

Parasite and host defense mechanisms: when *Toxoplasma* subjugates the brain

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Abstract

Brain is an immune-privileged organ for infection with the neurotropic protozoan parasite, *Toxoplasma gondii*. *T. gondii* escapes the immune system by establishing a dormant and lifelong bradyzoite cyst during chronic and latent phases within the brain and musculature of immunocompetent individuals. During acute stage *T. gondii* stimulates the host defense to display depressive behaviors over a background of sickness. Infection with *T. gondii* is initiated with its proliferative tachyzoite that acts on peripheral immune cells to produce the pro-inflammatory cytokines, most of them induce the behavioral signs of sickness. For instance, interleukin-(IL) 1 β , tumor necrosis factor- α and IL-6 are triggering most of sickness symptoms. Whereas, the cardinal inflammatory cytokine interferon- γ activates, indoleamine 2,3 dioxygenase (IDO), an enzyme mainly synthesized by phagocytic cells like macrophages and microglia. IDO activation causes tryptophan depletion and precipitates the depressive symptoms. On other hand, brain encysting with the bradyzoite is characterized by immunosuppression in form of reduced pro-inflammatory and heightened anti-inflammatory responses as evidenced by increased phagocytic activity and release of IL-10 and TGF- β . To conclude, *T. gondii* has multiple strategies to harness the host's immune mechanisms and behavior by the virtue of dual effects of tachyzoite-induced immune invasion intertwined with bradyzoite-induced immune evasion.

Background

The most prominent behavioral pattern of animals affected with viral, bacterial or parasitic infections is a set of organized symptoms called acute phase response or sickness behavior [1]. Sickness behavior is a normal host defense mechanism in form of an all-out effort against infection to keep the organism survival. If infection resolved, the sickness symptoms such as fever, lethargy, anorexia, reduced grooming and body's self-maintenance activities, gradually decline until vanish completely. It is assumed that sickness symptoms help enable the host to adapt better with an infection [2, 3]. In addition, sickness in response to infection is an adaptive change in an animal behavior, endocrine and autonomic nervous systems, triggered by stimulation of innate immune cells and release of pro-inflammatory cytokines such as IL-1, IL-6 and TNF- α [4]. The released cytokines from peripheral organs act locally on systemic organs and centrally on the brain to produce such well-coordinated response of sickness of the host to adapt with infection [1]. On other side, if infection does not terminate, depressive symptoms can arise within the context of sickness [5]. It has been reported that inflammation and its related cytokines mediate the interplay between the immune system and brain and involve in development of sickness and depression-related behaviors [4, 5]. Further, several experimental and clinical studies clarified the potential causal role of cytokines in depression and the effect of anti-cytokine therapy in improving depressive symptoms [6].

T. gondii as a unique model of an infectious disease paved the way for studying the interaction between immune and nervous systems. *T. gondii* is characterized by a persistence in the intermediate host by establishing the sedentary bradyzoite cyst in the cerebral and muscular tissues [7]. There is a general agreement towards the severity of behavioral changes caused by *T. gondii* is related to the brain tissue damage caused by the brain parasite burden and to the secondary changes of neurotransmitters

secretion and neuroinflammatory response [8]. Brain colonization with such a life long-lasting stage of *T. gondii*, seems to be one of the most crucial events which gathered a global attention of the public in multimedia and scientific community towards toxoplasmosis [9]. Infection with *T. gondii* is characterized by common behavioral changes such as increased exploration and lowered general anxiety, and predation that were not limited to felids [10]. On other hand, the parasite seems to have contrasting influences on the host, hence, a recent multi-parametric analysis revealed contrasting effects of *T. gondii* on the host behavior [9]. Thus, investigating the relationships among the host immune mechanisms and the developed physical symptoms of disease, anxiety, sickness-and depressive-like behaviors during toxoplasmosis may elucidate the role of infection-mediated inflammatory cytokine release and the parasite-host immune system interaction.

Here, we report this mini-review to address the following five-when inquiries; (i) when *Toxoplasma* infection is associated with heightened clinical signs and mortalities, (ii) when *Toxoplasma* infection is associated with diminished sickness symptoms and mortalities, (iii) when *Toxoplasma* -infected mice display sickness without depressive symptoms, (iv) when *Toxoplasma* get access in the brain to evade the immune system, and (v) When *Toxoplasma* manipulate the host by stage conversion strategy. This mini-review would answer these questions and provide an up-to-date summary of a sample of key studies that embodies the interplay among *Toxoplasma*, the immune system and the brain.

Methodology

Search Strategy and Data Extraction

A systematic search of the PubMed, Google Scholar, Cross Ref, Scopus, ISI web of Knowledge and Web of Science was carried out for articles published in English language until 20th September 2020. The search terms including indexing items as well as keywords to increase the output were *Toxoplasma*, latent infection, stage conversion, brain, behavior, and immunity. We also searched reference list of the state-of-the-art articles. We selected 74 research studies relevant to this review and wrote to some of main authors in the field for clarifying some of the negative or unpublished data.

1- When *Toxoplasma* infection is associated with heightened clinical signs and mortalities

The severity of illness or acuteness of toxoplasmosis is not only dependent on parasite strains but also on the immune status of the animal. For example, immunocompetent BALB/c mice overcome the acute phase of *T. gondii* infection and show an inverted-bell shaped curve for the number of clinical signs observed during the first three weeks of infection [11, 12]. The physical symptoms in immunocompetent BALB/c mice entail; arched back, erected fur, searching for warmness, sunken eyes, dropped upper eyelids, staggering gait, reduced locomotion, deficient touch and evacuation reflexes, and eventually the animal succumbs lying on belly [13, 14]. Whereas, in severe combined immunodeficient (SCID) and IFN- γ knockout mice always display the full repertoire of physical signs of toxoplasmosis, attaining the peak of signs at 10 days post-infection (dpi) and succumbing the acute stage within the second week of infection

[15–17]. Moreover, some studies suggest that changes in the host behaviors during *T. gondii* infection could be also related to the differences in mouse strain. For instance, Kannan et al. [18] reported in 7-months *T. gondii*-post-infected BALB/c mice, ME49 strain caused impairment of spatial working memory while Prognaud strain was associated with increased body weight and hyperactivity. Furthermore, provided that widespread brain pathology caused by infection with ME49 strain of *T. gondii*, immunocompetent C57BL/6 mice displayed motor incoordination in footprint test and reduced general locomotor activity but the spatial and recognition memories were not affected [19]. On other hand, no apparent clinical signs were recorded in immunocompetent BALB/c mice at 60 dpi with PLK strain of *T. gondii* [20]. Numerous studies illustrated that infection with *T. gondii* that does not cause mortalities may reveal changes in the behavioral repertoire of the rodent intermediated host, and induce manipulative changes that may increase the likelihood of horizontal transmission of infection [21]. Therefore, there is an apparent differences in the behavioral changes displayed by *T. gondii*-infected rodents that could be dependent on the parasite strain and animal species.

Besides the intensity of behavioral changes caused by *T. gondii*-infection in rodents may refer to the animal species or the parasite strain. Several research studies reported that there are some of these behavioral abnormalities are consistently found during *T. gondii* infection regardless of the animal species or the parasite strain such as; (i) memory deficits were observed in wild rats and emphasized by experimental approaches in both rats and mice [22–24], (ii) motor abnormalities were either in the form of lowered or enhanced generalized activities [25, 26], and (iii) reduced anxiety in open spaces, increased preference to novel food, and host manipulation in a form of diminished fear responses to cats' urine and fur [27–29]. Taken together, the immune potentiation of rodent host against *Toxoplasma* infection was associated with low mortalities, shortened acute phase responses and appearance of depression-related behaviors within the context of repertoire of physical signs of illness.

2- When *Toxoplasma* infection is associated with diminished sickness symptoms and mortalities

Bradyzoite is a slow replicating intracellular parasite-filled cystic form of *T. gondii* that has the potential to persist for the host lifespan. It has been reported that the elicited immune responses to *T. gondii* differ during the proliferative tachyzoite stage from those of the dormant bradyzoite stage [30]. It well-known that *T. gondii* strains have been classified according to its virulence into three distinct clonal lineages: high, medium and low virulent strains, or commonly written as type I, II and III of virulence, respectively [31]. However, a recently isolated strain of *T. gondii* belongs to type II clonal lineage named TgCatJpObi1 strain (Obi1), was identified, it has a robust ability of cyst formation within murine macrophages and human fibroblasts cells, without the exposure to stress factors required for bradyzoite induction [14]. It has also an *in vivo* ability of cyst formation in mice. Furthermore, Obi1 harbors higher gene expression of virulence factors, like Gra15, Rop5, 16, 17, and 18, when compared to ME49 strain, causes neither change in body weights, clinical signs of sickness, nor any mortalities in BALB/c mice [14]. To clarify this point, *T. gondii* undergoes genetic variability during the sexual reproductive stage of its lifecycle within the intestinal cells of feline main host, where shedding of diploid oocyst in the cat faeces which then undergo meiosis and sporulation in environment yielding haploid sporozoites [32]. Studied on *T. gondii* genetic

differences revealed that type I virulent strain harbor high levels of ROP18 and ROP16 (Reviewed by Hunter and Sibley [33]). Further, it was reported that within the same archetypical genotypes there are variations in some of the key effector proteins like rhoptries (ROP5, ROP16, and ROP18), and dense granular GRA3, and GRA15) families secreted by isolated *T. gondii* strains from stray cats [34]. Therefore, Salman et al. [14]. suggested that factors responsible for parasite virulence and those of bradyzoite cyst formation found in such field strain, are likely to affect independently the host. Or there may be other influencing factors, or additional genes that contribute to the differences in virulence between *T. gondii* genotypes [35, 36]. In line with this, a recent study has identified single factor that is only responsible for *T. gondii* bradyzoite cyst formation *in vivo* and *in vitro*, similar to a Myb-like transcription factor and abbreviated as BFD1 [37]. Therefore, to explain these mysterious characters of atypical strains of *T. gondii*, an additional experimental study with an extensive nature may be required. To sum, despite the identified virulence genes, diminished sickness symptoms and no mortality are exhibited by mice infected with a spontaneous cyst forming strain compared to the standard strain of the most similar clonal lineage.

3- When Toxoplasma-infected mice display sickness without depressive symptoms

Apart from the debate describing the depressive symptoms are either an adaptive or maladaptive to the Darwinian fitness, several retrospective studies simplified that the risk alleles responsible for depressive disorders are still existing and persisting in human population due to their significant role in host defense mechanisms against pathogens [38]. These embrace the combined set of immunological and behavioral responses of the host to fight infection and danger via recognition receptors called pathogen-associated, and damage-associated molecular patterns; PAMPs and DAMPs, respectively [38]. Furthermore, from the adaptive aspect, depressive behavior has been associated with acute phase response during the interaction of the host with the pathogen [39, 40]. During the course of *T. gondii* infection in immunocompetent mice, depressive symptoms appeared within the context of sickness signs. Therefore, there was some challenge in separating depressive symptoms from the physical symptoms of the disease, one strategy was to use a genetically modified animal model, as those deficient in innate or adaptive immunological responses to *T. gondii* infection such as severe combined immunodeficient (SCID), IFN- γ knockout mice in Table 1, Mahmoud et al. [16, 41] Rehan et al. [17] and TLR-2 knockout mice [41]. Reduced sucrose preference used to measure one of major depressive symptoms called anhedonia, and sometimes described as the hall mark of depression and forced swim test to assess despair-related behaviors [40]. Immunocompetent BALB/c mice during acute stage of *T. gondii*-infection exhibited anhedonia, assessed by reduced sucrose consumption, and despair-like behaviors, as evidenced by increased sedentary time in forced swim test [41]. However, in IFN- γ knockout (KO) BALB/c and Toll-like receptor (TLR)-2 KO C57BL/6 mice, both immune-deficient animals displayed neither despair-nor anhedonic-like behaviors, with or without *T. gondii* infection [17, 24]. In addition, TLR-2 KO mice exhibited an increased anxiolytic behavior in form of increased fear conditioning and reduced fear memory in conditioned fear test [24].

It could assume that heightened immune defense mechanisms were associated with depression-related behaviors during *T. gondii* infection, in support to this notion, *T. gondii*-infected BALB/ mice with type II colonial lineage strain (PLK) displayed increased anhedonic and despair symptoms, and enhanced conditioned fear responses during acute stage but not during chronic infection [12]. In parallel, deficiency of some components of normal behavior were also observed in immunodeficient mice model called an absent in melanoma 2 (Aim2) inflammasome, or mice deficient caspase 1 and caspase 11. Such mice showed anxiety-related behaviors during performing elevated plus maze and open field testing, but they performed normally in anhedonic and despair tests [42]. Taken together, these studies suggest that in case of *T. gondii*-infected immunocompromised mice, as a result of deficiency of either adaptive or innate immune components of immunity, displayed full repertoire of sickness symptoms, but reduced depression-related behaviors.

4. When *Toxoplasma gondii* get access in the brain to evade the immune system

In human and rodents *T. gondii* finds its convenience within the body by escaping the immune system to skeletal muscles and brain. Hence, nerve cells are the main focus of the neurotropic protozoan parasites *T. gondii* [43], so the brain is considered a predilection organ for the formation of lifelong bradyzoite cyst [44]. Therefore, infection mostly happens via oral intake of cyst or oocyst, rapidly liberates tachyzoites which penetrate the enterocytes and disseminate throughout the body of the host. An intracellular tachyzoite forms a membrane-bound parasitous vacuole (PV) and settles in immunologically protected sites like brain, muscles, retina and fetus [45–47]. *T. gondii* as an intracellular parasite secretes cyclophilin 18 (TgCyp18) which binds to a cysteine-cysteine chemokine receptor 5 (CCR5), CCL2 and CXCL10, that attract more leucocytes to the site of infection [48–50]. The released proteases process and produce proteins necessary for the penetration of parasite into the host cells [51]. After crossing the blood-brain barrier (BBB), tachyzoites settle in neurons, proliferate within the parasitous vacuole (PV) and then change to latent bradyzoite in different brain areas such as cerebral cortex and hippocampus in mice and human [52, 53]. In addition, *T. gondii* transmits vertically and results in a wide spread of bradyzoite cyst in brains of progeny or what is known as congenital toxoplasmosis [54]. Provided the existence of latent cyst in brains of congenitally infected mice, deficient short-term memory was not associated with changes in brain neurotransmitter concentrations [55].

On other aspect, *T. gondii* helps keep a state of balance or homeostasis with the immune system of the host. The parasite protects itself from host's inflammatory cytokines by creating the separating wall called parasitous vacuole (PV) [56]. A discharge of *T. gondii* organelles, called rhoptry (ROP) proteins, when released into cytoplasm of infected cells initiates the formation of PV, which would be ready to be occupied by *T. gondii* [57]. Secreted effector molecules of *T. gondii* and release of both pro-inflammatory and anti-inflammatory cytokines of the host are important to achieve this homeostasis with the host [47, 58]. Initially, *T. gondii* infection is manifested by release of inflammatory cytokines in acute phase followed by anti-inflammatory cytokines in latent phase of infection [59]. The, the production of IFN- α and IFN- β showed a simultaneous increase starting from 6 days post-infection (dpi) with *T. gondii* in the brain and 8 to 10 dpi in the spleen. Thereafter, an increase in IFN- γ expression was evident at 6 dpi in the spleen

but not before 10 dpi in the brain [41]. Where, the host major defense mechanisms against *T. gondii* multiplication depends on promoting natural killer (NK) and T-cell activities, and consequent IFN- γ production with subsequent activation of monocytes [60]. On other hand, after the parasite bypasses the acute stage, the production of antiinflammatory IL-10 and suppressor of cytokine signaling protein-1 (SOCS1) favors braking of the heightened immune responses and the survival of *T. gondii* within brain cells, and therefore help the persistence of bradyzoite cyst [60, 61]. To conclude *T. gondii* invade the nervous system in pursue to the immune-privileged environment. The integral host immunity and the parasite virulence factors are important to maintain this defense mechanisms of host-*Toxoplasma* balance.

5. When *Toxoplasma* manipulate the host by stage conversion strategy

To what extend *T. gondii* is a much an opportunistic parasite can be emphasized by its intrinsic ability to switch from tachyzoite to bradyzoite forms and *vice versa*. *Toxoplasma* perform stage conversion to match the host immune status and keep its survival. For instance, in immune-deficient conditions or under chronic immunosuppressive therapy, recrudescence of *T. gondii* occurs and is associated with severe symptomatic toxoplasmosis in human and encephalitis as in immunocompromised patients, AIDS in particular [62, 63]. Such evidence was reproduced after reactivation of chronic toxoplasmosis by two weeks-treatment with an immunosuppressant drug, dexamethasone in BALB/ mice. Despite bradyzoite marker Bag1 gene expression was not affected, Sag1, tachyzoite marker, mRNA expression showed a marked increase after dexamethasone treatment. Clear anhedonic and despair-related behaviors with minimal sickness symptoms were displayed by mice after recrudescence [20]. However, it was worthy to mention that the process of cyst rupture and formation may continue unabated even in the immune-competent host [61]. To conclude *Toxoplasma* get benefits and keep its survival with the host by such amazing reversibility from tachyzoite to bradyzoite stages and help manipulate the host immune system to reach immune balance (Fig. 1, [Ref, 64–66]).

Limitations

T. gondii-induced inflammation is likely to be suppressive to the immune brain environment. Few studies in mice showed that latent infection with *T. gondii* has positive or beneficial effects on brain dysfunctions or CNS insults. For instance, reduced stroke caused by permanent middle cerebral artery occlusion in chronically infected mice. That effect could be attributed to the immunosuppressive effect of *T. gondii* in brain environment via reduced nitric oxide production, or increased antiinflammatory cytokines production such as IL-10 and tumor growth factor- β [67, 68]. However, *T. gondii*-induced immune suppression would have mutual benefits gained by establishing the state of host parasite equilibrium. In addition, the protection against beta amyloid plaque deposition, in form of increased phagocytosis and degradation of amyloid beta in Alzheimer disease model, was found in chronic infection with ME49 strain but not with type II strain [69, 67]. The anti-Alzheimer effects were also specific to type II strain of *T. gondii* [70]. Therapeutics based on blocking the inflammatory response failed to prevent secondary infection with virulent strains of *T. gondii* [71]. Thus, further studies with intensive nature are still required to elucidate the mechanistic control of infection with this immune-encrypting protozoan parasite.

Future Directions

This mini-review focused on studying the crosstalk between immune responses and behavioral response during *T. gondii* infection in a mouse model, and to interrogate how much are the behavioral responses variable under the conditions of heightened and lowered immune responses. Many experimental trails for treating *T. gondii* have been focused on the acute course of infection, therefore, attention should be much paid to chronic, latent and congenital toxoplasmosis as they are considered the prolonged and predominant forms in human population [72]. Hence, latent infection with *T. gondii* has been described by immunosuppressive pattern within the brain. For example, several studies reported that chronic infection with *T. gondii* augmented anti-inflammatory cytokines IL-10 and TGF- β , and reduced nitric oxide (NO) productions [73, 74] Assessment of strains virulence of *T. gondii* on the basis of rodent host survival after infection with wild or field strains may still not enough criterion, an additional studies are required to deal with freshly isolated strains from the field. The therapeutic approach of using anti-cytokine treatment would be applicable. For instance, anti-inflammatory cytokine treatment may reduce the immune reactions and improve the *T. gondii* parasite burden in the brain.

Conclusion

This mini-review highlighted the manipulative role played by *T. gondii* on the rodent host and its crosstalk with host immunity with emphasis on a point; *T. gondii* subjugates the brain when succeed to set up this sedentary lifespan stage by bradyzoite encysting. If the activation of peripheral immune system continues unabated, as in case of immune-competent host, the conversion of *T. gondii* tachyzoite to bradyzoite cyst would be the ultimate strategy to escape such heightened host defense mechanisms. Finally, establishment of chronic infection in immunocompetent host by *T. gondii* in the form of bradyzoite cyst may outweigh the symptomatic stage of acute phase of infection. In fact *Toxoplasma* control or establishment of chronic infection is dependent on several factors such as those related to the host defense mechanisms, host species and genotype of infecting parasite strain.

Abbreviations / Acronym / Definitions

Abbreviation/Acronym	Definition
AMA	Amylopectin (glucose storage) granules
Aim2- inflammasome	An absent in melanoma 2 inflammasome
BBB	Blood-brain barrier
BAG1	Bradyzoite antigen-1, a bradyzoite marker of <i>T. gondii</i>
PAMPs	Pathogen- associated molecular patterns
CCR5	Cysteine-cysteine chemokine receptor 5
CCL2 or MCP1	The chemokine (C-C motif) ligand 2 or/ monocyte chemoattractant protein 1 or/ small inducible cytokine A2.
CXCL10	C-X-C motif chemokine ligand 10 or/ Interferon gamma-induced protein 10 (IP-10) or / small-inducible cytokine B10
PMA1	The P-type H ⁺ -ATPase
DAMPs	Damage-associated molecular patterns/ or danger- associated molecular patterns
ENO	Enolase
TgCyp18	<i>T. gondii</i> cyclophilin 18
dpi	Days post-infection
TgIF	Eukaryotic initiation factor
ELISA	Enzyme-linked immunosorbent assay
GRA(s)	Dense granules of <i>Toxoplasma gondii</i>
5-HT	5-hydroxytryptamine, or serotonin
HVA	Homovanillic acid, a dopamine catabolite
IL	Interleukin
IFN-β	Interferon-beta
IFN-γ	Interferon-gamma
IDO	Indoleamine dioxygenase
LDH	Lactate dehydrogenase
KO	Knockout
MIC	Microneme
MyD88	Myeloid differentiation primary response 88

NO	Nitric oxide
NK	Natural killer
PV	Parasitophorous vacuole
ROP	rhoptry organelles
SAG1	Surface antigen-1, a tachyzoite marker for <i>T. gondii</i>
SCID	Severe combined immunodeficiency
SOCs	suppressor of cytokines signaling 1 protein
SRS	SAG1-related sequence
TNF- α	Tumor necrosis factor – alpha
TGF- β	Tumor growth factor-beta
<i>T. gondii</i>	<i>Toxoplasma gondii</i>
TLR	Toll-like receptor

Declarations

Author Contributions

M.E.M. and Y.N. developed the idea of review, and M.E.M wrote the first draft. All authors read, revised and agreed the submitted version of the manuscript.

Conflict of interests

All authors declare the absence of any conflict of interests.

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Tables

Table 1 Comparison between chronic and acute stage of infection with <i>Toxoplasma</i> in immunocompetent and immunocompromised conditions				
Chronic stage in immunocompetent host	acute stage or reactivation in immunocompromised host	Parasite strain	Intermediate host	Reference
Bradyzoite stage dominates; elevated Bag1 expression	Tachyzoite stage dominates evidence; increased Sag1 expression	Type II, PLK	Female BALB/c	Mahmoud, et al., 2016
Proinflammatory cytokines return to basal level	Increased IFN-IL-6 and IFN- γ	Type II, PLK	Female BALB/c	Mahmoud et al., 2015
Depressive symptoms appear within the context of Sickness symptoms	Depressive symptoms are covered by Sickness symptoms	Type II, PLK	Female BALB/c	Mahmoud et al., 2015
Inhibition of neuronal function via bradyzoite	Inhibition of neuronal function via tachyzoite			Haroon et al., 2012
Intracellular bradyzoite in brains brain cells have high levels of dopamine	homovanillic acid (HVA)/ dopamine, a catabolite of dopamine, was increased			McConkey et al., 2013, Mahmoud, 2016
unchanged IDO activity in mice	Enhanced IDO activity in mice			Mahmoud et al., 2016
Kynurenine/ Tryptophan (Kyn/Trp ratio returned to basal level) in mice	elevated Kyn/Trp ratio in mice		Female BALB/c	Mahmoud et al., 2016
		Type II, PLK	Male C57BL/6 mice	Ihara et al., (2015)
Absent anhedonic and despair-related behaviors	Existent depressive symptoms over a background of sickness	Type II, PLK		Mahmoud et al., 2017
Gut flora have an increase of Bacteroidetes compared with controls	Gut flora have potentiated increase of Bacteroidetes compared with controls			Severance et al., 2018
Brain inflammation and perivascular cuffing	Intracerebral cyst rupture, and increased inflammation and perivascular cuffing after reactivation	Type II, ME49		Jost et al., 2007, Martynowicz et al., 2019
Brain (mainly in hippocampi) and spleen have highest parasite load in C57BL/6 mice	Brain (mainly in amygdala) in C57BL/6 mice	Type II, ME49		

Bodyweight loss in mice	Type II, ME49	Female BALB/c and Male C57BL/6	Gatkowska et al., 2012, Salman et al., 2020
Abbreviations; interleukin, IFN: interferon, Kyn: kynurenine, Trp; Tryptophan			

Figures

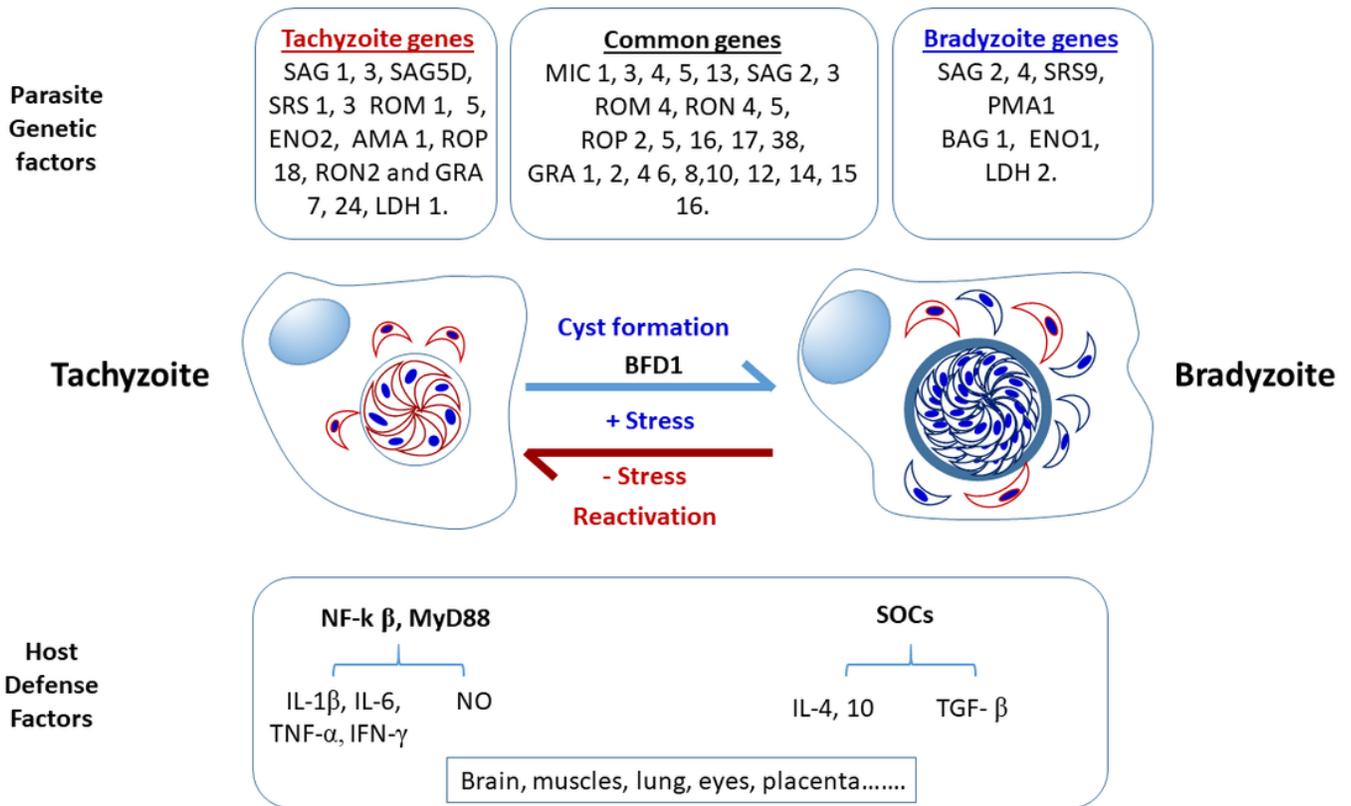


Figure 1

Parasite-host immune-balance. After *Toxoplasma* infection is tachyzoite stage predominates the acute phase response, bradyzoite cyst formation happens via BFD-1 if the immunocompetent intermediate hosts bypass the acute stage. In case of lowered host defense mechanisms reactivation of tachyzoites occur (Ref. 62-64).