Seroprevalence of Theileria Infection in Goats in Hubei Province, China based on Circulating Antibodies

Houqiang Luo\textsuperscript{1,3}, Kun Li\textsuperscript{3,4}, Hui Zhang\textsuperscript{5}, Yanfang Lan\textsuperscript{2}, Ping Gan\textsuperscript{1}, Xiong Xiong\textsuperscript{2,4}, Haigang Wu\textsuperscript{3} and Jiaxiang Wang\textsuperscript{6*}

\textsuperscript{1}Animal Science Department, Wenzhou Vocational College of Science and Technology, Wenzhou 325006, People’s Republic of China; \textsuperscript{2}College of Veterinary Medicine, Huazhong Agricultural University, Wuhan 430070, People’s Republic of China; \textsuperscript{3}Jiangxi Animal Disease Prevention and Control Center, Nanchang, Jiangxi 330096, People’s Republic of China; \textsuperscript{4}Animal husbandry technology extending stations, Yichang 443000, People’s Republic of China; \textsuperscript{5}College of Animal Science and Veterinary Medicine, Xinyang Agriculture and Forestry University, Xinyang, Henan, 464000, People’s Republic of China; \textsuperscript{6}College of Animal Science, Yangtze University, Jingzhou 434023, People’s Republic of China

\textsuperscript{*}Corresponding author: chviolet@tom.com (HQL); wangjiaxiang1109@163.com (JXW)

\textbf{ARTICLE HISTORY (16-142)}

Received: June 18, 2016
Revised: October 13, 2016
Accepted: October 14, 2016
Published online: November 17, 2016

\textbf{Key words:}
ELISA
Goat
Prevalence
Theileria

\begin{abstract}
The word-wildly spread Theileria, is an economical important parasite to sheep and goats production. However, a little is known about whether goats in Hubei province are exposed to Theileriosis. Therefore, a survey was conducted to investigate the prevalence of Theileria infection in goats in Hubei province, China. A total 912 and 1047 serum samples were collected in 2014 and 2015, respectively and were assayed for Theileria antibodies by ELISA. The results showed that the prevalence was 17.0 and 21.8\% in 2014 and 2015, respectively, which had revealed a growing trend of Theileriosis infection in goats in Hubei province.
\end{abstract}

\begin{introduction}
Tick-borne hemoproteozan Theileria parasites are widely distributed in tropical and subtropical climates (Aydin et al., 2013). Theileria species are a cause of clinical and subclinical infections in many wild and domestic animals including sheep and goats (Aydin et al., 2013; Li et al., 2014). Theileria infection is important in animal production for its high mortality and morbidity leading to heavy economic losses in epidemic regions (Aydin et al., 2013). Tian et al. (2013) reported Theileriosis as a severe and often lethal disease, contributing a severe restriction for the development of livestock industry of small ruminants in northwest China.

Hubei province located in the central of China, is a big agricultural province in this country. The suitable climate and fertile land accomplish the abundant agricultural resource including a large number of 4.70 million (available at http://data.stats.gov.cn/easyquery.htm?cn=E0103; National Bureau of Statistics of China) goats in this province. Goat fur, milk, meat are important products to herdsman, so any goat disease may lead to potential threat to goat production. However, until now scarce information is available about the prevalence of Theileria infection in goats in Hubei. The object of current research was to execute epidemiological investigation of Theileria infection in goats in this area.

\end{introduction}

\begin{methods}
Samples Collection: Blood samples were collected from the jugular vein by local veterinary practitioners from 912 goats in 2014 and 1047 goats in 2015 in Hubei province (Table 1). After collection, all the samples were centrifuged at 10000xg for 10min, and serum was separated and stored at -20\degree C till further analysis.

Determination of antibodies against Theileria: Antibodies against Theileria were determined by a commercial enzyme linked immunosorbent assay (ELISA) kit (goat theileriosis-Ab Test Kit, Shanghai Yu Ping biotechnology limited company) according to the manufacturer's instructions. The CUT OFF value was calculated based on the optical density (OD) values according to the formula: CUT OFF= the average OD 450 of negative controls + 0.15. To ensure validity, the average OD 450 of negative controls was ≤0.10; the average OD 450 value of positive was ≥1.00. The results
\end{methods}
were interpreted as negative when the OD 450 value of sample <CUT OFF and positive when the OD 450 value of sample ≥CUT OFF.

**Statistical analysis:** Statistical analysis of *Theileria* prevalence was performed by chi-square test with SPSS (Statistical Analysis System, Version 17.0). The differences were considered statistically significant at 5% level of significance (P<0.05).

**RESULTS**

In 2014, a total of 155 out of 912 serum samples were tested positive for antibodies against *Theileria*. The regional seroprevalence of *Theileria* was 100% (6/6). The prevalence of *Theileria* infection in each region of Hubei province ranged from 8.5% to 30.1%, and differences among the regions were found statistically significant (P<0.01). On genders basis, the prevalence was 17.2% and 16.8% in male and female goats, respectively. In different seasons, the prevalence was ranged from 5.1% to 29.2% with a significant difference in the different seasons (P<0.01) (Table 1).

In 2015, a total of 228 out of 1057 serum samples were detected positive for antibodies against *Theileria*. The regional seroprevalence of *Theileria* was 100% (6/6). In all of the 6 counties, the prevalence was ranged from 9.4% to 39.4% and there was significant difference in the different counties (P<0.01). On genders basis, the seroprevalence was 20.6% and 23.2% in male and female goats, respectively. In different seasons, the prevalence of *Theileria* infection was ranged from 14.4% to 30.9% and the difference in the different seasons were found significant (P<0.01) (Table 1).

**DISCUSSION**

As a word-wide spread parasite, *Theileria* has been reported in many countries and areas (Aydin et al., 2013). Previously, Alyasino and Greiner (1999) reported a prevalence of 59.9% (547/913) of *Theileriosis* in Awassi sheep in Syria by IFAT (indirect immunofluorescence antibody test). Altay et al. (2005) reported that 54.03% (67/124) of sheep were detected positive for *Theileria ovis* antibodies by employing nested PCR method. Previously, *Theileriosis* was reported in Sichuan, Qinghai, Gansu, Liaoning, Inner Mongolia, Shaanxi, and Ningxia in China with a prevalence of 36%-100%; mortality as high as 17.8% to 75.4% (Li et al., 2012).

According to Li et al. (2014), *Theileria* infection in goats was 80% (40/52) in Dawu, 50% (28/56) in Suizhou and 50% (24/48) in Suixian, in China by using method of microscopic examination of thin blood smears. *Theileria* infection in goats in Hubei province, are significantly lower than that in previous studies (Alyasino and Greiner, 1999; Altay et al., 2005; Li et al., 2014). The possibly reason may be the differences in geographical conditions, climatic changes, environment and detection methods (Li et al., 2015). Though, a relatively low prevalence was presented herein, the results revealed a growing trend of *Theileria* infection in goats (Table 1).

In different counties, there was significant difference (P<0.01) of *Theileria* infection in goats in 2014 and 2015, respectively (Table1), as this protozoan disease is an epidemic disease (Li et al., 2012). In different genders, the difference of the prevalence of *Theileria* between male and female goats were not statistically significant (P>0.05) (Table 1), which may demonstrate that male and female goats are equally prone to *Theileria* infection. In different seasons, the prevalence of *Theileria* were significantly higher in spring and summer than that in autumn and winter (P<0.01) (Table 1), the reason may because of that the activity of ticks is highly relevant to temperature (Guo et al., 2002) and *Theileria* are mainly transmitted by tick vectors and a number of them are highly pathogenic for goats (Tian et al., 2013; Li et al., 2014). The infected animals may transmit this protozoa by ticks to other animals, contributing to the high prevalence in spring and summer. Once infected with *Theileriosis*, sheep and goats depicted pyrexia (40-42°C), pale mucous membranes, enlarged superficial lymph nodes, respiratory distress (Tageldin et al., 2005). The high prevalence in current results may lead to enormous economic losses due to high mortality and morbidity (Tian et al., 2013).

**Table 1**: Seroprevalence of *Theileria* infection in goats in different counties, genders and seasons in Hubei province

<table>
<thead>
<tr>
<th>County</th>
<th>Positive serum / Total Samples</th>
<th>Seroprevalence (%)</th>
<th>Positive serum / Total Samples</th>
<th>Seroprevalence (%)</th>
<th>Growth rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>27/100</td>
<td>27.0</td>
<td>61/181</td>
<td>33.7</td>
<td>24.81</td>
</tr>
<tr>
<td>B</td>
<td>17/201</td>
<td>8.5</td>
<td>22/233</td>
<td>9.4</td>
<td>10.59</td>
</tr>
<tr>
<td>C</td>
<td>35/177</td>
<td>19.8</td>
<td>41/155</td>
<td>26.5</td>
<td>33.84</td>
</tr>
<tr>
<td>D</td>
<td>22/73</td>
<td>30.1</td>
<td>37/130</td>
<td>28.5</td>
<td>-5.32</td>
</tr>
<tr>
<td>E</td>
<td>43/318</td>
<td>13.5</td>
<td>39/277</td>
<td>14.1</td>
<td>4.44</td>
</tr>
<tr>
<td>F</td>
<td>11/43</td>
<td>25.6</td>
<td>28/71</td>
<td>39.4</td>
<td>53.91</td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>69/401</td>
<td>17.2</td>
<td>122/591</td>
<td>20.6</td>
<td>19.77</td>
</tr>
<tr>
<td>Female</td>
<td>86/511</td>
<td>16.8</td>
<td>106/456</td>
<td>23.2</td>
<td>30.10</td>
</tr>
<tr>
<td>Season</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Spring</td>
<td>38/156</td>
<td>24.4</td>
<td>51/178</td>
<td>28.7</td>
<td>17.62</td>
</tr>
<tr>
<td>Summer</td>
<td>81/277</td>
<td>29.2</td>
<td>93/301</td>
<td>30.9</td>
<td>5.82</td>
</tr>
<tr>
<td>Autumn</td>
<td>31/381</td>
<td>8.1</td>
<td>59/411</td>
<td>14.4</td>
<td>77.78</td>
</tr>
<tr>
<td>Winter</td>
<td>5/98</td>
<td>5.1</td>
<td>25/157</td>
<td>15.9</td>
<td>211.76</td>
</tr>
<tr>
<td>Total</td>
<td>465/2736</td>
<td>17.0</td>
<td>684/3141</td>
<td>21.8</td>
<td>28.24</td>
</tr>
</tbody>
</table>

* Differences among the regions were found statistically significant in 2014 (P<0.01, χ²=32.353; df=5). * Differences among the regions were found statistically significant in 2015 (P<0.01, χ²=63.953; df=5). * Differences among the seasons were found statistically significant in 2015 (P<0.01, χ²=66.467; df=3). * Differences among the seasons were found statistically significant in 2015 (P<0.01, χ²=36.094; df=3).
Although *Theileria* was recognized to affect small ruminants, however, it was particularly less studied in goats (Taha *et al.*, 2011). The results of our serologic survey may provide a theoretical basis for the prevention of *Theileriosis* in this area.

**Acknowledgements:** This study was supported by the Wenzhou city public welfare science and technology plan projects (N20140041) and Startup Project of Doctor scientific research of Wenzhou Vocational College of Science and Technology in 2016 (201604).

**Authors’ contribution:** HQL, KL and JXW contributed in the plan of the study while HQL, KL, HZ, YFL, XX and JXW preformed the trial. HQL, KL, HGW and JXW analyzed the data. HQL and KL wrote the manuscript. All authors read and approved the final manuscript.

**REFERENCES**


