commonly used serological tests include the indirect haemagglutination assay, indirect fluorescent antibody assay (IFA), direct agglutination test (DAT), Latex agglutination test (LAT), and Enzyme-Linked Immunosorbent assay (ELISA) [19].

Risk factors associated with transmission of Toxoplasma gondii

T. gondii is prevalent in Libya in both humans and animals [20,21]. Several studies have been conducted on women in childbearing age (pregnant women, non-pregnant), psychosocial patients, schoolchildren and blood donors to determine the prevalence of infection. Most of the studies were carried out in the east, west and north of Libya, whereas few studies conducted in the south. The results showed significant differences from region to region (Table 1). The main source of infection to herbivores is ingestion of oocysts contaminated in grasses, feed and water. Therefore, toxoplasmosis in animals is significantly associated with presence of cats in the farms [7]. Most of the acquired infections of T. gondii in herbivores (Sheep, goat, cattle, camel and equine) are subclinical. However, fever, ataxia and retinal degeneration and encephalomyelitis may develop. Severity of toxoplasmosis in ovine is associated with the stage of pregnancy[22]. The most devising outcome of toxoplasmosis is miscarriage or abortion which is particularly important in humans and domestic livestock. Additionally, it can cause a wide variety of neurological disease especially when transmitted congenitally. In Libya, limited studies are made on the prevalence of toxoplasmosis among animals. El-Gomati et al (2008 and 2010) [24,25] found 40.7% and 35% T. gondii infection among sheep and mice in Tripoli respectively.

(Table 1) Prevalence of T. gondii in Libya

<table>
<thead>
<tr>
<th>Region/ Year</th>
<th>Sample size</th>
<th>Population</th>
<th>Assay</th>
<th>Seroprevalence (%)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tripoli 1987</td>
<td>2000</td>
<td>adult males</td>
<td>Latex</td>
<td>51.6</td>
<td>[44]</td>
</tr>
<tr>
<td></td>
<td>300</td>
<td>adult females</td>
<td>Agglutination (LA)</td>
<td>43.4</td>
<td>[36]</td>
</tr>
<tr>
<td></td>
<td>1980</td>
<td>schoolchildren</td>
<td>indirect haemaggulitation test (IHA)</td>
<td>43.7</td>
<td>[33]</td>
</tr>
<tr>
<td>Benghazi 1991</td>
<td>369</td>
<td>pregnant women</td>
<td>ELISA</td>
<td>47.4</td>
<td>[33]</td>
</tr>
<tr>
<td>Tripoli 2008</td>
<td>692</td>
<td>women who suffered spontaneous abortion</td>
<td>IgM 17.6, IgG 45</td>
<td>[45]</td>
<td></td>
</tr>
<tr>
<td>Tripoli 2008</td>
<td>474</td>
<td>non-pregnant</td>
<td>IgG 18.14</td>
<td>[46]</td>
<td></td>
</tr>
<tr>
<td>Benghazi 2011</td>
<td>143</td>
<td>pregnant women with previous adverse pregnancy outcome.</td>
<td>IgM 8.4, IgG 44.8</td>
<td>[39]</td>
<td></td>
</tr>
<tr>
<td>Tripoli 2014</td>
<td>300</td>
<td>psychiatric patients</td>
<td>ELISA</td>
<td>IgG 50.3</td>
<td>[31]</td>
</tr>
<tr>
<td></td>
<td>300</td>
<td>control volunteers</td>
<td>Latex</td>
<td>46.7</td>
<td>[31]</td>
</tr>
<tr>
<td>Benjawad 2015</td>
<td>280</td>
<td>pregnant</td>
<td>ELISA</td>
<td>IgM 3.57, IgG 37.14</td>
<td>[32]</td>
</tr>
<tr>
<td></td>
<td>250</td>
<td>non-pregnant</td>
<td>IgM 3.6, IgG 37.20</td>
<td>[32]</td>
<td></td>
</tr>
<tr>
<td>Alkhoms 2015</td>
<td>361</td>
<td>pregnant women</td>
<td>ELISA</td>
<td>IgG 39.3</td>
<td>[36]</td>
</tr>
<tr>
<td>Misurata 2015</td>
<td>300</td>
<td>pregnant women</td>
<td>Latex Elecsys-Cobas e analyzer</td>
<td>26.7</td>
<td>[47]</td>
</tr>
<tr>
<td>Sebha 2016</td>
<td>267</td>
<td>pregnant women</td>
<td>ELISA</td>
<td>IgM15.37,IgG 25.89</td>
<td>[49]</td>
</tr>
</tbody>
</table>
Recently, Al-Mabruk et al (2013) [26] found significantly higher seroprevalence (71%) of antibody of Toxoplasma in sheep in Tripoli. They consider sheep are suitable host for T. gondii in Libya. Worldwide, prevalence of the toxoplasmosis is measured by detection of specific anti-Toxoplasma IgG antibodies varies between 1% and 100 depending on different factors such as geographical location, age, habit of eating raw meat or unwashed fruit and vegetables, sociocultural and nutritional habits, general level of hygiene and contact with domestic cats. [2,27, 28].

**Geographical location:** Prevalence of infection with T. gondii varies widely between countries and often from one country to another or even within the same country or between different communities in the same region. This wide variability may be attributed to differences in climatic conditions, cultural differences regarding hygienic and feeding habits. A higher sero-prevalence is associated with humid areas which are favourable for sporulation of oocysts that voided in cat feces compared to arid areas. Moreover, oocyst survival increases in humid conditions where it can remain viable in a moist environment for more than a year [29,30,31]. In Libya, results of several studies showed high prevalence of parasite in the eastern and western regions compared to the southern region of the country. This can be attributed to the difference in environmental conditions, such as the high rainfall, and high humidity, making it an environment suitable for the sporulation and surviving of oocysts. However, hot and dry environment as such in the southern regions of the country could be unfavouring the development and surviving of the parasite which may explain the moderate to low incidence of T. gondii in the southern regions [32].

**Age:** Previous studies conducted in Libya reported high rate of positivity of T. gondii among the older age group [20,30,31,32,33,34,35, 36,37]. This association does not mean that older age is a risk factor predisposing to infection but might be explained by the older the person the longer time being exposed to the causing agent and may retain a steady level of anti-toxoplasma IgG in serum for years [36]. On contrary, results of studies done in Benghazi and Sebha Cites showed a clear declination of seropositivity with age [22,38, 49]. This difference may be due to wide variations in age groups used in these studies. This could be explained by that older women are more likely to have been exposed to any one of the risk factors than younger women as a result of longer exposure time. In addition, decrease of immunity in old ages; increase the chance for more exposure to infection [30,32].

**Consumption of undercooked meat:** Meat of warm-blooded animals and birds has been considered a major source of Toxoplasma infection especially in countries that consumed raw or undercooked meat. Besides the consumption of T. gondii tissue cysts contained in meat, meat-derived products, or offal can be an important source of infection in humans [29,30]. Virtually, all edible portions of an animal can harbor viable T gondii organisms. It can also be transmitted by containers, knives or other utensils, cutting boards or other preparation surfaces contaminated with raw meat. Touching unwashed hands to the face after meat preparation is another source of infection. Type of meat consumed should also be considered since pork, mutton and farm chickens are commonly infected, while beef meat are rarely infected [30]. In their study, Gamal & Jaroud (2015) [36], reported significantly higher seropositivity rate (70.0%) among women consuming sheep meat compared to those (54.0%) consuming cow meat. The method and degree of meat cooking have an effect on seropositivity to T. gondii. Previous studies have shown that people who consuming undercooked meat have higher risk of infection than those eating well cooked meat [36]. In a study conducted by El-sayed et al. (2016), the sero-positivity of T.gondii was high among pregnant women eating roasted mutton or processed meat as hamburger, minced meat and Sharrrma which may be insufficiently cooked [30,36]. Raw or undercooking meat consumption was positively associated with T.gondii infection. Several studies identified an association between eating raw meat and T.gondii seropositivity [37,39].
On the other hand, transmission of *T. gondii* infection through uncooked meat in Libya is uncommon because most people preferred to eat well cooked meat [30,34,40]. Generally, thorough cooking is always preferred in Libya, and therefore the most possible way of transmission is probably through handling of raw contaminated meat, during food preparation. Local meat, sheep, goat or camel might be contaminated with oocysts due to poor hygiene during handling of meat from slaughterhouse to kitchen. In addition, in Libya, consumption of lamb is greater than that of beef. These trends may have increased exposure to toxoplasma because lamb has a higher risk of infection than beef or poultry [39].

**Eating raw or unwashed fruit and vegetables:** Contaminated fruit and vegetables by cat faeces have been considered source of *T. gondii* infection, especially in developing countries. This may attributed to their contamination with oocysts that carried by soil and water, in addition to, poor hygienic measures in these places such as consumption of unwashed vegetables. Several previous studies [31,37,49] and have found an association between the seropositivity among personals and intake of raw vegetables if proper hygiene is lacking and ingestion of oocysts occurs. On contrary, results of other studies found no such association [30].

**Drinking water:** Drinking of contaminated water is another source of *T. gondii* infection as oocysts of *T. gondii* can remain infective in water for a long time (i.e., under optimal conditions several months or even years) [41]. They are not killed by chemical and physical treatments currently applied in water treatment plants, including chlorination and ozone treatment [42]. So, high prevalence was observed among people who drunk untreated water that may have high risk of contamination by oocysts [30]. In a study conducted by Gamal and Jaroud (2015), the higher prevalence rates of *T. gondii* were reported in women using rain water (68.0%) and well water (60.0%). Meanwhile lower prevalence was in women using general network water (33.0%).

Corresponding finding was reported by Elsaid et al. (2014) in psychiatric patients in Tripoli. **Consumption of unpasteurized milk:** The infection via milk is not an important risk factor because tachyzoites which may pass in milk is very sensitive to environmental conditions and is usually killed rapidly outside the host. Also, tachyzoites were suggested to be rare cause of acquired toxoplasmosis in humans after the consumption of unpasteurized goat’s milk. However, a significant association between the consumption of raw milk and infection was recorded [30]. Generally, most of Libyan population consumes pasteurized milk. However, the consumption of raw milk in rural and some suburban areas is common [37] thus, consumption of non-processed milk from several animals is a potential source of *T. gondii* transmission [43].

**Contact with domestic cats:** Contact with cats and cats excrement have been considered as major risk factors for acquiring infection as oocysts that voided in feaces are main source of infection for human and animals [30]. The association between the infection and contact with cats was observed in several studies conducted in Libya [22,32,33,36,49]. On the contrary, other studies found no association between seroprevalence of toxoplasmosis and contact with cats. [20, 30,31,35,39, 37]. In addition, contact with dogs showed significant association with infection; therefore dogs might be an important route of *T. gondii* transmission [31]. However, the acquisition of cats as pets is not common practice in Libya and most of cats are straying. Stray cats were found in farms, gardens or may enter houses from time to another to obtain their food from remains of raw meat thrown in the garbage. This could increase the chance of infection especially for children living in houses with soil floor or playing in farms during picnics [30,39]. Hence, the cats play a central role in the epidemiology of *T. gondii* and constituting the only known source of environmental contamination with the infective oocyst stage. A high risk is thus imposed on human communities that come into contact with cats [15]. Cats may also play indirect role in transmission of toxoplasma through contaminated meat, vegetables and fruits.

**Contact with contaminated soil:** The soil consider as risk factor for transmitting *T. gondii* due to their contamination by oocysts dispersed in the excrement of stray cats. Soil contaminated with cat’s faeces may play role in transmitting infection through contamination of raw vegetables or water [30]. Therefore, contaminated fruit and vegetables by cat faeces and poor hand hygiene are important in parasite transmission [37].

**Blood transfusion:** Transmission of *T. gondii* through blood transfusion though uncommon, it is theoretically possible if the donor has recently acquired a *Toxoplasma* infection. However, several studies in Libya showed no significant association between *T. gondii* seropositive and blood transfusion [30,37].

**Conclusion:** *Toxoplasma gondii* is a widely prevalent parasite among both humans and animals that is potentially responsible for significant morbidity and mortality. While occurrence of the infection is well documented in Libya, the main sources of infection remain unclear. Based on available data, several risk factors of toxoplasmosis in Libya were identified including contact with cat and consumption of raw unwashed fruit and vegetables, consumption of undercooked or unhygienic prepared meat, drinking untreated water, older age, and geographic location. Therefore, knowledge of these risk factors could help to further reduce the burden of toxoplasmosis in Libya, raising health awareness, and designating of control strategies. Besides, attention should also be given to environmental sampling in order to develop adequate transmission models between animals, the environment and people. Furthermore, more comprehensive epidemiological studies are needed to guide decision makers to adopt and implement control programs involving both the medical and veterinary sectors.

References:
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In the city of Misrata, Libya, and its relationship with pregnancy. The second year.