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COMPARATIVE ASPECTS REGARDING THE PRESENCE OF BEHAVIORAL CHANGES OF AGGRESSIVE TYPE IN SEROPOSITIVE *TOXOPLASMA GONDII* INDIVIDUALS - HUMANS AND CATS

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Abstract

The aggressive behavior encountered in the feline patient, represents a major problem in the approach and care handling for this patient, hard quest for a complete clinical and neurological evaluation, also in applying some investigation methods and running a full internal and external set of analysis: blood workouts, ultrasounds, X-rays, MRIs. In this study, we searched for triggers, causes and effects that give the aggressive state of the feline patient and we found that toxoplasmosis, hyperthyroidism, epilepsy, dental pain, arthritis, discomfort from coinfections with other bacteria, viruses, parasites, trauma, lack of sensitive reactions and cognitive response malfunctioning in geriatrics, could all contribute for this behavior. In this comparative study of human, feline and other mammals with latent infection with T. gondii, we found common aggression traits presented in some psychiatry studies linked to the human latent infection, as well as in feline and other mammals brain studies with toxoplasmosis confirmed with different laboratory technics.

Key words*: aggressivity, serology, test panel, coinfections.*

INTRODUCTION

There are different types of feline aggressivity: Among cats (eg 2 males, mating of fighting for territory); in fear or defensive (danger: a person, animal, object or sound based); for territory (other cats passing by, or even dogs or humans by selection, new cat, change of territory, new human in the family); playful type (could easily turn into a fight with deep marks and broken house objects); redirected type (most dangerous, has one target - animal or human- but if the cat cannot reach it, it will attack the next thing coming, other cat or human); by over-petting (when it is too much, one should better stop the petting- the cat will let you know); induced by pain or other stimuli (inflammatory muscles, osteoarthritis, frustration, closed environment for too long, otitis, dental or fight injuries); The maternal type (protective mothers will attack- contact in the first days of litter should be avoided); Idiopathic type (explained after thorough medical exams, clinical history of the pet – they easily attack, bite the owner, the hyperexcitability status is high for long periods of time – very important to establish the difference between redirected and idiopathic aggression features, for it is decreases the comfort of both cat and owner for sharing the same environment – many of them end up being euthanized by request; The hunting type is easily understandable, even between the kittens in their play time.

Common features of aggression found in humans and cats

Based on studies performed on humans and cats, it is a proven fact that there are common traits of expressed aggressive behavior, witch in time these types of aggression got classified more extensively, but we can speak about psychiatric disorders like: impulsivity, violence, chronic depression, self-inflicted or suicidal tendencies. Gregg & Siegel (2001) studied aggressive behavior on criminals and thieves, first class with a predator aggressive features, second with defensive reactions. They studied the neurological pathways of aggression and found that medial and lateral hypothalamus, the amygdala and the central grey periapeduct activity are responsible for this behavior. Amygdala is known to be the center for processing information regarding sensitive, visceral, interceptive, cognitive and

emotional stimuli, and the role of being able to change the animal's behavior via hypothalamus and PAG accordingly. Berenreiterova et al. (2011) has a study on rats infected with *T. gondii*, that suggests that the tropism of the parasite's cysts is distributed mainly in amygdala, hypothalamus, olfactive area, sensitive and motor areas, but the localization was not restricted just for these areas, so it is not preferential.

***Toxoplasma gondii*. Short general characterization**

Antropozoonotic parasitic agent, a unique protozoa member of *Toxoplasmatinae* Subfam., *Sarcocystidae* Fam., *Eimeria* Subord., *Eucocidia* Order, *Coccidia* subclass, *Sporozoa class*, *Apicomplexa* Phillum. *T. gondii* is a 5 μm parasite, it infects nearly one third of the global population of individuals at this moment, 80% of the French population, 60% of American population as studies suggest. In our country there are no actual per country screening or other registered information about the prevalence of *T. gondii,* except some local and the ones found among pregnant women region surveys (44%) (Jaroslav et al., 2014). And for the cats we are trying to have a more elaborate picture of this influential parasite.

Infection pathway of *T. gondii* in the intestinal wall and the immune response of the host

When parasites take over the intestinal wall. they break the tight bound between the enterocytes. This parasite uses different strategies to infest the enterocytes, followed by strong reactions like the release of nitric oxide, some cytotoxic molecules causing neuropathic damage. Enterocytes then release chemokines and cytokines attracting leukocytes, macrophages and dendritic cells (activating self-immunity and their microbiocidal direct function). Interleukins like IL-12 activate CD4 immune adaptative response.

T lymphocytes with NK cells and NKT, in the presence of IL-15, release IFN γ activating the defense mechanism for expelling the parasite out of the enterocytes. B cells are activated also for releasing antibodies that could cross the epithelial barrier by transcytosis and reach the lumen. Excepting the bactericidal factor, if IFNγ is uncontrollable in its release, it could damage the integrity of intestinal epithelium. The intraepithelial lymphoid cells are cytotoxic for the infected enterocytes and may produce TGF - β limiting the production of IFN-γ (Schulthess, J. et al., 2008).

Immune response in neurotransmitted *T. gondii*

T. gondii acute phase activates a powerful response with the release of IFN γ, IL-12 and CD8+ T lymphcells. The production of IFNγ is most essential for stopping *T. gondii* tachizoytes replication, it controls the infection in CNS and prevents toxoplasmic encephalitis, easily transforming them in cysts without letting them spread. For triggering this pathway, *T. gondii* has a gene called profiline, inducing the expression of IL-12 (Plattner et al., 2008). Same IFNγ maintains the low number of tachizoytes and prevents the reactivation of the cysts in the tissues at the time of infestation. IFNγ has its own secondary clinical depression side effect when used as treatment for antitumor purpose, it degrades the tryptophan, the main precursor for serotonin, the one responsible for the depressive state. *T. gondii* infestation upon IFN's secretion could affect indirectly some cognitive functions. Graeff's work says that the serotonin level could be having a role in the initiation of the fear behavior, but this is not always proven fact, for the animals or humans. Dynamic of antibodies response. The medium dynamics of different isotypes, but their level can differ from patient to patient and used serological technique. IgM can be detected even years after initial detection (Figure 1) (Robert-Gangneux, Florence, and Marie-Laure Dardé, 2012).

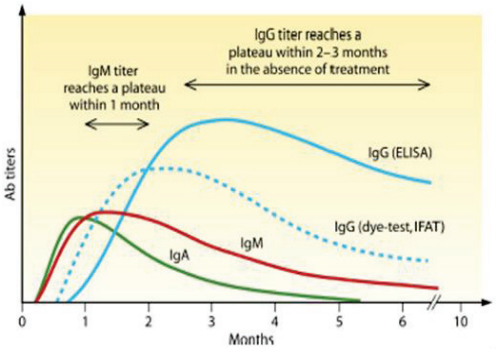

Figure 1. *T. gondii* antibody response by Robert-Gangneux, Florence, and Marie-Laure Dardé, 2012

MATERIALS AND METHODS

For the in-house analysis in the Veterinary medical laboratory of Faculty of Veterinary Medicine, sampling and processing of feline samples, a fresh sample was worked on, without refrigeration, almost instantly.
For testing and detection of IgG antibodies of *T. gondii* in domestic cats either serum, plasma or blood was used, samples being processed by specialized personnel, schooled in the authorized laboratory and according to protocols and operational procedures.
The period taken into account for testing IgG antibodies for *Toxoplasma gondii* was between 01.01.2015 - 13.05.2018.
The total number of processed samples for qualitative detection of IgG antibodies against *T. gondii* was of 544 felines, presented in the clinic, for different affections: neurology and coordination with paralysis or limping, cardiology, ophthalmology, stomatology, or kept under surveillance and treatment for general, internal or metabolically diseases.
Toxoplasma/Chlamidophyla determinations are based on antigen-antibodies reactions.
Having as principle an immunoenzymatically method, **IMMUNOCOMB** is a plastic device, resembling a comb, on which **Toxo/Chlamidophyla** is fixed.
The serum, plasma or blood samples are place in the wells of the multicompartment developing plaque.

RESULTS AND DISCUSSIONS

For the purpose of determining the IgG Toxoplasma or Chlamydia titer, sample color intensity is compared on the corresponding dents of ImunoComb with the series of spots on its scale (included in the kit). The inferior spot represents *Toxoplasma* test. The middle spot represents *Chlamydia* test. Results for each disease is evaluated separately. Sample color intensity is compared with that of Positive Control (C+) included in the kit, for determining the titer. Positive Control for *Chlamydia* and *Toxoplasma* is calibrated at a 1:32 titer. The samples with a higher or equal color intensity with that of the positive control are considered positive.

RESULTS AND DISCUSSIONS

From the total of 544 domestic feline tested for IgG *Toxoplasma* antibodies detection, 258 were positive and 286 negative, test made during 01.01.2015-13.05.2018 in the Faculty of Veterinary Medicine of Bucharest Laboratory.

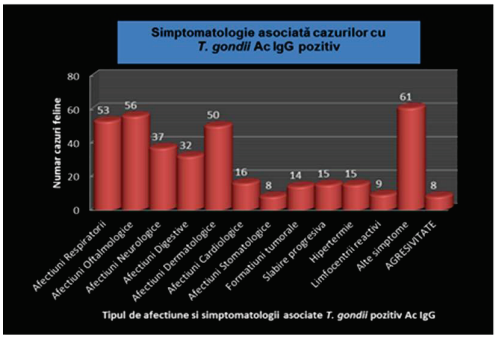

Figure 2. Main clinical signs in the presence of *T. gondii* IgG positive

In describing the symptomatology, for each category, we have extracted significant data in the moment of patient presentation, by clinicians, as follows (Figure 2):
Respiratory affections: cough, dyspnea, bronchitis, infections of the superior respiratory tract (nasal secretions, sneeze, blockage with ocular connections, often with feline infectious rhinotracheitis caused by type I Herpesvirus), broncho-pneumonia, interstitial pneumonia, infiltrations at broncho-alveolar level, pharyngitis, laryngitis, often associated with *Streptococcus, Pasteurella or Chlamydophila*.
Ophthalmological affections: *T.gondii* specific uveitis, severe secretions, anisocoria, iritis, corneal ulcerations, cataract, kerato-conjunctivitis, glaucoma, sinechia, iris vaso-dilatations, tapetum negrum hyperpigmentation, crystalin anterior chamber deposits, corioretinits, blepharitis.
Neurological affections: often have continued the ophthalmological tableau with mydriasis, photophobia, delayed attention, menace, other neuropathies were described as facial paralysis, balance loss, delayed proprioception on some limbs, grand mall type seizures, muscular tremor, exaggerated aggressivity or affection, depreciation alternated with excitability, affection of some cranial nerves, neuronal

parameters modifications, vestibular syndrome, spasms, dromomania, encephalitis.

Digestive affections: can be caused by the reaction of the digestive mucosa at the moment of parasite entry, or by association with Coronavirus or any other opportunist virus, *Helicobacter* dormand or ascarids, hematophague by egg ingestion, excessive licking.

Dermatological affection: pruriginous dermatitis, especially in areas where licking is impossible: itching, otitis, hypertrichosis (lateral, abdominal or on limbs), ulcerations, wounds, grating, eosinophilic complex, all being related to the activation phase of IgE given by the presence of *T. gondii*.

Cardiological affections: encountered in positive IgG antibodies for *T gondii* - arterial hypertensions, aortic, septum and left ventricle hypertrophia, congenital, cardiomyopathy - testing through SNAP pro-BNP is recommended.

Stomatological affections were gingivitis, some proliferative, stomatitis, plaque and halitosis, which appeared in associations with a *Calicivirus*, retrovirus FIV or FeLV.

Tumors: frequent in association with seropositivity were mammary tumors, abdominal formations, fribrosarcoma, squamocellular carcinoma, hepatic.

Progressive weight loss: manifested due to depreciated state by the parasitic infection, intestinal discomfort, pancreatitis, associated gastritis, biliary sludge, cholestasis, steatosis, angiocolitis, neurological or thyroid disfunctions.

Not rarely, there are mentioned falling from the balconies of the houses, blaming sexual instincts, birds chasing, exploring the surroundings, but what if the parasite impairs the equilibrium ways, the sight with its **ophthalmic deposits**, what if the topography of the cysts are affecting the decision making side of cognitive functions, amygdala, hypothalamic area could be affected by the cysts insertion, the inflammatory responses and lesions of the chemical imbalance they create.

Also, trauma and hormonal changes, such in **GnRH cells**, they might lose their original place once the **cysts** could trigger a mixt signal for secretion for **more androgenic** hormones and **less estrogens** and **progesterone**, the result might turn into an **aggressive behavior** (Figure 3).

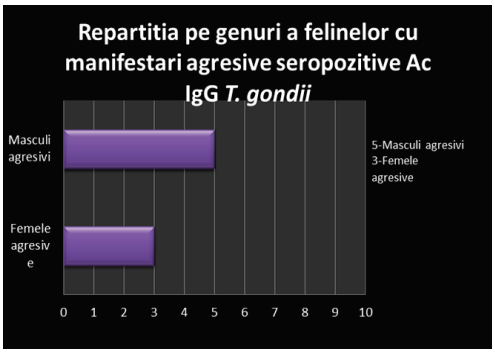

Figure 3. Gender distribution for positive IgG *T. gondii* with aggressive behavior

Because of such hormonal changes, the imbalance could cause **infertility**, **abortion** can be present in some cases, kittens can be severily immunodepressed at birth, therefore any viral agent could be fatal. *Herpesvirus* is active, they could go blind, especially if they are sold very early from their mother and the maternal immunity fails to be fully installed. From our analysis after serology testing of latent *T. gondii*, we conclude that from the point of age, cats were mostly manifesting symptoms between **11 months** and **2 years** of age. For the old ones, the number of testing itself drops, because they usually come for checking bigger issues like **tumors**, **degenerative processes**, or the situation where they stop coming. The youngest we've tested, and got a positive, at 2 months of age, also **positive** on *Coronavirus*, and 2-year-old ones, we can blame all this on a poor immunity settled. If the IgG antibodies are mainly transmitted from the mother, then the antigens for *Coronavirus* come from external grounds, an opportunistic virus that could be fatal.

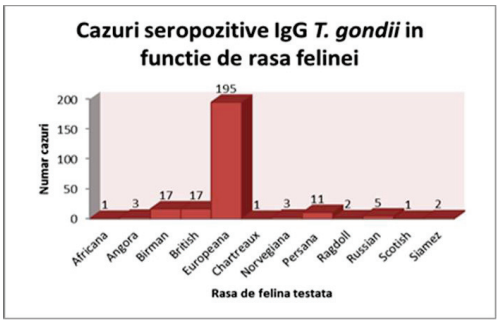

Figure 4. Breed distribution for *T. gondii* IgG

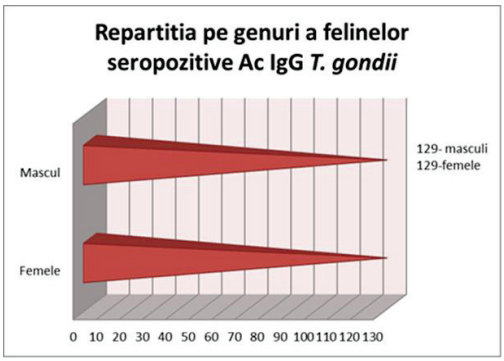

Figure 5. Gender distribution for *T. gondii* IgG

The 8 positive felines for latent *T. gondii* that manifested an aggressive behavior are the ones that the blood collection could be performed. Most definitely, the percentage is bigger if at least once in a cat's lifetime we test for these *T. gondii* antibodies, we recommend testing as soon as the cat gets into a household, for *T. gondii* is a zoonosis. These positive cats were very aggressive, some with epileptic crisis, anisocoria and ocular discharge, otitis, one was positive on latent *Coronavirus*, and some were said to be consuming fresh, uncooked cattle meat. On the biochemistry panel, a high Glucose was registered as a pattern of distress and may also from inflammation, also from a high level of glutamate changing the biochemistry of the nervous central system infested with the parasites.

For the serology testing we used immunochromatography tools same as mentioned for latent *T. gondii*, like **SNAP Combo FelV/FIV™** and also latent **IgG *Coronavirus*** fast test, with one possitive test of latent ***Calicivirus*** confirmed by the owner of one of the cats, we mentioned it for it could also decrease the immunity barrier of that cat, making room for other coinfections to manifest. For **FCoV, FPV** and **Giardia** and for **SNAP Giardia™ antigens** we worked with fresh **feces samples**, with same testing protocol described for any other **qualitative test**. For any enteropathy disorder we recommended this testing and we had a great prevalence of viral infections, the only problem was that we could **not test for *T. gondii* antigens** and neither the microscopical exams for feces, but this is part of a futured protocol of catching this parasite in action at the site, where it riches the enterocytes and then passing through all the barriers, masking itself in immunity cells. As it is mentioned, there is a great amount of 2 or more pathogens in association, the symptomatology is common for all of them and might not be tested at a first consult. The owners usually come either to soon, either too late when all internal organs are severely compromised: renal failure, hepatic damage, cardiopathies, pneumonia, etc. From the **negative cases (286)** for latent *T. gondii*, **4** of them were aggressive, but the cause for the aggressivity came from elsewhere, as it follows.

One **case of aggressive** feline tested **negative** for latent ***T. gondi***i and *Chlamydophila* spp., but had a caught based on the microbiology swab sample that grew positive for Streptococcus spp. and *Pasteurella* spp. Other pathogens were not asked for testing.

One case also **negative** for *T. gondii*, was a feline with renal lithiasis, discovered on the ultrasounds, assuming the pains were on a high level so the discomfort made the cat be aggressive idiopathically.

One case had a **positive *Criptococcus neoformans***, diagnosed by MV phd student Dobre Patricia from the Pathological Anathomy Dep., 2018, after extracting a CSF from the cat and analyzing it with cytology, the necropsic exam discovered multiple lesions in the cerebral tissue. We tested the fluid together and found a negative result for **FCoV IgG** and a **negative antigen for Rabies**.

The cat was euthanized for it was very dangerous to be around people, it attacked it's owners and everyone around. As a contamination way was supposed to have been pigeon feces from the balcony of the owner's house. And **the last case** was a fall from the balcony, **constipated** so again the disconfort factor present, both latent *T. gondii* and *FCoV* were **negative**, and with no other symptoms.

For the protocol to be complete in testing for *T. gondii,* we recommend testing in the laboratory for feces microscopy and **antigens, the antibodies, IgM, IgG** in the blood and if the symptoms persist, and neurological signs do not disappear**, PCR** from **CSF** is in order, to surprise any fragment of the parasite. If **MRI** with some contract substance could reveal the cysts, we could soon be able to understand better were they are located and maybe how

they affect the functions of our patients, and then treat accordingly.

CONCLUSIONS

The aggressive behavior remains a common subject for future study, both in veterinary neuropathies and human neuropsychiatry, and also a very important approach for the means of contention for investigating, applying a therapy and especially settling some living arrangements with its owner.

Research on damaged cerebral area and tissue, given by this parasite, had been shown to choose a special placing in the amygdala, hypothalamic, retrobulbar, areas responsible for processing the received signals, the response to them from different cortical areas, for cats or humans, and even for different other species naturally or artificially infected in the laboratory. The presence of *T. gondii* parasite disrupts neural mechanism, cognitive, visual, changes the brain biochemistry in the way that might affect the survival skills set of an individual, homeostasis and hormonal balance, involving decision making when to retreat, attack, defense, impulsivity or even self-harm. From our current study we can conclude that *T. gondii* has no gender affinity, that **common European race** is mostly predominant in testing numbers for positive latent *T. gondii,* the symptoms connect among themselves for all the epithelial surfaces, the respiratory with ocular and dermatology, where all the eosinophilic reactions take place, and the cycle is never interrupted without a proper treatment, we wonder if the toxoplasmic cat changes the symptoms when stressed, if a IgM reaction to the parasite reappears in the blood stream, or it changes biochemistry where the cysts are located. Co-infectious with viruses and bacteria could easily be found in immuno- depreciated by *T. gondii* cats, there might be a way to trigger a new wave of cysts replication, this is only a future idea to follow, but for sure it makes it easier for the new pathogen to insert. When in consult, after taking the patient's history, obvious symptomatology, vaccination status, temperature, dehydration status, dietary habits, plants in the house, other incidents or new cat, or any animal in the parameter, trauma, hormonal status, new changes in behavior, we recommend a full serology screening of the animal, especially when aggressive. After a full history panel of checking for latent and antigen forms of any viruses, parasites and bacteria available in the laboratory, if the symptoms are persistent and a certain disease might be present, it's mandatory that molecular biology should be next on the testing protocol, that is why we insist on the good communication between the veterinarian and the laboratory staff.

REFERENCES

Abdoli, Amir. (2014). *Toxoplasma, testosterone, and behavior manipulation: the role of parasite strain, host variations, and intensity of infection*. Frontiers in biology 9.2: 151-160.

Barnard, Christopher J. (2011). *Animal behaviour: mechanism, development, function and evolution*. Pearson Education

Berenreiterová, Miroslava, et al. (2011). *The distribution of Toxoplasma gondii cysts in the brain of a mouse with latent toxoplasmosis: implications for the behavioral manipulation hypothesis*. PloS one 6.12: e28925.

David, Clément N., et al. (2016). *GLT-1-dependent disruption of CNS glutamate homeostasis and neuronal function by the protozoan parasite Toxoplasma gondii. PLoS pathogens* 12.6 e1005643.

Fallahi, Shirzad, et al. (2017). *A literature review on maternal-fetal and reproductive disorders of Toxoplasma gondii infection*. Journal of gynecology obstetrics and human reproduction.

Gregg, Thomas R., and Allan Siegel. (2001). *Brain structures and neurotansmitters regulating aggression in cats: implications for human aggression*. Progress in Neuro-Psychopharmacology and Biological Psychiatry 25.1 (2001): 91-140.

Hetts, Suzanne (1999). Pet behavior protocols: What to say, what to do, when to refer. *AAHA press*, 1999.

Hunsperger, Robert W., and Verena M. Bucher. (1967). *Affective behaviour produced by electrical stimulation în the forebrain and brain stem of the cat*. Progress în Brain Research. Vol. 27. Elsevier, 103-127.

Joiner, Keith A., and David S. Roos (2002). *Secretory traffic in the eukaryotic parasite Toxoplasma gondii: less is more*. J cell Biol157.4: 557-563.

Julio-Pieper, Marcela, et al. (2011). *Exciting times beyond the brain: metabotropic glutamate receptors in peripheral and non-neural tissues*. Pharmacological reviews 63.1:35-58.

Mathai, Ashwin Jacob, et al. (2016). *Reciprocal moderation by Toxoplasma gondii seropositivity and blood phenylalanine–tyrosine ratio of their associations with trait aggression.* Pteridines 27.3-4: 77-85.

McConkey, Glenn A., et al. (2013). *Toxoplasma gondii infection and behavior–location, location,*

location? Journal of Experimental Biology 216,1 (2013): 113-119.

Mitra, Rupshi, Robert Morris Sapolsky, Ajai Vyas (2013). *Toxoplasma gondii infection induces dendritic retraction in basolateral amygdala accompanied by reduced corticosterone secretion.* Disease models & mechanisms 6.2: 516-520.

Oktenli, Cagatay, et al. (2004). *Transient hypogonadotrophic hypogonadism in males with acute toxoplasmosis: suppressive effect of interleukin-1â on the secretion of GnRH*. Human Reproduction 19.4: 859-866.

Prandovszky, Emese, et al. (2011). *The neurotropic parasite Toxoplasma gondii increases dopamine metabolism*. PloS One, 6.9: e23866.

Robert-Gangneux, Florence, and Marie-Laure Dardé (2012). *Epidemiology of and diagnostic strategies for toxoplasmosis.* Clinical microbiology reviews, 25, 2: 264-296.

Schulthess, J., et al. (2008). *Mucosal immunity in Toxoplasma gondii infection*. 389-395.

Mendez, Oscar A., Anita A. Koshy. (2017). *Toxoplasma gondii: Entry, association, and physiological influence on the central nervous system.* PLoS pathogens 13.7 (2017): e1006351.

Xiao, Jianchun, et al. (2014). *MicroRNA-132 dysregulation in Toxoplasma gondii infection has implications for dopamine signaling pathway*. Neuroscience 268: 128-138.

http://vetexpert-rf.ru/express-test/toxoplasmaAb/

https://phys.org/news/2017-12-toxoplasmosis-exploits-immune-cells-brain.html#jCp

https://journals.plos.org/plospathogens/article?id=10.1371/journal.ppat.1006739

https://doi.org/10.1371/journal.ppat.1006351.g001

http://www.msunites.com/toxoplasma-gondii-parasite-and-neurological-disorders-research/