

# Seroprevalence of *Toxoplasma gondii* in donkeys (Aquus asinus) from Grenada, West Indies

Kathryn O'Connell<sup>1</sup>, Alfred Chikweto<sup>1</sup>, Keshaw Tiwari<sup>1</sup>, Mahesh Deokar<sup>2</sup> and Ravindra N. Sharma<sup>1</sup>

<sup>1</sup>Pathobiology Department, School of Veterinary Medicine, St. George's University, Grenada, WEST INDIES <sup>2</sup>Anatomy, Physiology and pharmacology Department, School of Veterinary Medicine, St. George's University, Grenada, WEST INDIES

Corresponding author: RN Sharma; Email rsharma@sgu.edu

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#### ABSTRACT

The objective of the present study was to estimate seroprevalence of *Toxoplasma gondii* in donkey from Grenada, West Indies. Using a modified agglutination test (MAT), sera of 37 donkeys were examined for evidence of exposure to *T. gondii*. Two (5.4%; 95 percent confidence interval: -1.88% to 12.68%) of the 37 donkeys were seropositive with titers of 50 in one donkey and 100 in the other. The lower Seroprevalence coupled with non consumption of meat and milk from donkey in Grenada suggests that donkeys are not important in the epidemiology of *T.gondii* in this country.

Keywords: Toxoplasma gondii, seroprevalence, donkeys, Grenada

*Toxoplasma gondii* is an obligate intracellular parasite that causes the zoonotic disease, toxoplasmosis. About 30-50% of the world's population is estimated to be seropositive (Flegr *et al.* 2014; Furtado *et al.* 2011) All warm-blooded animals including mammals and birds are susceptible to *T. gondii* (Robert and Darde, 2012). Cats are important in the life cycle of *T. gondii* because they are the only known definitive hosts capable of shedding environmentally resistant oocysts in nature (Dubey, 2010). Ingestion of oocysts shed by cats is the main source of infection for herbivores, where tissue cysts develop in infected animals

Humans can get infection through ingestion of oocysts, in contaminated water, vegetations and soil or from milk from Toxoplasma positive goats and donkeys (Dubey, 2010; Francesca, 2014) or from tissue cysts contained in undercooked meat of Toxoplasma positive animals and birds (Flegr *et al.* 2014). In most cases, infection with *T. gondii* is usually asymptomatic in humans with a competent immunity. Although primarily asymptomatic in healthy humans, toxoplasmosis is responsible for 10-30% of fatality in AIDS patients and if first contracted during pregnancy it can cause abortion or congenital problems in

the fetus. [(Flegr *et al.* 2014; Furtado *et al.* 2011; Robert and Darde 2012; Dubey 2010).

Infection of donkeys with *Toxoplasma gondii* has been reported in Italy (Francesca *et al.* 2014; Machakova *et al.* 2014), Turkey (Balkaya *et al.* 2011), China (Na Yang *et al.* 2013; Miao *et al.* 2013), Brazil (De Oliviera *et al.* 2013), Egypt (El-Ghaysh, 1998; Fauad *et al.* 2010), USA (Dubey *et al.* 2014), Mexico (Cosme *et al.* 2015) and in Spain (Garcia *et al.* 2012)

To our knowledge, there has been no report of *T. gondii* in donkeys from the Caribbean region. The aim of this study was to determine the seroprevalence of *T. gondii* in donkeys from Grenada, a small island country in east Caribbean.

#### **MATERIALS AND METHODS**

#### **Geographic location of Grenada**

Grenada, Petite Martinique and Carriacou make up the triisland country of Grenada, located in the southern end of



the West Indies, about 160 km north of Venezuela. The climate is tropical with a wet and humid season occurring between June and December.

## Sample collection and serology

Approximately 5 mL of blood was collected from each of 37 donkeys in Carriacou by jugular venipuncture and allowed to clot. All samples were centrifuged at 1000 g and serum was separated and stored at -20 °C until tested at the diagnostic laboratory of the St. George's University, School of Veterinary Medicine. The samples were collected between September and October during 2013 and 2014 from Carriacou. This is the only part of Grenada where sufficient numbers of donkeys are domesticated. Serum levels of *T. gondii* IgG antibodies were detected using the Modified Agglutination Test (MAT) as described by Dubey and Desmonts (1987). Sera for the MAT test were diluted two-fold serially from 1:25 to 1:3200.

## **RESULTS AND DISCUSSION**

Antibodies to *Toxoplasma gondii* were found in 2 (5.4%; 95 percent confidence interval: -1.88% to 12.68%) of 37 donkey serum samples tested, with titers of 50 in one donkey and 100 in the other.

A low to very high seroprevalence of *T.gondii* antibodies in donkeys have been reported from different parts of the world. Very high seropositivity was found in Egypt (45 % in serum and 46.3% in milk (Fouad et al. 2010). In another study in Egypt, El-Ghaysh (1998) found 65.6% positivity in blood. Very high Seroprevalence has also been reported from Brazil, 43.2% (De Oliviera et al. 2013) and Turkey 62.0% (Balkaya et al. 2011). Moderate Seroprevalence was found in China 20.3% (Miao et al. 2013) 23.6% (Na yang et al. 2013) Italy 25.0% (Francesca et al. 2014) and Spain 20.0% (Garcia et al. 2012). Similar to our study, low seroprevalence of T.gondii in donkeys was reported from the USA 6.3% (Dubey et al. 2014) Mexico, 10.9% (De Oliviera et al. 2013) and Italy, 5% to.8% (Francesca et al. 2014). The variation in seroprevalence in different counties may be due to methods of husbandry, and contact of cats with donkeys and their feed. We are not aware of the status of cats on the island of Carriacou, however food animals from Grenada and Carriacou had higher seroprevalence rates (Dubey et al. 2005; Chikweto et al. 2011; Sharma *et al.* 2015). It is possible that husbandry and the environment of the donkeys could limit their contact with cats, infected feces or contaminated feed sources.

 Table 1: Studies on seroprevalence of T.gondii antibodies in various hosts in Grenada, WI

Host	Seroprevalence (%)	Reference
Pregnant woman	57.0	Asthana et al. 2006.
Pet cat	36.0	Dubey et al. 2009
Feral cat	28.0	Dubey et al. 2009
Free range chicken	52.0	Dubey et al. 2005.
Rat	0.8	Dubey et al. 2006
Owned dog	25.0	Sharma et al. 2014
Stray dog	33.4	Sharma et al. 2014.
Goat	42.8	Chikweto et al. 2011
Cattle	8.4	Chikweto et al. 2011
Sheep	44.1	Chikweto et al. 2011
Pig	24.3	Sharma et al. 2014
Mongoose	17.0	Choudhary et al. 2013

Prior studies conducted on *T. gondii* antibodies from various animal species and pregnant women in Grenada showed a wide range of seroprevalence rates (Asthana *et al.* 2006; Dubey *et al.* 2009; Dubey *et al.* 2005; Dubey *et al.* 2006; Sharma *et al.* 2014; Chikweto *et al.* 2011; Sharma *et al.* 2015; Choudhary *et al.* 2013). (Table1).

This is the first study on *Toxoplasma gondii* seroprevalence in donkeys from Grenada. However, we found the seroprevalence in donkeys to be lower (5.4%) than most other animal species in Grenada. In various part of the world, the vast majority of donkeys are mainly used for draught, meat and milk production. In many countries now donkey is being kept as pet. Donkey meat is delicacy in most counties. Recently the use of donkey milk for children suffering from cow milk allergy is being advocated (Machakova *et al.* 2014; Fouad *et al.* 2010).

## CONCLUSION

Since in Grenada meat and milk from donkeys are not consumed, coupled with our finding of low seroprevalence of *T.gondii* in donkeys, it is suggested that donkeys may not be important in the epidemiology of *T. gondii* in Grenada.

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## REFERENCES

- Asthana, P.S., Macpherson, C.N.L., Weiss, S.H., Stephens, R., Denny, T.N., Sharma, R.N. and Dubey, J.P. 2006. Seroprevalence of *Toxoplasma gondii* in pregnant women and cats in Grenada, West Indies. *J. Parasitol.*, **92**(3): 644-645.
- Balkaya, I., Babur, C., Celebi, B. and Utuk, A.E. 2011. Seroprevalence of Toxoplasmosis in donkeys in Eastern Turkey. *Israel J. Vet. Med.*, 66(2): 39-42.
- Cosme, A.E., Domingo, A.E. and Jitender, D. 2015. Prevalence of Toxoplasma gondii antibodies in domestic donkeys (Equus asinus) in Durango, Mexico slaughtered for human consumption. *BMC Vet. Res.*, **11**: 6.
- Chikweto, A., Kumthekar, S., Tiwari, K.P., Nyack, B., Deokar, M., Stratton, G., Macpherson, C., Sharma, R. and Dubey, J.P. 2011. Seroprevalence of *Toxoplasma gondii* in pigs, sheep, goats, and cattle from Grenada and Carriacou, West Indies. *J. Parasitol.*, **97**: 950-951.
- Choudhary, S., Zieger, U., Sharma, R.N., Chikweto, A., Tiwari, K., Ferreira, L.R., Oliveira, S., Barkley, L., Verma, S.K., Kwok, O.C.H., Sharma, R.N., Su, C. 2013. Isolation and RFLP genotyping of *Toxoplasma gondii* from mongoose (*Herpestes auropunctatus*) in Grenada, West Indies. *J. Zoo* Wildlife Med., 44(4): 1127-30.
- De Oliviera, E., de Albuquerque, P.P., de Souza, Neto O.L., Faria, E.B., Junior, J.W., Mota, R.A., Pedro, P.F., de A, Orestes L., de S.N., Eduardo, B.F., Jose, W.P., Jun and Rinaldo A.M. 2013. Occurrence of antibodies to *Toxoplasma gondii* in mules and donkeys in the Northeast of Brazil. *J. Parasitol.*, **99**(2): 343-345.
- Dubey, J.P., Ness, S.L., Kwok, O.C., Choudhary, S., Mittal, L.D. and Divers, T.J. 2014. Seropsitivity of *Toxoplasma gondii* in domestic donkeys (*Equus asinus*) and isolation of *T. gondii* from farm cats. *Vet. Parasitol.*, **199**(1-2): 18-23.
- Dubey, J.P., lappin, M., Mofya, S., Chikweto, A., Baffa, A., Doherty, D., Shakeri, J., Macpherson, C.N.L., Sharma, R.N. 2009. Seroprevalence of *Toxoplasma gondii* and concurrent *Bartonella* spp., feline immunodeficiency virus, and feline leukemia virus infections in cats from Grenada, West Indies. *J. Parasitol.*, **95**(5): 1129-1133.

- Dubey, J.P., Bhaiyat, M.I., de Allie, C., Macpherson, C.N.L., Sharma, R.N., Sreekumar, C., Viana, M.C.B., Shen, S.K., Kwok, O.C.H., Miska, K.B., Hill, D.E. and Lehmann, T. 2005. Isolation, tissue distribution and molecular characterization of *Toxoplasma gondii* from chickens in Grenada, West Indies. *J Parasitol.*, **91**(3): 557-60.
- Dubey, J.P., Bhaiyat, M.I., Macphereson, C.N.L., DeAllie, C., Chikweto, A., Kwok, O.C.H., Sharma, R.N. 2006. Prevalence of *Toxoplasma gondii* in Rats (*Rattus norvegicus*) in Grenada. *J. Parasitol.*, **92**(5): 1107-8.
- Dubey, J.P. Toxoplasmosis of animals and humans, 2nd ed. Florida: CRC Press, 2010.
- Dubey, J.P. and Desmonts, G. 1987. Serological responses of equids fed *Toxoplasma gondii* oocysts. *Equine Vet. J.*, **19**: 337–9.
- El-Ghaysh, A. 1998. Seroprevalence of *Toxoplasma gondii* in Egyptian donkeys using ELISA. *Vet. Parasitol.*, 80: 71-73.
- Flegr, J., Prandota, J., Sovic'kova, M. and Israili, Z.H. 2015. Toxoplasmosis – A Global Threat. Correlation of Latent Toxoplasmosis with Specific Disease Burden in a Set of 88 Countries. *PLoS ONE*, 9(3): e90203.
- Francesca, M., Simona, N., Roberto, P., Lind, M., Mina, M., Iolanda, A., Federica, S., Carlo, D.A. and Dubey, J.P. 2014. Detection and genotyping of *Toxoplasma gondii* DNA in the blood and milk of naturally infected donkeys (*Equus asnus*). *Parasit Vectors*, **7**: 165.
- Furtado, J.M., Smith, J.R., Belfort, R., Gattey, D. and Winthrop, K.L. 2011. Toxoplasmosis: A Global Threat. J. Glob. Infect. Dis., 3(3): 281–284.
- Fouad, H.M., Saleh, N.M.K., Khalil, H.H.M. and Morsy, T.A. 2010. Anti-*Toxoplasma gondii* antibodies in working donkeys and donkey's milk in greater Cairo. Egypt. *J. Egypt Soc. Parasitol.*, **40**(2): 459.
- Garcia- Bocanegra, I., Cabezon, O., Arenas- Montes, A., Carbonero, A., Dubey, J.P., Perea, A. and Almeria, S. 2012. Seroprevalence of *Toxoplasma gondii* in equids from Southern Spain. *Parasitol Int.*, **61**: 421-424.
- Machakova, T., Bartova, E., DILoria, A., Sedlak, K., Mariani, U., Fusco, G., Fulgione, D., Veneziano, V. and Dubey, J.P. 2014. Seroprevalene of *Toxoplasma gondii* in donkeys (*Equus acinus*) in Italy. J. Vet. Med. Sci., **76**(2): 265-267.
- Miao, Q., Xi, W., Li-Na, S., Ya-Ting, F., Fei-Zhou, Y., Jian-Fa, Y., Xing-Quan, Z. and Feng-Cai, Z. 2013. Seroprevalence of *Toxoplasma gondii* in horses and donkeys in Yunnan Province, Southeastern China. *Parasit. Vectors*, 6: 168.
- Na, Yang., Ming-Yang, m., Gao-Ming, Y., Guo-Xin, Z., Hong-Kui, L. and Jian-Bin, H. 2013. Seroprevalence of *Toxoplasma* gondii in slaughtered horses and donkeys in Liaoning Province, Northeastern China. *Parasit. Vectors*, 6: 140.

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- Robert-Gangneux, F. and Dardé, M. 2012. Epidemiology of and Diagnostic Strategies for Toxoplasmosis. *Clin. Microbiol. Rev.*, **25**(2): 265-89.
- Sharma, R.N., Gabriel, O., Tiwari, K., Chikweto, A., Bhaiyat, M.I., DeAllie, C., Paterson, T. 2014. Prevalence of *Toxoplasma gondii* antibodies in stray and owned dogs of Grenada, West Indies. *Vet. World.*, 7: 661-664.
- Sharma, R.N., Tiwari, K., Chikweto, A., DeAllie, C., Bhaiyat, M.I. 2015. Prevalence of antibodies to *Toxoplasma gondii* and *Neospora caninum* in pigs in Grenada, West Indies. *Open J. Vet. Med.*, 5: 138-141.