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Seroprevalence of Listeriosis, Toxoplasmosis and Brucellosis in Saanen X Kilis and Angora Goats in Ankara

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The objective of the present study was to determine prevalence of infection in Saanen x Kilis and Angora goats resided in Ankara with a selected group of infectious organisms. Clinical symptoms produced by Listeriosis, Toxoplasmosis and Brucellosis in goats are seen sometimes similar. Both of these zoonotic diseases possess risk for people and may cause sometimes common clinical findings. Only limited information is available in Ankara, Turkey regarding the diagnosis, presence and sero-epidemiology of these infections. Therefore we also present in this work, serological herd level analysis for detection of all aforementioned organisms.

Sera of a total of 137 goats, comprising 74 Saanen x Kilis and 63 Angora goats were analyzed for presence of antibodies against *Listeria monocytogenes, Toxoplasma gondii* and *Brucella melitensis* by use of Osebold agglutination assay, Sabin feldman dye test and Micro Agglutination Tests were used, respectively. The overall prevalences of Listeriosis, Toxoplasmosis and Brucellosis were 58.39%, 81.75% and 24.08%, respectively. Among 74 Saanen x Kilis goats the seropositivity rates of Listeriosis, Toxoplasmosis and Brucellosis were 60.81%, 81.0827% and 20.27%. Among 63 Angora goats tested the seropositivity rates of aforementioned microorganisms were 55.5%, 82.53% and 28.57%, respectively.

The results of this study suggest the presence of Listeriosis, Toxoplasmosis and Brucellosis may be prevalent in this area in goats and all of these infections should be considered by both veterinarians and public health officials.

Key Words: *Listeriosis, Toxoplasmosis, Brucellosis Saanen x Kilis, Angora goats, Seroprevalance, Ankara.*

Ankara'da Saanen X Kilis ve Angora Keçilerinde Listeriozis, Toxoplazmozis ve Brusellozis'in Seroprevalansi

Bu çalışmada Ankara'da Saanen x Kilis ve Angora keçilerinde seçilmiş bir grup bulaşıcı mikroorganizma ile infeksiyon prevalansının belirlenmesi amaçlanmıştır. Listeriozis, Brucellozis ve Toxoplasmozis infeksiyonları tarafıdan keçilerde oluşturulan semptomlar kimi zaman benzerlik göstermektedir. Tamamı zoonotik olan bu hastalıklar insan sağlığı için risk teşekkül etmekte ve ortak bir bulgu olarak keçilerde aborta neden olmaktadır. Ankara, Türkiye'de söz konusu hastalıkların tanısı, varlığı ve sero-epidemiyolojisi ile ilgili yalnızca sınırlı bilgi mevcuttur. Dolayısıyla bu araştırma ile aynı zamanda yukarıda belirtilen mikroorganizmaların sürü bazında analizleri serolojik düzeyde gerçekleştirilmiştir.

Yetmiş dört Saanen x Kilis ve 63 Angora olmak üzere toplam 137 keçiye ait serum örneği *Listeria monocytogenes, Toxoplasma gondii* ve B*rucella melitensis*'e karşı antikorların belirlenmesi amacıyla sırasıyla Osebold agglutination testi, Sabin Feldman Dye testi ve Mikro Aglütinasyon Testine tabi tutulmuştur. Listeriozis, Toxoplazmozis ve Brucellozis'in toplam prevalansları sırasıyla % 58.39, % 81.75 ve % 24.08 olarak bulunmuştur. Yetmiş dört Saanen x Kilis melezi keçide seropositiflik oranları Listeriozis, Toxoplazmozis ve Brucellozis için sırasıyla % 60.81, % 81.0827 ve % 20.27 olarak saptanmıştır. Altmış üç Angora keçisinde yukarıda sözü edilen mikroorganizmalara karşı test edilen seropositiflik oranları sırasıyla %,55.5 % 82.53 ve % 28.57 olarak belirlenmiştir.

Bu çalışmanın sonuçlarına bakıldığında Listeriozis, Toxoplazmozis ve Brucellozis infeksiyonlarının bu yöredeki keçilerde varlığı belirlenmiş ve bu infeksiyonların tamamının hem Veteriner Hekimler, halk sağlığı yetkilileri ve hayvan yetiştiricileri tarafından dikkate alınması gerektiği kanaatine varıldı.

Anahtar Kelimeler: *Listeriozis, Toksoplazmozis, Brusellozis, Saanen x Kilis, Angora keçileri, Seroprevalans, Ankara.*

Introduction

Listeriosis, caused by *Listeria monocytogenes*, has a worldwide sporadic distribution frequently in temperate climates (1-4). Listeriosis occurs commonly in ruminants, indeed may also effect fowl and human beings as well (2). The disease shows 3 distinct clinical syndromes: abortion after the infection of the pregnant uterus, septicemia in fetuses and/or neonates, and encephalitis in adult animals (3, 5). Encephalitic listeriosis has been reported more commonly in sheep than in goats, although it may effect all goat breeds.

Toxoplasma gondii, an obligate and intracellular parasite, is the aetiological agent of Toxoplasmosis. In goat and sheep abortion and associated neonatal mortality is common (6, 7).

Brucella melitensis, is the main causative agent responsible for caprine brucellosis (8). The agent is highly pathogenic for humans and appears to naturally exist in the Mediterranean area, and is widespread throughout the world. Abortion, orchitis, epididymitis, and retained plasenta may be noticed as clinical signs of the disease (8).

The purpose of this study was to demonstrate the prevalence of infection in Saanen x Kilis and Angora goats resided in Ankara with a selected group of infectious organisms. Therefore we also present in this work, serological herd level analysis for detection of Listeriosis, Toxoplasmosis and Brucellosis. Moreover we aimed to provide useful data on recognizing the presence and epidemiology of all 3 diseases together.

Materials and Methods

Clinical samples and herd history

The present study was carried out in three different locations in Ankara. In the first part of the study blood samples were collected from a total of 59 Saanen x Kilis goats at the Animal Science Department Enterprise, Faculty of Agriculture University of Ankara. The herd comprised 45 female and 14 male goats at the age of 1 year-old (n=23), 2-years-old (n=18), 3 years-old (n=13), 4 years-old (n=3) and 5 years-old (n=2). The herd had no history of previous abortions nor had signs of current illness condition, as detected by complete physical examinations. The rest of the blood samples were collected from a commercial farm with 15 Saanen x Kilis goats at the age of 1 to 4 years of age.

In the third part of the study a total of 63 blood samples were collected from an equivocal number of Angora goats resided at 5 different locations in Ankara. All samples were collected during visits provide at local commercial farms. At the time of sampling physical examination including respiratory rate and rectal temperature was performed for all goats. Age distribution of the goats was as follows: 1 year-old (n=25), 2-year-old (n=18), 3 year-old (n=9), 4 year-old (n=6), 5 year-old (n=3), 6 year-old (n=1) and 7 year old (n=1). Of the 63 goats, 11 had a history of abortion. All goats were otherwise healthy and there was no sign of any disease condition.

Blood examination

Diagnosis of Listeria monocytogenes infection

Osebold Agglutination Test (OAT)

Determination of *L. monocytogenes* “O” antibodies was carried out with the method described previously (9). The test antigen in OAT was prepared in Refik Saydam National Hygiene Center, Department of Communicable Diseases Research. All the analyses were carried out in 3 different steps. On first occasion, the whole cell antigens were prepared from *Staphylococcus aureus* (ATCC 29213) strains as described previously (9). At the next step, the antigens were prepared from *L. monocytogenes* 1/2a, 1/2b, 4b, 4c and 4d strains and were combined within the same suspension. Agglutination test was performed as a last step after absorption of sera samples with *S. aureus* antigen (9). Titers > 1/100 were considered as positive.

Diagnosis of Toxoplasma gondii infection

Sabin-Feldman Dye Test (SFDT) Protocol: Sera samples were analyzed against anti-*T.gondii* antibodies in fourfold dilutions at 1/16; 1/64; 1/256 and 1/1024 according to the modified standard Sabin-Feldman dye test (SFDT) (10).The SFDT result was suggested positive, even if more than 50% of tachyzoits were unstained at ≥1:16 dilution as detected under the light microscope (x 400).

Diagnosis of Brucella infection

Brucella Micro Agglutination Test (MAT): The test antigen was prepared in Refik Saydam National Hygiene Center, Department of Communicable Diseases Research. The MAT was performed with regard to the prior description (11). Two-fold serial dilutions of sera, ranging from 1:2.5 to 1:40, were prepared in microtiter plates within phenol saline. Fifty µl *B. abortus* S99 antigen solution stained with Safranin-O was added to each well including diluted serum and within the plate covering lid. Positive and negative controls were included for each of the running test. Phenol saline and the antigen were included as the negative control covering wells. The results were evaluated following 18 h of incubation at 37°C. Positive (large diffuse red mat) or negative (compact red dot) results were evaluated within the agglutination results.

Results

Osebold test for Listeria infection

Of the 137 goat sera tested against *L. monocytogenes* “O” antibodies; 80 (58.39%) were seropositive, regarding the overall prevalence. Among 74 Saanen x Kilis goats 45 (60.81%) were seropositive . The titers of 45 seropositive goats were as follows; 1/100 for 32 and 1/200 for 13 goats, respectively (Table 1). Among 63 Angora goats tested 35 were seropositive (55.5%) with titers as follows; 1/100 for 17, 1/200 for 14 and 1/400 for 4 (Table 1).

Table 1. Anti-*L.monocytogenes* “O” antibody titers in Saanen x Kilis and Angora goats.

	1/100	1/200	1/400	Total
Goats				
Saanen x Kilis	32	13	-	45
Angora	17	14	4	35

SFDT test for Toxoplasma gondii

By use of SFD test, the overall prevalence of Toxoplasmosis was 81.75% (112/137). Among 74 Saanen x Kilis goats 60 (81.0827%) were seropositive. The titers of 60 seropositive goats were as follows; 1/16 for 23, 1/64 for 30, 1/256 for 4, 1/1024 for 3 (Table 2). Among 63 Angora goats tested 52 were seropositive (82.53%) with titers as follows; 1/16 for 19, 1/64 for 23, 1/256 for 4 and 1/1024 (Table 2).

Table 2. Anti-*T.gondii* antibodies in Saanen x Kilis and Angora goats.

	1/16	1/64	1/256	1/1024	Total
Goats					
Saanen x Kilis	23	30	4	3	60
Angora	19	23	6	4	52

MAT test for Brucella infection

By use of MAT test the overall prevalence of Brucellosis was 24.08% (33/137). Among 74 Saanen x Kilis goats 15 (20.27%) were seropositive. The titers of 15 seropositive goats were as follows; 1/20 for 3, 1/40 for 2, 1/80 for 6, 1/160 for 3 and 1/640 for 1 (Table 3). Among 63 Angora goats tested 18 were seropositive (28.57%) with titers as follows; 1/20 for 1, 1/40 for 4, 1/80 for 8, 1/160 for 2 and 1/640 for 3 (Table 3).

Table 3. Brucella antibody titers in Saanen x Kilis and Angora goats.

	1/20	1/40	1/80	1/160	1/640	Total
Goats						
Saanen x Kilis	3	2	6	3	1	15
Angora	1	4	8	2	3	18

Discussion

Presence and epidemiology of Listeria monocytogenes infection in Saanen x Kilis and Angora goats

To the present authors' knowledge there has been limited epidemiological data available regarding Listeriosis infection in goats in Ankara and in Turkey. Therefore the real prevalence remains unclear. In Aydın provience by use of Osebold method *L. monocytogenes* ''O'' antibodies were detected 35 (35%) out of 100 sheeps with titers of 1/100 (27%), 1/200 (8%) (12). In a previous study sera of 50 Angora goats, analyzed for the presence of antibodies against Listeria monocytogenes by the Osebold method on a commercial farm in Ankara revelaed 23 (46%) seropositive samples (13). In the latter study titrations of seropositive samples were found 1/100 (20%), 1/200 (24%) and 1/400 (2%) (13). In the present study the overall prevalence of the 137 goat sera tested was 58.39% for Listeriosis. Among 74 Saanen x Kilis goats 45 (60.81%) were seropositive with titers of 1/100 (43.24%) and 1/200 (17.56%). Among 63 Angora goats tested 35 were seropositive (55.5%) with titers as follows; 1/100 (26.98%), 1/200 (22.2%) and 1/400 (0.6%).

In conclusion higher rates of seropositivity against *L. monocytogenes* in goats resided in Ankara may suggest that *L. monocytogenes* infection is prevalent and should be taken into consideration by veterinarians and public health officers.

Presence and epidemiology of Brucellosis in Saanen x Kilis and Angora goats

In Kayseri region 12 (7.79%) out of 154 sheep showed seropositivity against Brucellosis (14). Another study undertaken in Aydin province revealed 2% seropositivity for Brucella antibodies in sheeps by use of agglutination tests (12). By use of tube agglutination test in Kars city 3 (2.91%) out of 103 sheep were found seropositive against Brucellosis (15). In the present study by use of MAT the overall prevalence of Brucellosis was 24.08% (33/137). Among 15 (20.27%) seropositive Saanen x Kilis goats, titers were 1/20 for 3, 1/40 for 2, 1/80 for 6, 1/160 for 3 and 1/640 for 1 sera samples. Among 18 (28.57%) seropositive Angora goats tested titers were; 1/20 for 1, 1/40 for 4, 1/80 for 8, 1/160 for 2 and 1/640 for 3 sera samples. In contrast to the previously reported seroprevalence studies in Turkey among different regions in sheeps, the overall prevalence detected in goats was markedly higher. Given the seroprevalence rates in the present study and the lack of detailed epidemiological studies in goats in Turkey, this fact indicates the necessity of further studies with greater goat populations to better understanding of the aetiology of this disease, which will become the prior subject of our future studies.

Presence and epidemiology of Toxoplasma gondii infection in Saanen x Kilis and Angora goats

To the present authors' knowledge there has been limited data available regarding the seroepidemiology of Toxoplasmosis in goats in Ankara and in Turkey. In Angora goats resided in Eskisehir, anti-*Toxoplama gondii* antibodies were detected in 43 (43.87%) out of 98 sera samples (16). In a previous study performed on 68 Angora goats in Ankara vicinities, 37 goats (54%) were found to be positive for *T. gondii* antbodies by use of Sabin Feldman Dye test (17). In another seroprevalence study in Kayseri of the 154 sheep tested by use of Sabin Feldman Dye test, 52 (33.76%) were found to be seropositive against Toxoplasmosis (14). Similarly in another seroprevalance study in Kars city, Turkey, 53 (51.45%) out of 103 sheep were found to be positive against *T. gondii* antbodies (15). In Nigde, a central Anatolian city in Turkey, by use of Sabin Feldman Dye test, 56 out of 110 sheep and 19 out of 46 goats (41%) had seropositivity against Toxoplasmosis (18). Another prevalence study performed in Cankiri revealed 24 (63.15%) seropositive samples out of 38 goats (19). In the present study a seropositivity rate of Toxoplasmosis was 81.75% (112/137). Among 74 Saanen x Kilis goats 60 (81.0827%) were seropositive with of 1/16 for 23, 1/64

for 30, 1/256 for 4, 1/1024 for 3 sera samples. Among 63 Angora goats tested 52 were seropositive (82.53%) with titers as follows; 1/16 for 19, 1/64 for 23, 1/256 for 4 and 1/1024 sera samples. In contrast to the previous reports, the overall prevalence determined in the present study was markedly higher. Although, as aforementioned above, both the previous and the present study had the same methodology, the variable seroprevalence rates may be attributable to the geographical differences.

Conclusion

The results of the present study further confirm the presence of Listeriosis, Brucellosis and Toxoplasmosis infections in goat populations in Ankara, Turkey. The difference in seroprevalence rates in goats between the present work and the previous studies from Turkey may be attributable to difference in localities where samples have been taken. However in the present study and in most of the other studies reported similar methodologies, i.e. Sabin Feldman Dye Test for Toxoplasmosis, Agglutination tests for Brucellosis and Osebold method for Listeriosis. In addition, this variation may also be related to the age, breed, sex and sampling methods of the animals and husbandary practices. In the present study none of the animals included had prior abortion history, nor had relevant clinical signs, although indicating the subclinical presence of those agents and exposure of the animals. High seropositivity rates should not always be evaluated by means of real infection, suggesting that the animals may be exposed to the disease previously and recovered.

References

1. Erdogan HM, Cetinkaya B, Green LE, Cripps PJ, Morgan KL.: Prevalence, incidence, signs and treatment of clinical listeriosis in dairy cattle in England. Vet Rec 2001; 149: 289-293.
2. George LW. Diseases presenting principally with brain stem and cranial nerve dysfunction. Listeriosis. In: Smith BP. (Ed) Large Animal Internal Medicine. 2nd Edition, St Louis, Missouri: Mosby-Year Book, INC, Carlisle Communications Ltd., 1996; 1001-1091.
3. Johnson GC., Maddox CW., Fales WH., Wolff W.A., Randle R.F., Ramos JA., Schwartz H., Heise KM., Baetz AL., Wesley IV., Wagner, DE. Epidemiologic evaluation of encephalitic listeriosis in goats. J Am Vet Med Assoc 1996; 208: 1695-1699.
4. Miettinen A., Husu J., Tuomi J. Serum antibody response to *Listeria monocytogenes*, Listerial excretion, and clinical characteristics in experimentally infected goats. J Clin Microb 1990; 28: 340-343.
5. Seimiya Y, Ohshima KI., Itoh H., Murakami RK. Listeric septicemia with meningitis in a neonatal calf. J Vet Med Sci 1992; 54: 1205-1207.
6. Eckert Y, Kutzer E., Rommel M., Bürger HJ., Körting W. Veterinarmedizinische Parasitologie. Begründet von Josef Boch und Rudolf Supperer. Berlin: Verlag Paul Parey, 1992.
7. Dubey JP., Beattie CP. Toxoplasmosis of animal and man. Florida: CRC press. inc. Boca Raton, 1988.
8. Arda M., Minbay A., Leloglu N., Aydın N., Istanbulluoglu E., Akay O., Ilgaz A., Izgur M., Diker S. *Ozel Mikrobiyoloji* 4. Baskı, Ankara: Medisan Yayın Serisi, Medisan Yayınevi, 1997.
9. Osebold J., Aalund O., Chrisp C.E. Chemical and immunological composition of surface structures of *Listeria monocytogenes*. J Bacteriol 1965; 89: 84-86.
10. Feldman HA., Lamb GA. A micromodification of the Toxoplasma Dye Test. J Parasitol 1966; 52: 415
11. Baum M., Zamir O., Bergman-Rios R., Katz E., Beider Z., Cohen A., Banal M. Comparative Evaluation of Microagglutination Test and Serum Agglutination Test as Supplementary Diagnostic Methods for Brucellosis. J Clin Microbiol 1995; 33: 2166-2170
12. Pasa S., Kılıc S., Babur C., Ocal N., Gazyagci, S. Aydın yoresinde koyunlarda Listeriosis ve Brucellosis'in seroprevalansı. Etlik Vet Mikrobiyol Derg 2003; 14(1-2): 11-17.
13. Borku MK., Ural K., Gazyagci S., Ozkanlar YE., Babur C., Kılıc S. Serological Detection of Listeriosis at a Farm. Turk J Vet Anim Sci 2006; 30: 279-282.
14. Inci A., Aydin N., Babur C., Cam Y., Akdogan C., Kuzan S. Kayseri yoresinde sıgır ve koyunlarda Toksoplazmoz ve Brusellozis uzerine seroepidemiyolojik araştırmalar. Pendik Vet Mikrobiyol Derg 1999; 30(1):41-46.
15. Aslantas O., Babur C. Kars Yoresinde sıgır ve koyunlarda Bruselloz ve Toksoplazmoz uzerine seroepidemiyolojik araştırmalar. Etlik Vet Mikrobiyol Derg 2000;11(1-2): 47-55.
16. Babur C., Piskin FC, Biyikoglu G., Dundar B., Yarali C. Eskisehir Cifteler harasi Ankara Kecilerinde Anti-*Toxoplasma gondii* antikorlarının Sabin-Feldman Dye Test (SFDT) ile araştirilmasi. Turkiye Parazitol Derg 1999; 23(1):72-74.
17. Yagci S., Babur C., Karaer Z., Cakmak A. Ankara yoresinde kecilerde Toxoplasmosis. Etlik Vet Mikrobiyol Derg 1997; 1(9): 94-98.
18. Karatepe B, Babur C, Karatepe M, Cakmak A, Nalbantoglu S. Seroprevalance of toxoplasmosis in sheep and goats in the Nigde province of Turkey Indian Vet J 2004; 81: 974-976.
19. Babur C, Inci A., Karaer Z. Cankiri yoresinde koyun ve kecilerde *Toxoplasma gondii* seropozitifliğinin Sabin-Feldman boya testi ile saptanmasi. Türkiye Parazitol Derg 1997; 21(4): 409-412.