

Prevalence and spatial distribution of brucellosis in goats in the southernmost provinces of Thailand in 2014

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Abstract

Brucellosis in goats is an important zoonotic disease in Thailand, where a national brucellosis control and eradication program has been in place since 2003. This study aimed to determine prevalence of brucellosis in goats in the five southernmost provinces that form the country's major goat raising area. A descriptive study was conducted in 2014 using laboratory data from the Veterinary Research and Development Center (Lower-southern Region). The study included goats in the five provinces from herds with a minimum size of five adult goats aged ≥ 6 months, an approximate average herd size for the area. All goats were sampled for brucellosis testing, resulting in 15,281 goat serum samples from 845 herds. These herds were located in 42 out of 55 districts. Two hundred and thirty-five samples (1.54%) tested positive after screening with the modified Rose Bengal Test. Only 67 (0.44%) of these tested positive using the Complement Fixation Test and were identified as having brucellosis. Brucellosis was found in goats in every province. These goats were from 15 herds in 10 districts. Overall, herd prevalence and overall individual prevalence of brucellosis in the goats in this study were 1.78% (95%CI: 1.03%-2.98%) and 0.44% (95%CI: 0.34%-0.56%), respectively. The mean within-herd prevalence at overall-herd and positive-herd levels were 0.18% and 10.00%, respectively. The prevalence of brucellosis in goats in the southernmost provinces of Thailand was generally low. The control program should be continued and strengthened to further reduce the prevalence of brucellosis in goat herds in this area.

Key words: Brucellosis, Prevalence, Goat, Modified Rose Bengal test, Complement fixation test

Introduction

Brucellosis is an important bacterial zoonotic disease that mainly affects cattle, sheep and goats. It is caused by various species of *Brucella* spp. Among them, *B. melitensis* is the species that most commonly causes brucellosis in goats and sheep, and that has the highest potential for zoonotic infection. Brucellosis has a worldwide distribution. Serological tests are commonly used for the diagnosis of brucellosis because they have good sensitivity are cheap, and are practical to apply on a large scale. The main serological tests used for diagnosis of brucellosis in goats and sheep are the modified Rose Bengal test (mRBT) as a screening test and the complement fixation test (CFT) or enzyme linked immunosorbent assay (ELISA) as the confirmatory test (Ferreira et al., 2003).

Brucellosis is endemic in Thailand, where it particularly affects cattle and goats. However, goats have been almost exclusively the source of human infections (BoE, 2007-2013). The country implemented a national brucellosis control and eradication program (NBCEP) in 2003 to minimize the economic impact on livestock and to prevent a public health risk. The program includes animal identification and farm registration, serological surveillance, culling and compensation, vaccination of cattle and buffaloes with the *Brucella abortus* strain 19 vaccine, movement control, certified brucellosis-free farms, and improved laboratory diagnosis (Ekgat et al., 2011). Currently, every veterinary diagnostic laboratory in the Department of Livestock Development (DLD) in Thailand is working under the same standards. The laboratories conduct serial testing by first screening with mRBT and then confirming using CFT for diagnosis of brucellosis in goats and sheep. In 2010, the national brucellosis surveillance showed a nationwide estimated prevalence of brucellosis at the herd and animal levels in sheep and goats of 6.9% and 1.5%, respectively. The herd and animal level estimates of 1.58% and 0.76% for the lower southern region and 4.02% and 0.52% for the upper southern region were lower than in any other regions (Ekgat et al., 2011).

The five southernmost provinces are located in the lower southern region of Thailand. This represents the country's major goat raising area, where goat consumption is high (DLD^A, 2014). In 2004, a goat-associated human brucellosis outbreak occurred in this region (BoE, 2004). In addition, these provinces share a border with a neighboring country, where there were reports of significantly high prevalence of brucellosis in goats in their border areas (Baimiya et al., 2015). Accordingly, the brucellosis status of goats in the five southernmost

provinces should be periodically updated. Thus, this study aimed to determine the prevalence and describe spatial distribution of brucellosis in goats in this particular area of Thailand in 2014.

Materials and methods

Study area

The five southernmost provinces comprise the Satun, Songkhla, Pattani, Yala and Narathiwat Provinces. The estimated goat population in these areas in 2014 was 173,933 goats in 28,574 herds, accounting for 37.14% of country's goats (DLD^A, 2014). The government veterinary diagnostic laboratory that has served this region since 2013 is the Veterinary Research and Development Center (Lower-southern region), located in Songkhla Province.

Study design and samples

This descriptive study was conducted using the laboratory database of the Veterinary Research and Development Center (Lower-southern Region). The data comprised demographic and laboratory test results from individual goats. The study population was goats in the five southernmost provinces from herds with a minimum size of five adult goats aged ≥ 6 month-old, an average herd size for the area. All goats were sampled for brucellosis testing.

Serological tests and definition of brucellosis goat/herd

The antigen used for the mRBT was produced by the Bureau of Veterinary Biologics, Department of Livestock Development, Thailand. CFT antigen was prepared from *B. abortus* strain 99 (Antifix, Synbiotics Corp. Lyon, France). The mRBT was performed following the standard procedure (OIE, 2012). Briefly, 75 μ L of sera and 25 μ L of the antigen were mixed. The plates were shaken for four minutes and any agglutination that appeared within this time was recorded as a positive reaction. The CFT was performed on microplate by the standard procedure (OIE, 2012). Briefly, 25 μ L of inactivated test serum was diluted for serial doubling dilutions. Then, 25 μ L of antigen were added to all wells except the serum control well and 25 μ L of the complement was added to each well. After the plates were incubated at 4°C overnight, 25 μ L of sensitized sheep red blood cells (SRBCs) was added to all wells and the plates were re-incubated at 37 °C for 30 minutes. The results were read after the plates were left to stand at 4°C for 2-3 hours to allow unlysed SRBCs to settle. A positive result was

revealed by the absence of hemolysis. Sera giving a titer equivalent to ≥ 20 international complement fixation test units per ml (ICFTU/ml) were considered positive. Only the sera with a positive reaction in the mRBT were tested with the CFT for confirmation of brucellosis. Samples that were positive in both tests were defined as brucellosis-positive. Any herd with at least one goat positive for brucellosis was defined as a brucellosis-positive herd.

Statistics

Descriptive statistics, frequency, average, and geographic mapping were used to describe brucellosis prevalence, mean within-herd prevalence and visualized spatial distribution. Percentages of brucellosis-positive individuals and herd levels were estimated at the 95% confidence interval. The average percent positive within each herd represented the mean within-herd prevalence (Epi InfoTM7). Spatial distribution of brucellosis occurrence by district was depicted on a map (ArcGIS).

Results

A total of 15,281 goat serum samples from 845 herds were included in this study. These were broken down into 2,635 serum samples from 45 herds in Satun, 1,918 serum samples from 65 herds in Songkhla, 1,250 serum samples from 110 herds in Pattani, 4,874 serum samples from 273 herds in Yala and 4,604 serum samples from 352 herds in Narathiwat. These accounted for 8.79% (15,281/173,933) of the goat population. Two-hundred-and-thirty-five serum samples were positive after screening with mRBT and 67 (28.51%) of those were confirmed positive with CFT.

Herd and individual prevalence

Overall prevalence of brucellosis in goats with 95% confidence at herd and individual levels was 1.78% (1.03%-2.98%) and 0.44% (0.34%-0.56%), respectively (Table 1). The highest prevalence was found in the Songkhla Province.

Mean within-herd prevalence

Brucellosis prevalence within each herd was described by mean within-herd prevalence (Table 2). The overall mean within herd prevalence at the overall-herd and positive-herd levels were 0.18% and 10.00%, respectively. The mean within-herd prevalence by province

amongst positive herds ranged from 1.54 to 13.81. Songkhla Province had the highest mean within-herd prevalence.

Spatial distribution of brucellosis in goats

Samples were collected from 42 out of the 55 districts in the area. Brucellosis in goats was found in 10 districts (Figure 1). Brucellosis presented in one district each in the Satun and Songkhla Provinces. The disease presented in two districts in Pattani and three districts in the Yala and Narathiwat Provinces. The districts where brucellosis was identified in the latter three provinces were adjacent.

Discussion

Approximately 9% (15,281/173,933) of goats in the southernmost provinces of Thailand were examined for brucellosis in 2014. The prevalence was generally low and lower than previous surveys. The overall prevalence rates of 0.44% (95%CI 0.34%-0.56%) and 1.78% (95%CI 1.03%-2.98%) for the individual and herd levels, respectively, were well below those from the previous surveys of Thailand's southern region done in 2009 and 2010, which showed levels of 0.62% and 1.40% for individual prevalence and 3.33% and 8.46% for herd prevalence (Chumek and Jeenpun, 2012). A recent survey of brucellosis in goats in western Thailand, the country's second most prominent goat production area, found significantly high individual and herd prevalence rates of 5.08% and 18.39%, respectively (Antarasena et al., 2013). Moreover, a neighboring country that shares border with Thailand at its southernmost provinces also reported higher prevalence figures. There, the overall individual and herd prevalence rates for brucellosis in goats, based on sero-surveillance data from 2000-2009, were 0.91% (95%CI 0.86%-0.96%) and 7.09% (95%CI 6.27%-7.98%), respectively (Baimiya et al., 2015). The decline in brucellosis prevalence in the southernmost provinces of Thailand could be the outcome of the NBCEP, which has been in place for over a decade. Certified brucellosis-free goat farms in the area have grown in number (and this study could have a selection bias if samples were mainly from brucellosis-free farm) (DLD^B, 2014). Standardization of laboratory diagnostic testing has also helped reduce false positive diagnosis. In serial testing, any positive results from mRBT screening must be confirmed by CFT. In this study, this practice reduced the number of false positives from mRBT by about three fourths.

Despite a decline in brucellosis prevalence in goats in the southernmost provinces, the spatial distribution has not changed. This present study showed similar results to a 2009-2010 sero-survey of brucellosis in goats in southern Thailand, which indicated that the Songkhla Province was the area with the highest brucellosis prevalence (Chumek and Jeenpun, 2012). That province also had the highest mean within-herd prevalence, which suggested a delay in disease detection and control. Furthermore, the present study showed that brucellosis was found in every province and mostly occurred in adjacent districts. Due to a lack of data, this study was unable to describe whether this occurrence happened by chance. Factors associated with brucellosis clustering in space were shown by a spatial analysis of ruminant brucellosis in the Kafr El Sheikh Governorate of the Nile Delta, Egypt in 2008 (Hegazy et al., 2011). The study revealed brucellosis clusters near animal markets and areas with high livestock density. These results were in accordance with studies in several settings showing that animal movement and large herds are associated with brucellosis (Coelho et al., 2007; Solorio-Rivera et al., 2007). In addition, a 2006 study of risk factors associated with brucellosis at the herd level in goats in Chinat Province, Thailand identified farms with a history of abortion during the previous year as risk factor (Suddee et al., 2011). Information from that study, coupled with previous research on risk factors of brucellosis in goats, suggested that brucellosis prevention and control activities needed to be strengthened in areas and herds that were at risk in the southernmost provinces of Thailand. Particular measures included quick disease detection and culling, and promotion and expansion of the number of voluntary certified brucellosis-free farms.

Conclusion

The prevalence of brucellosis in goats in the five southernmost provinces of Thailand was generally low and has declined from earlier periods. The NBCEP should be continued and strengthened in high prevalence areas to further reduce the prevalence of brucellosis or free brucellosis from goat herds in this area.

Acknowledgements

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Table 1 Prevalence of brucellosis in goats in the five southernmost provinces of Thailand at individual and herd levels, 2014

Provinces	Individual				Herd			
	Total	Positive			Total	Positive		
	No	No	%	95%CI	No	No	%	95%CI
Satun	2,635	1	0.04	0.00-0.25	45	1	2.22	0.06-11.77
Songkhla	1,918	39	2.03	1.47-2.80	65	2	3.08	0.37-10.68
Pattani	1,250	2	0.16	0.03-0.64	110	2	1.82	0.22-6.41
Yala	4,874	13	0.27	0.15-0.47	273	5	1.83	0.60-4.22
Narathiwat	4,604	12	0.26	0.14-0.47	352	5	1.42	0.52-3.48
Total	15,281	67	0.44	0.34-0.56	845	15	1.78	1.03-2.98

Table 2 Mean within-herd prevalence of brucellosis in goats in the five southernmost provinces of Thailand at overall-herd and positive-herd levels, 2014

Provinces	Overall-herd			Positive-herd		
	Total	Prevalence		Total	Prevalence	
	No	Mean	SD	No	Mean	SD
Satun	45	0.03	0.23	1	1.54	NA
Songkhla	65	0.42	3.01	2	13.81	14.48
Pattani	110	0.11	0.84	2	6.27	0.55
Yala	273	0.22	1.74	5	11.96	5.63
Narathiwat	352	0.14	1.34	5	9.70	6.40
Total	845	0.18	1.59	15	10.00	6.81

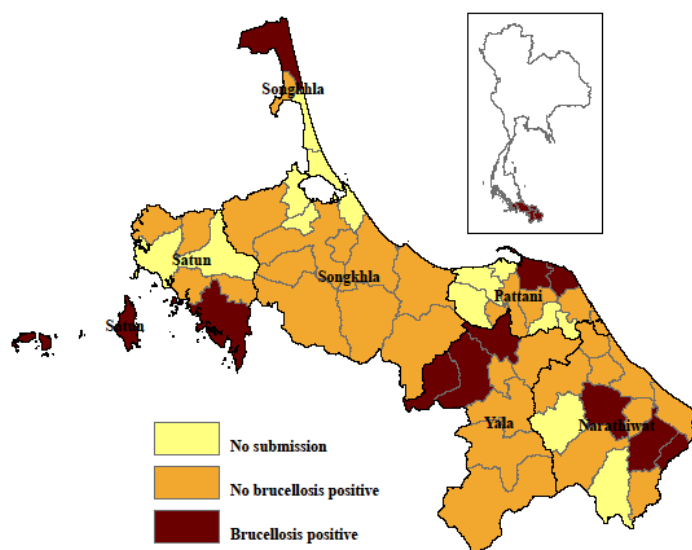


Figure 1 District map showing the distribution of brucellosis in goats in 10 districts of the five southernmost provinces of Thailand, 2014

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ความชุกและการกระจายเชิงพื้นที่ของโรค布鲁เซลโลสิสในแพะ ในพื้นที่จังหวัดชายแดนภาคใต้ของประเทศไทยใน พ.ศ. 2557

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บทคัดย่อ

โรค布鲁เซลโลสิสในแพะเป็นโรคติดต่อระหว่างสัตว์และคนที่มีความสำคัญในประเทศไทย จึงมีโครงการควบคุมและกำจัดโรคในระดับประเทศขึ้นตั้งแต่ พ.ศ.2546 การศึกษารั้วนี้มีจุดประสงค์เพื่อแสดงความชุกของโรค布鲁เซลโลสิสในแพะ ใน พ.ศ.2557 ในพื้นที่ 5 จังหวัดชายแดนภาคใต้ของประเทศไทยซึ่งเป็นแหล่งเลี้ยงแพะที่สำคัญของประเทศ โดยศึกษาข้อมูลรายตัวแพะและผลการทดสอบโรค布鲁เซลโลสิสจากห้องปฏิบัติการศูนย์วิจัยและพัฒนาการสัตวแพทย์ภาคใต้ตอนล่าง เลือกเฉพาะฝูงที่มีแพะอายุไม่น้อยกว่า 6 เดือน 5 ตัวขึ้นไป ซึ่งเป็นขนาดฝูงแพะเฉลี่ยในพื้นที่ และแพะทุกตัวได้ทดสอบโรค布鲁เซลโลสิส จำนวน 15,281 ตัว จาก 845 ฝูง ของพื้นที่ 42 อำเภอ ใน 55 อำเภอ พบให้ผลบวก 1.54% (235/15,281) โดยการตรวจคัดกรองด้วยวิธี modified Rose Bengal Test และมีการตรวจยืนยันด้วยวิธี Complement Fixation Test วินิจฉัยว่าเป็นโรค布鲁เซลโลสิส 0.44% (67/15,281) พบแพะที่เป็นโรคในทุกจังหวัดกระจายอยู่ใน 15 ฝูง ของ 10 อำเภอ ซึ่งส่วนใหญ่เป็นอำเภอที่มีพื้นที่ติดกัน ค่าความชุกในภาพรวมในระดับฝูงและรายตัว มีค่า 1.78% (95%CI: 1.03%-2.98%) และ 0.44% (95%CI: 0.34%-0.56%) ตามลำดับ ส่วนค่าเฉลี่ยในภาพรวมของความชุกของโรค布鲁เซลโลสิสภายในฝูงจากจำนวนฝูงทั้งหมดและจากจำนวนฝูงที่ให้ผลบวกเท่านั้น มีค่าเท่ากับ 0.18% และ 10% ตามลำดับ กล่าวได้ว่าความชุกของโรค布鲁เซลโลสิสในแพะในพื้นที่ 5 จังหวัดชายแดนภาคใต้ใน พ.ศ.2557 อยู่ในระดับต่ำ ควรดำเนินการมาตรการควบคุมโรคที่มีอยู่ต่อไปอย่างต่อเนื่อง และมุ่งเน้นให้มีความสำคัญกับการดำเนินการในพื้นที่เสี่ยงเพื่อลดความชุกของโรค布鲁เซลโลสิสในฝูงแพะในพื้นที่นี้ต่อไป

คำสำคัญ: บรูเซลโลสิส ความชุก แพะ modified Rose Bengal Test, Complement Fixation Test