Article Title: Investigation and prevalence of hard ticks infestation in ruminant farm animals in the United Kingdom

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ABSTRACT

Ticks are haematophagic ectoparasites of domestic, wild animals and humans so an important part of the rationale is the risk of zoonotic disease. Globally, tick infestation causes considerable economic losses of approximately US$ 14-19 billion annually, although scientific data is limited. This study aimed to measure tick infestation amongst ruminant animals in the UK, to investigate tick prevalence, identify tick-species and risk factors associated with high tick prevalence. An online questionnaire and tick samples were used to estimate tick prevalence and identify risk factors. Tick infestation was distributed across the UK, with a higher prevalence in England (47%), Wales (28%) and Scotland (21%) compared to the Isle of Man (4%) and Northern Ireland (2%) at the time the study was conducted. Sixty-five percent of farmers that responded to the questions reported that they had previously had a tick-infestation in their herd or flock. The dominant tick species found was *Ixodes ricinus* (73%), followed by *Ixodes hexagonus* (18%) and *Dermacentor reticulatus* (10%). Upland farming (44%), not performing acaricide treatment (57%) and the presence of domestic pets and wildlife (67%) were significant risk factors. Although the prevalence rate is lower compared to other countries, this study provides evidence that tick infestation in livestock is a concern for UK farmers and warrants further investigation.

Keywords: *Ixodes ricinus*, ruminants, ticks, prevalence, risk factors and United Kingdom

Introduction

Ticks are blood-sucking arthropods that range between 0.5-11mm in size, and are found in almost every region of the world [1,2], including the United Kingdom (UK). They belong to two families Ixodidae (hard ticks) and Argasidae (soft ticks) of the order Ixodida and class Arachnida. Ticks are ectoparasites of domestic, wild animals and humans and present a zoonotic risk through transmission of pathogens of public health importance [3,4]. They are commonly found on cattle, deer, moose, rodents, squirrels, horses, cats, dogs and birds [5]. According to the European Scientific Counsel for Companion Animal Parasites (ESCCAP), ticks are active from March to June in the UK, during the season of spring to early summer. However, some ticks become active from August to October when the climate can be cooler than spring [6]. Ticks are biological vectors of pathogens of many human and animal tick-borne diseases (TBDs), including *Anaplasma phagocytophilum*, tick-borne macular fever, tick paralysis, babesiosis, tick-borne encephalitis, Q-fever, haemorrhagic fever and louping ill [7]. Approximately, 10% of the recognised 867 tick species are reported as vectors for TBDs [8].

Ticks are becoming much more common across the UK, particularly in heathland, moorland, grassland and woodland sites with their numbers increasing due to an increase in the number and distribution of deer [9]. Ticks that infest animals and humans belong to genera *Rhipicephalus*, *Haemaphysalis*, *Hyalomma* and *Ornithodoros* [10]. The most common tick species reported in the UK are *Ixodes ricinus, Ixodes hexagonus*, *Ixodes canisuga* [11]. Within the UK, the Tick Surveillance Scheme (TSS) run by Public Health England (PHE) is a citizen science programme that relies on the submission of ticks by members of the public and veterinary professionals for identification [12]. A total of 4172 ticks were submitted to TSS from humans and animals between 2005 and 2009 with 81% being identified as *Ixodes ricinus* [12]. Furthermore, since 2013, Larkmead and researchers at the University of Bristol have been

collecting ticks in the UK to help PHE in the identification of tick species and the diseases they transmit, with 89% ticks identified as *Ixodes ricinus* [2]. Tick-borne pathogens such as *Theileria ovis*, *Theileria recondita*, *Babesia motasi*, *B. crassa*, *B. capreoli* and *B. canis* were associated with tick species including *Haemaphysalis punctata*, *Ixodes ricinus* and *Dermacentor reticulatus* that were collected from sheep and dogs in Wales and southern England [7, 13]. Amongst wild animals, it was reported that 11% of red deer were infected with *Babesia divergens* and 16% were infested with *B. odocoilei* on farmland in the UK, hence wild deer are a potential reservoir for livestock transmitting *B. divergens* with red water fever [13].

Ticks and TBDs affect 80% of the world's cattle population and are widely distributed globally [14]. Tick infestation causes considerable economic losses in livestock animals, recently, the global economic loss has been estimated at USD $14-19 billion per year worldwide [15]. Damage is caused to animals in two ways, either direct or indirect. Direct damage causes impairment of the animal's growth and skin lesions, whereas indirect damage causes the transmission of variety of pathogens including rickettsia and other types of bacteria, protozoa and viruses to animals [15]. Tick infestation is a significant cause of economic losses in the dairy industry worldwide due to decreased milk yield in infested animals [16]. Sutherst *et al.*, 1983 and Norval *et al.*, 1988 have reported a 4 g loss in live mass gain caused by *Rhipicephalus appendiculatus* in Africa [17] and 0.6-1.5 g caused by *Boophilus microplus* in Australia [18]. In addition, heavy infestation of ticks in sheep may lead to anaemia [7]. In cattle, babesiosis manifests by clinical signs of fever including temperatures up to 41°C, depression, weakness, hemoglobinuria and anorexia [19].

Risk factors associated with tick prevalence on UK farms have been investigated and included large flock sizes, upland farming and the presence of sheep on cattle farms [20]. However, there are country to country variations in the risk factors associated with tick

prevalence [14, 21] and this needs to be considered when determining future studies on control and prevention measures against ticks. In the UK, the only authorised acaricides for controlling ticks in sheep are deltamethrin, alphacypermethrin pour-on or high-cis cypermethrin and diazinon plunge dips, which afford upto 6, 8 or 12 week of protection against ticks [7]. An infested animal can be treated with Ivermectin (0.2 mg/kg) and spraying of Deltamethrin (1%) twice every 14 days in the surrounding environment. These two treatments are the recommended options for the control of tick infestation amongst ruminant species [22].

In the UK, the total number of cattle (*Bos taurus*) is estimated to be 9.6 million, with sheep and lambs (*Ovis aries*) estimated at 22.8 million [23]. Goat (*Capra hircus*) production is less important in the UK and the total number of goats is estimated to be 105,029 [24]. There is no doubt that ruminant farm animal production is important to the UK, with 2 million cattle slaughtered in 2020. Livestock animals are reared according to strict legislation and under bio-secure conditions to limit the incidence of infectious diseases. Any condition that is detrimental to the health and welfare of the animal can impact on production and this is why the incidence of ticks and TBDs are important. However, data on tick infestation in ruminant farm animals in the UK is limited. Previous studies in the UK on tick infestation, largely focused on domestic animals, mainly dogs [25, 26]. This study therefore aimed to identify the most prevalent tick species, determine tick infestation amongst ruminant farm animals (cattle, sheep and goats) in the UK, and determine the risk factors associated with high prevalence.

Materials and Methods

Study area and period

Questionnaires and samples were collected from March to June 2021. The study area was first stratified into 5 regions based on the responses to the questionnaires obtained: Wales, England, Scotland, Northern Ireland and the Isle of Man (Fig. 1).

Study population

Ruminant farm animals including cattle, sheep and goats of all ages, sex, breed and body conditions were included in this study. There were no exclusion criteria. The inclusion criteria were that the respondents were aged over 18 and currently farmed ruminant animals in the UK.

Study design

A quantitative research design, using an adapted method from previous studies [27, 28] were used. Convenience sampling strategy was used in the study. A web-based questionnaire survey, generated using Microsoft forms (Office 365) was used to collect data from respondents concerning tick infestation problems. The questionnaire was divided into four sections which contained 10 open-ended and 23 closed questions: (i) Farm-related information, (ii) tick-related information on ruminant breeds, domestic and wild animals, (iii) risk factors associated with tick prevalence, (iv) a request for farmers to send in tick samples for speciation. The questionnaire was promoted in a monthly e-bulletin by the Farmer's Union of Wales. The questionnaires were also distributed with help from Hartpury Agri-Tech centre and through social media apps including Facebook, Twitter and LinkedIn. Microsoft Excel was used to collate the data obtained from the questionnaires.

The estimation of the prevalence of tick infestation was calculated according to the formula [29].

$$\text{Prevalence} = \frac{\text{Number of animals found positive} \times 100}{\text{Total number of animals studied}}$$

Collection and identification of tick samples

Tick sampling instructions, risk assessment, tick removal tools, stamped addressed envelopes and tick data collection sheet were provided to farmers, who indicated on their questionnaire that they would be prepared to collect and submit tick samples prior to sampling. The farmers were asked to remove ticks from the whole-body of ruminant animals including cattle, sheep and goats and then leave them in a secure location for 1 hour. The ticks were then sent to the laboratory. The collected ticks were preserved in 70% ethyl alcohol. Tick samples were identified using light microscopy (VWR, Leicester, UK) with the species of tick identified using the Bristol University Tick identification guide [30].

Ethical Considerations

The research compiled with all national regulations and local ethical approval was obtained from Hartpury University.

Statistical analysis

Statistical Analysis was carried out using Statistical Package for the Social Sciences (SPSS) [31]. The effect of age, breed, sex and other risk factors on tick prevalence was determined using the non-parametric Kruskal Wallis H test. To identify the effect of the different seasons (summer/spring/autumn/winter) on the prevalence of tick infestation, a post-hoc (Tukey HSD) test was used following the paired sample t-test. The accepted significant value was set at P=0.05.

Results

Questionnaire respondents

A total of 104 questionnaires were completed, with all respondents providing valid postcodes to the district level. Responses were received from farmers living in England (43%), followed by Wales (30%), Scotland (21%), Isle of Man (4%) and Northern Ireland (2%). Farmers reported previous occurrences of tick infestation in their herds or flocks and the overall prevalence of tick infestation was found to be 65% (n=68), with the prevalence of ticks higher in England (47%), compared to Wales (28%), Scotland (21%), Isle of Man (2%) and Northern Ireland (1%), but considering the total numbers of questionnaires received from the various geographical locations this result is to be expected. Of the total respondents, 27% (n=28) farmed breeding animals including cattle and sheep, 21% (n=22) were mixed farms including cattle, sheep and goats, 18% (n=19) farmed dairy, 6% (n=6) farmed sheep; and only 2% (n=2) farmed beef and were store farms. In total, 41% (n=43) farmed only cattle, 28% (n=29) farmed only sheep, 6% (n=6) farmed only goats and 25% (n=26) farmed all three ruminant animals (cattle, sheep and goats). The highest number of animals in a herd or flock was reported for breeding ewes (n=2300), followed by lambs (n=2000). Cattle herd sizes were reported to be lower in number (n=150), compared to sheep with the respondents not specifying goat herd size. The percentage of ruminant farm animals sold in the last 12 months was 55% (n=57) with 30% (n=31) sold at market and 25% (n=26) sold privately. The percentage of ruminant farm animals purchased in the last 12 months was 34% (n=35) with 19% (n=20) purchased at market and 14% (n=15) purchased privately.

Tick species

A total of 1026 ruminants (1000 sheep and 26 cattle) were examined on eight livestock farms in five different regions: Isle of Man (n=1000), Wales (n=12), England (n=11), Northern Ireland (n=2) and Scotland (n=1). In total, 40 ticks were collected belonging to three species. *Ixodes ricinus* (n=29; 73%) was the most common species, followed by *Ixodes hexagonus* (n=7; 18%) and *Dermacentor reticulatus* (n=4; 10%). A Kruskal-Wallis H test showed that there was a statistically significant difference between prevalence of the three examined tick species, $P<0.001$.

Tick-species collected from different body parts

Tick-species were collected from different body parts such as under armpits (40%), ear (28%), behind the shoulder (20%), around eyes (8%) and neck (5%). There was a statistically significant difference between armpits and other body parts examined for ticks, $P<0.001$, with a mean rank tick species of 8.50 for under armpits, 22.00 for ear, 31.50 for behind the shoulder, 37.00 for around eyes and 39.50 for neck.

Prevalence of tick infestation in ruminant farm animals

Tick prevalence was significantly higher in female (72%) compared to male (55%) animals, $P<0.001$, with a mean rank of 45.50 for female animals and 11.50 for male animals. It was also observed that older animals, aged 1-3 years (44%) and animals more than 3 years of age (35%), carried more ticks than animals less than 1 year of age (21%). The data showed a significant difference between agewise tick prevalence, $P<0.001$, with a mean rank of 7.50 for animals aged <1 year of age, 29.50 for animals aged 1-3 years and 56.50 for animals aged >3 years. The prevalence of tick infestation was found to be higher in Angus cattle (46%) compared to other cattle breeds, followed by ewes (43%) compared to other sheep breed and British

Primitive goat (42%) compared to other goat breeds. The results between the breeds were statistically significant, $P<0.05$.

Tick-borne diseases (TBDs)

Thirty-four respondents (74%) reported TBDs within their herd of flock. The most common TBDs was louping ill in sheep (22%), followed by tick-borne fever in cattle (20%), lyme borreliosis in cattle (13%), tick pyaemia in sheep (11%) and bovine babesiosis in cattle (9%; Fig. 2). The difference was statistically significant between louping ill and other TBDs examined in ruminant animals, $P<0.001$.

Symptoms of tick infestation and control measures

Behavioural changes in tick infested animals, including weight loss and drop in milk yields (35%; n=20), loss of condition or apathy (33%; n=19), general irritation or scratching (32%; n=18) were reported in 55% (n=57) of questionnaires. Almost 43% (n=45) of farmers reported that they use acaricide to control ticks e.g. Crovect, Ectofly, Zermasect, Spotinor and Bayticol insecticide spray (42%; n=19), injectable Imizol and Ivermectin (31%; n=14) and 27% (n=12) reported that they treat infested animals with gold fleece shower dip to control ticks. The results were statistically significant between insecticide spray over injections and dips, ($P<0.05$).

Season

Ticks were reported in all months throughout the year. A Kruskal-Wallis H test showed that there was not a statistically significant difference in prevalence between seasons, $P>0.05$. A Tukey's post-hoc test revealed that there was no difference in prevalence between seasons, between Apr-Jul and Aug-Nov ($P>0.05$), Aug-Nov and Dec-Mar ($P>0.05$) and Dec-Mar and Apr-Jul ($P>0.05$).

Risk factors associated with tick prevalence

There were several risk factors associated with tick prevalence with 67% (n=70) reporting the presence of domestic pets and wildlife on their farms, with the frequency of reporting ticks on wildlife and domestic pets at 58% (n=60), indicating that this could be a potential source for ticks on ruminant animals. In addition, 71% (n=74) reported water sources such as ponds, rivers and lakes in close proximity to their farms. In terms of farm descriptions, 44% (n=46) reported their farm as upland, 23% (n=24) as lowland, 22% (n=23) as shared grazing, 39% (n=41) reported their farms in higher level health and assurance schemes. Fifty-seven per cent of farmers (n=59) do not use acaricide to control ticks on their farms. All of these were significant risk factors for tick presence in ruminant farms animals on UK farms, ($P<0.05$) and the risks associated with these factors requires further investigation.

Discussion

Ticks are considered to be the most important vector of disease-causing pathogens in domestic and wild animals [32, 33]. Many researchers [7, 20, 34] have identified the prevalence of tick infestation in companion animals in the UK but the data from livestock is more limited. This study gives an indication of tick prevalence in livestock within the UK as tick samples were obtained from cattle and sheep in England, Wales, Scotland, Northern Ireland and Isle of Man, although a limited number of samples were obtained from each location. Tick infestation and TBDs leads to a decrease in milk yield, meat, fur and skin production [35, 36]. Recently, a study was conducted in the UK which revealed that the prevalence of ticks reported on sheep farms was 44% whereas it was 33% on cattle farms [20]. Approximately, 80% of the world cattle population are infested with ticks, particularly in tropical and sub-tropical countries (14, 22] including Pakistan, India, Bangladesh [37] and Egypt [38]. In Pakistan, a study was conducted and a total of 1050 cattle, 1400 sheep and goats were examined, out of which 75.1% cattle (n=789) and 51.6% goats (n=723) were found to be infested with ticks [39]. In a cross-sectional survey conducted by Sen *et al.*, (2012), a total of 1095 cattle were examined for tick infestation, of which 77.6% (n=850) were found to be infested with ticks [40]. These infestation rates are much higher than what is observed in UK studies, despite this, tick infestation and TBDs in ruminant farm animals are still a concern for UK farmers and warrant further investigation.

The results of the present study indicated that there are differences in the prevalence which is linked to geographical location, it was found that there was a higher prevalence of tick infestation in England (47%), compared to Wales (28%), Scotland (21%), Isle of Man (4%) and Northern Ireland (2%), however, this is perhaps not surprising as the majority of respondents in the present study were based in England. Lihou *et al.*, (2020) reported the prevalence of tick infestation on sheep and cattle farms was highest in Scotland (20%, 10%),

followed by Wales (16%, 7%) and northern England (16%, 6%) in a more thorough study [20]. The difference in prevalence between the studies, could be due to the sample size, as the total number of respondents in the present study is 104, whereas the Lihou *et al.,* (2020) study showed a larger sample size of 964, therefore sample size needs to be considered as an important factor for the difference in the results obtained. In addition, the tick population is affected by many factors including weather, climate and sampling strategy weaknesses [41, 42]. The distribution and abundance of a range of tick species have been attributed to change in factors such as climate, travel, management, habitat, economic patterns and increasing number of wild hosts particularly deer (11, 12, 43]. There is an association between ambient temperature and tick prevalence [44]. In the present study tick infestation was found to be higher in warmer months between April-July, compared to the colder months in Autumn between August-November and Winter between December-March. This agrees with Lihou *et al.,* (2020), who reported that the prevalence of ticks reported each month follows a normal distribution throughout the year with highest proportion reported during May-July [20].

The prevalence of tick infestation in different ruminant species, revealed that 4% of the total observed animals (n=1026) were found to be infested with ticks, with the highest rate in cattle (81%), followed by sheep (2%). The most prevalent tick species affecting ruminant animals was *Ixodes ricinus* (73%; n=29), followed by *Ixodes hexagonus* (18%; n=7) and *Dermacentor reticulatus* (10%; n=4). The results were statistically significant ($P<0.05$) with *Ixodes ricinus* being statistically higher than the other two species. Similar results were found by Cull *et al.,* (2018), who revealed that 4173 records were submitted to the TSS between 2010-2016 from companion animals including cats and dogs, the most frequently recorded tick species was *Ixodes ricinus* (59%), followed by *Ixodes hexagonus* (33%) [45]. The results are also in agreement with the findings of Smith *et al.,* (2011) who recorded that *Ixodes ricinus* (72%) is widespread across the UK, followed by *Ixodes hexagonus Leach* (22%), there was also five

cases of *Dermacentor reticulatus* in South-East England found [25]. Jameson and Medlock (2011) examined tick specimens sent to the Health Protection Agency, which was the agency PHE replaced, from academics, veterinarians and wildlife charities. Results confirmed that 81% of ticks submitted were *Ixodes ricinus*, confirming the dominance of this species in the UK [12]. This is also confirmed by Lihou *et al.* (2020) cites the work of Milne (1950) where it was demonstrated that *Ixodes ricinus* is a widespread tick species in the UK, that causes disease in livestock [20]. Similar findings were reported by Liddell *et al.*, (2020), where 719 *Ixodes ricinus* were collected from sheep in the South-West in Dartmoor National Park in Devon [34]. Exact percentages of tick species in ruminant farm animals from previous published literature is very limited. However, Gray *et al.*, (2019) reported that *Babesia venatorum* predominantly infest domestic sheep and wild deer in the UK [46]. These differences in tick numbers obtained between regions and studies could arise from differences in animal breeds, due to the type of pasture and location [47].

There was a significant difference ($P<0.05$) between tick infestations and sex, age and breed of the animal. The infestation rate was higher in female animals (72%) compared to male animals (55%), moreover, tick infestation was higher in animals aged 1-3 years (44%), followed in order by those aged >3 years (35%) and lowest in animals aged<1 year (21%). The infestation rate was higher in Angus cattle (46%), followed in order by breeding ewes (43%) and British primitive goat (42%). This agrees with the findings of Kabir *et al.*, (2011) who found that tick infestation rates were higher in female animals (59.37%), when compared to male animals (35.83%) [38]. Musa *et al.*, (2014) reported significantly higher prevalence (55.8%) in animals aged 3 to 7 years [48]. There is no data linking breed of the host animal to tick prevalence reported previously to our knowledge.

Overall, the prevalence of TBDs were significantly different ($P<0.05$) including, louping ill in ruminant sheep (22%), followed by tick-borne fever (20%) and lyme borreliosis in cattle

(13%), tick pyaemia in sheep (11%) and bovine babesiosis in cattle (9%; Fig. 2). Louping ill virus (LIV) is endemic to the UK, causing illness and even death in livestock especially sheep [49, 50]. LIV is mainly detected in sheep, red grouse, cattle, goats and deer and is associated with locations in Wales, Dartmoor, North Lancashire, Scotland, Cornwall and Ireland [7, 51] and flock morbidity ranges between 5 to 60% in tick infected areas [7]. The clinical signs associated with LIV in sheep include seizure activity, opisthotonos, ataxia and tremors [7]. Naïve sheep of all ages are at risk of LIV, whereas in infected areas young lambs are protected by colostral antibodies, therefore most cases of LIV occur in yearling sheep and weaned lambs. Furthermore, lambs that have poor intake of colostrum such as orphan lambs and multiple births are at risk of LIV [7]. Approximately, 30% of the tick infested lambs develop tick pyaemia [52]. However, the results are different compared to the findings of Lihou et al., (2020) who reported most common TBD in sheep was tick-borne fever (1.5%), whereas bovine babesiosis was most common in cattle (0.8%) [20]. Furthermore, the results are not in agreement with the findings of Johnson *et al.,* (2020) who noticed bovine babesiosis in five cattle in a herd of twenty in Southern England [53]. It has been reported that *Ixodes ricinus* is considered to be the primary vector for lyme borreliosis, which is caused by bacterium *Borrelia burgdorferi*, responsible for >1000 confirmed laboratory cases each year in Wales and England [54, 55]. Despite differences between studies, these variations may be due to differences in geographic focus, climate, time of year and examined sample population.

Analysis of the questionnaire revealed that tick infestation and TBDs in ruminant farm animals are treated with Acaricide spray such as Crovect, Ectofly, Zermasect, Spotinor and Bayticol (42%), injectable Imizol and Ivermectin (31%) and Gold fleece shower dip (27%) with statistically significant results of different brands used ($P<0.05$). A study conducted in the UK examined ectoparasites on goats, with 48% of farmers responding that preventive treatments against ectoparasites were given on their farms, of which 32% used ivermectin [56].

It is notable that for ruminant farm animals, the presence of domestic pets and wildlife on the farm (67%) were significant risk factors for tick infestation, likely due to ticks being present on these animals. The results are in accordance with Gilbert, (2010) who noticed that deer are an important host for ticks in Scotland [57], and deer are particularly attracted towards certain plants such as Chicory, Meadow Foxtail [58] and Buckwheat [59], which might be sufficient in quantity in the present study area. The present study also revealed that upland farming (44%) and not performing acaricide treatment (57%) were the significant risk factors for tick infesting ruminant farm animals. Lihou *et al.*, (2020) described upland farming as an important risk factor for tick infestation on sheep and cattle [20].

The present study is limited by a relatively small number of tick samples received and a lack of information regarding tick samples and TBDs infesting ruminant goats. Future research is needed where ticks are commonly found, particularly in the Lake District, Scotland, the New Forest, the Thetford Forest and the Yorkshire Moors [60]. The TSS, is the only scheme that records tick distribution on a national scale which is operated by PHE [45], therefore accurate data is needed to estimate tick prevalence in the regions where ticks are more common in the UK. Future research should also focus on a range of diagnostic tests used to detect TBDs in livestock, either through blood samples or polymerase chain reaction (PCR), in order to identify and distinguish between different types of tick-borne diseases in ruminant farm animals.

In conclusion, this study demonstrates tick infestation and risk factors associated with high tick prevalence in ruminant farm animals. Although the prevalence rate is lower compared to other tropical and sub-tropical countries, this study provides evidence that tick infestation in livestock is a concern for UK farmers and warrants further investigation.

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Authors contribution

The study was conceived and designed by SI and LW. Data collection and statistical analysis was performed by SI. The first draft of manuscript was written by SI. JS and LW revised the manuscript critically for important intellectual content and were involved in the revision of the manuscript. All authors read and approved the final version of the manuscript.

Conflict of Interest

Authors state no conflict of interest.

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Fig. 1. Map of United Kingdom showing the locations where tick samples were collected by livestock farmers from ruminant farm animals including cattle and sheep

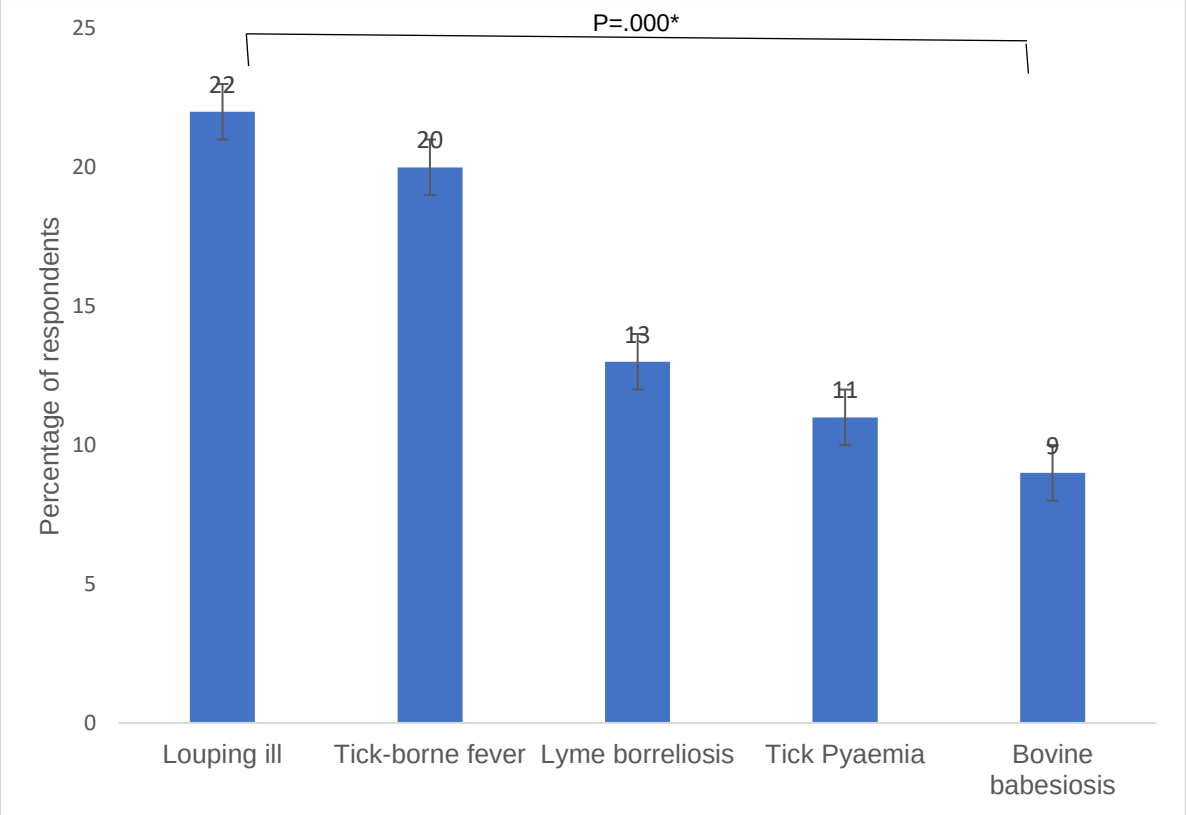

Fig. 2. The Prevalence (%) of TBDs reporting Louping ill (22%), tick-borne fever (20%), Lyme borreliosis (13%), Tick Pyaemia (11%) and Bovine babesiosis (9%) in cattle and sheep in the UK. * denotes statistically significant results P<0.05