Tick-borne encephalitis in Europe: Review of an emerging zoonosis

Tekenencefalitis in Europa: overzicht van een opkomende zoönose

[1]S. Roelandt, [2]P. Heyman, [1]P. Tavernier, [1]S. Roels

[1]WILDSURV project - Unit of Pathology –
Operational Direction Interactions and Surveillance
Veterinary and Agrochemical Research Centre (CODA/CERVA)
Groeselenberg 99, B-1180 Brussels

[2]Queen Astrid Military Hospital –
Research and Reference Laboratory for Vector-borne Diseases
Bruynstraat, Nederoverheembeek, B-1120 Brussels

sophie.roelandt@var.fgov.be

ABSTRACT

Tick-borne encephalitis virus (TBEV) is a highly pathogenic flavivirus and the most important arthropod-borne virus in Europe. Considerable increases in human tick-borne encephalitis (TBE) incidence in endemic countries and the emergence of the disease in several Western and Northern European countries, as well as in the European canine population, have made TBEV an increasing public health risk. Autochthonous TBE has never been reported in Belgium, despite the presence of suitable climate, environment, vectors and hosts. Though Belgian citizens are increasingly traveling to endemic areas, clinicians do not routinely test for TBE in human meningoencephalitis cases. In Belgium, TBE is not notifiable and surveillance is currently almost non-existent. If TBE were to emerge in Belgium, it could pose a significant threat to public health. Targeted serological screening of sentinel animals (dogs and wildlife) would contribute in a cost-effective way to a continuous epidemiosurveillance program for TBEV in Belgium.

SAMENVATTING

Tekenencefalitis virus (TBEV) is een sterk pathogeen flavivirus en het belangrijkste door arthropoden gedragen virus in Europa. Een aanzienlijke toename van de incidentie van menselijke tekenencefalitis (TBE) in endemische landen en het opduiken van de ziekte in verschillende West- en Noord-Europese landen en in de Europese hondenpopulatie hebben van TBEV een groeiend probleem voor de volksgezondheid gemaakt. Autochtone TBE werd nog niet gerapporteerd in België ondanks de aanwezigheid van een geschikt klimaat, milieu, vectoren en gastheren. Hoewel Belgische burgers in toenemende mate naar endemische gebieden reizen, onderzoeken clinici niet routinematig de aanwezigheid van TBE bij menselijke gevallen van meningo-encefalitis. In België is TBE niet aangifteplichtig en surveillance is momenteel bijna onbestaande. Als TBE zou opduiken in België zou het een belangrijke bedreiging voor de volksgezondheid kunnen vormen. Het gericht serologisch screenen van "sentineldieren" (honden en wilde fauna) zou op een kostenefficiënte manier bijdragen tot een continu epidemiologisch bewakingsprogramma voor TBEV in België.

INTRODUCTION

Tick-borne encephalitis virus (TBEV) is the most important arthropod-borne virus in Europe (Ramelow *et al.*, 1993; Herpe *et al.*, 2007). In Europe, the Western subtype of this highly pathogenic neurotropic flavivirus is carried by *Ixodes ricinus* (Kreil *et al.*, 1997; Labuda and Randolph, 1999; Stjernberg *et al.*, 2008). Tick-borne encephalitis (TBE) has become a considerable public health risk in several European countries (Süss *et al.*, 1997a), with currently 3000 hospitalized cases per year (Haglund, 2002). In many patients the disease results in long-term sequelae and disability (Donoso Mantke *et al.*, 2008a).

Recent increases and fluctuations in human incidence in Central and Eastern European countries (e.g. Switzerland, Germany, Poland, Baltic States) (Süss, 2008a/b) and the emergence of the disease in Finland (Jääskeläinen *et al.*, 2006), Norway (Csàngo *et al.*, 2004; Skarpaas *et al.*, 2004/2006), Denmark (Skarphédinsson *et al.*, 2005; Skarpaas *et al.*, 2006) and France (Herpe *et al.*, 2007) have sparked international concern and research. TBE is also emerging in Europe's canine population, and the numbers of clinical cases in dogs are expected to increase (Leschnik *et al.*, 2002; Beugnet and Marié, 2009).

This paper aims to highlight important features of TBE epidemiology, the clinical course in humans and

the surveillance possibilities for this tick-borne flavivirus. Afterwards, it will enter the discussion of whether Belgium could be at risk for TBE and whether national veterinary surveillance, in addition to medical surveillance, could be of benefit.

ETIOLOGY

TBEV is a small spherical enveloped RNA virus that belongs to the genus *Flavivirus* (*Flaviviridae*), which contains many neurotropic pathogenic arthropod-borne viruses. Its genome consists of a 10.5kb (approximately) single positive strand of RNA, encoding three structural and seven non-structural proteins (Heinz and Allison, 2000; Heinz, 2003; Gould *et al.*, 2004). Three lineages of TBEV have been described, namely the Western subtype transmitted by *I. ricinus* ticks and the Siberian and Far Eastern subtypes transmitted by *I. persulcatus* (Gubler *et al.*, 2007; Donoso Mantke *et al.*, 2008a). Besides the co-circulation of all subtypes in Estonia and Latvia (Golovljova *et al.*, 2004) and one focus of the Siberian subtype discovered in Finland (Jääskeläinen *et al.*, 2006), only the Western subtype is present in Europe (Heinz, 2008). Hereafter, only the Western subtype will be discussed, unless specified otherwise.

EPIDEMIOLOGY

Vector *Ixodes ricinus*

The castor-bean tick *I. ricinus* is the most important and most common European tick species and also the only tick in nature capable of sustaining infection with the Western TBEV subtype (Labuda and Randolph, 1999; Heinz, 2008). *I. ricinus* is a three-host tick with three life stages (larva, nymph, adult) and a two-year-long life cycle (six months to eight years) during which TBEV infection is passed transstadially, horizontally and transovarially. Horizontal tick-to-tick transmission occurs through viremic hosts or through simultaneous co-feeding of larvae with nymphs on the same host (Labuda and Randolph, 1999; Donoso Mantke *et al.*, 2008a; Heinz, 2008). TBEV infected ticks can be co-infected with many pathogens, but mainly with *Borrelia* spp., *Babesia* spp., *Anaplasma* spp. or *Coxiella burnetii* (Süss, 2003; Heinz, 2008).

Vertebrate hosts

I. ricinus has more than 300 known natural hosts, including mammals, birds and reptiles. Most of these hosts are subclinically infected with TBEV and become immune for life (Leschnik *et al.*, 2002; Süss, 2003).

Small rodents such as field mice (*Apodemus* spp.) show asymptomatic viremia and constitute competent reservoirs for immature ticks (Černý, 1975; Süss, 2003; Heinz, 2008), with rapid population turnover and a constant supply of susceptible individuals (Rizzoli *et al.*, 2004). Additionally, TBEV is able to replicate in immunocompetent cells in the skin of these mice, including immune or aviremic animals. The infected cells migrate to lymph nodes and back to feeding sites of uninfected co-feeding ticks. This allows non-viremic transmission between simultaneously co-feeding nymphs and larvae (Labuda *et al.*, 1993; Labuda and Randolph, 1999).

Large mammals such as roe deer (*Capreolus capreolus*) and domestic ruminants mainly serve as tick amplifying hosts. They are considered incompetent (i.e. unable to transmit), as they only develop low virus titers, though non-viremic co-feeding transmission may be possible (Labuda *et al.*, 1993; Süss, 2003; Rizzoli *et al.*, 2004). TBEV is excreted in the milk of viremic cows, goats and sheep and can be transmitted to humans this way (Gould *et al.*, 2006; Heinz, 2008). Birds also host immature *I. ricinus* and experience a short TBEV viremia. They seem to contribute to the spread of infected ticks (Gould *et al.*, 2004/2006; Heinz, 2008).

Dogs appear to be only occasional hosts for TBEV, though they may carry infected ticks from endemic to non-endemic areas and "into close vicinity of man" (Grešíková *et al.*, 1972a; Leschnik *et al.*, 2002). Although in 50% of dogs there is seroconversion without clinical signs (Grešíková *et al.*, 1972b; Klimeš *et al.*, 2001; Leschnik *et al.*, 2002), TBEV can nevertheless cause pyrexia, lethargy, inappetance and multifocal neurological signs (Kirtz *et al.*, 2001; Björsdorff, 2002). Humans are accidental dead-end hosts for ticks and for TBEV as, despite noticeable viremia, humans do not transmit the disease (Süss, 2003; Heinz, 2008).

Environment

Around 90% of TBEV endemic foci fall within specific climatic boundaries, such as the 7-8°C annual isotherm and the 800mm per annum precipitation minimum, resulting in the high soil humidity and 92% relative humidity required by *I. ricinus* for survival (Gritsun *et al.*, 2003; Labuda and Randolph, 1999; Heinz, 2008). Infected *I. ricinus* are often found questing for a host in mixed deciduous woodland with dense, humid ground layers and in animal feeding and resting places (Randolph, 2001; Heinz, 2008). Co-feeding of larvae and nymphs can only occur in areas where rapid autumnal cooling inhibits questing of unfed larvae, which will go into diapause until the next spring. At this time, rapidly rising temperatures will cause simultaneous reactivation of larvae and nymphs (Randolph, 2001; Gritsun *et al.*, 2003). Nymphal and larval seasonal feeding patterns will then overlap and this leads to co-feeding transmission (Labuda and Randolph, 1999). Without co-feeding, TBEV does not persist and an endemic focus cannot develop (Randolph, 2002; Gould *et al.*, 2004).

Distribution and incidence

The distribution of TBEV (all 3 subtypes) spans almost the entire Southern part of Eurasia, presently

from Alsace-Lorraine/Southern Norway (Donoso Mantke *et al.*, 2008a) to Vladivostok/Northeastern China/Japan, and it consists of up to 30,000 endemic foci (Korenberg and Kovalevski, 1999; Gritsun *et al.*, 2003). Worldwide, 10,000-12,000 cases of human TBE are reported annually (Süss, 2003), including more than 3,000 cases in Europe (Haglund, 2002). European TBEV is endemic in 27 countries (Süss, 2008a), of which the Central European countries, the Baltic States and Russia are most severely affected (Süss *et al.*, 1992; Korenberg and Kovalevski, 1999). In many of these countries TBE accounts for 50% of all cases of central nervous system (CNS) infection (Kaiser, 2008; Kunz, 2008). Western European incidence is currently <4 cases/100,000 inhabitants (Gubler *et al.*, 2006; Stjernberg *et al.*, 2008). However, in Great Britain, Ireland, Iceland, Belgium, the Netherlands, Spain and Portugal no autochthonous TBE cases have been reported to date (Donoso Mantke *et al.*, 2008a; Kunz, 2008).

During the last three decades, the TBE incidence has risen dramatically (2- to 17-fold) in most of the affected countries (e.g. Lithuania 1033%, Germany 574%, Europe as a whole 400%) (Randolph and Rogers, 2000; Stjernberg *et al.*, 2008; Süss, 2008a/b). Recently, new endemic foci have appeared in France, Greece, Denmark, Norway (Donoso Mantke *et al.*, 2008a), Italy (Pugliese *et al.*, 2007), Sweden (Stjernberg *et al.*, 2008), Finland (Jääskeläinen *et al.*, 2006) and Germany (RKI, 2009), and new risk areas are being discovered every year. TBE has rapidly become a growing public health problem (Süss, 2008a/b) and is currently the most important vector-borne viral infection in Europe (Labuda *et al.*, 2006; Donoso Mantke *et al.*, 2008a).

Several authors have discussed the many etiological factors that may cause a rise in incidence, prevalence and distribution of vector-borne diseases (VBDs), and specifically of TBE. These include various sociological, technological and ecological factors (Table 1). Whereas recent TBE emergence in Scandinavia has been linked predominantly to climate change (milder winter and earlier spring) (Lindgren and Gustafson, 2001), expansion of roe deer and tick populations (Randolph, 2001) and increased awareness (Haglund, 2002), increases in incidence in certain areas of Germany during the 1990s seem to be related to increased surveillance and improved diagnostics (Randolph, 2001). As opposed to this, in Eastern European countries the political change caused by the end of communism also resulted in many agricultural and social changes (Randolph, 2001). As a consequence, the increased consumption of unpasteurized milk and the increased use of forests and previously abandoned countryside areas for food collecting or leisure activities have led to increased exposure to *I. ricinus* and TBEV (Randolph and Rogers, 2000; Beltrame *et al.*, 2006). Finally, data indicate that the dramatic increase of human TBE cases in several European countries during the hot summer of 2006 can be explained mainly by an increase in human outdoor activity in response to the unusual weather of that year (Randolph *et al.*, 2008).

Modeling and risk maps

Geographic information and remote sensing systems (GIS and RS) offer great opportunities for environmental health research, particularly of VBDs and zoonoses such as TBE (Labuda and Randolph, 1999; Rinaldi *et al.*, 2006). TBE risk maps can be drawn based on satellite-derived GIS and RS data on environmental and climatic characteristics and based on virus and host survival requirements. Current TBE risk areas can be predicted with 85% accuracy (Randolph and Rogers, 2000; Randolph, 2000/2001; Figure 1) and risk maps based on reported human TBE cases are published regularly (Figure 2).

Risk maps give a good but incomplete picture of the European situation, as they usually do not take into account the prevalence of TBEV in the tick population or animal seroprevalence (Süss, 2003; Rendi-Wagner, 2004). During the 20th century, modest global warming combined with changing human behavior have resulted in TBEV spread, new foci and increased TBE incidence (Randolph and Rogers, 2000; Randolph, 2004; WHO, 2004).

Predictions for TBEV distribution during the next 80 years have been attempted based on different climate change scenarios. On the basis of these predictive models, it could be speculated that endemic regions may further disperse geographically in any one, two or even three directions − eastwards, northwards and/or westwards (Randolph and Rogers, 2000; Randolph, 2001; S. Randolph, personal communication, 2009) − and that none of the possible (combinations of) directions can be ruled out. This evolution seems to be taking place in Sweden, Germany and France (Kirtz *et al.*, 2001; Lindgren and Gustafson, 2001; Van Der Poel *et al.*, 2005).

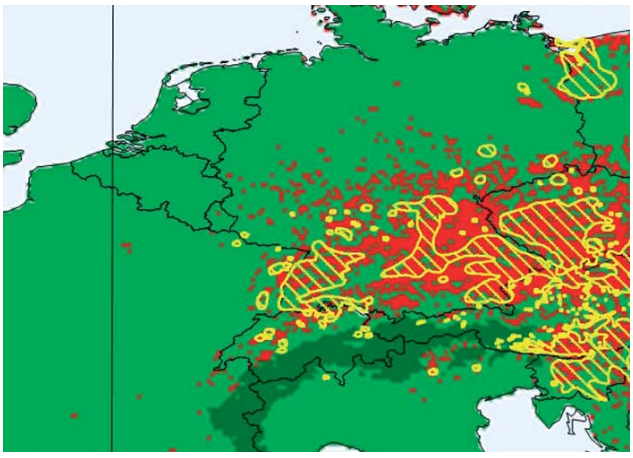

Figure 1. Satellite-derived predicted current distribution of Tick-borne encephalitis in Western and Central Europe (red, 2000) compared with established foci (yellow, mapped 1997) (From Randolph and Rogers 2000 and Randolph 2001, with permission from The Royal Society and Prof. S. Randolph).

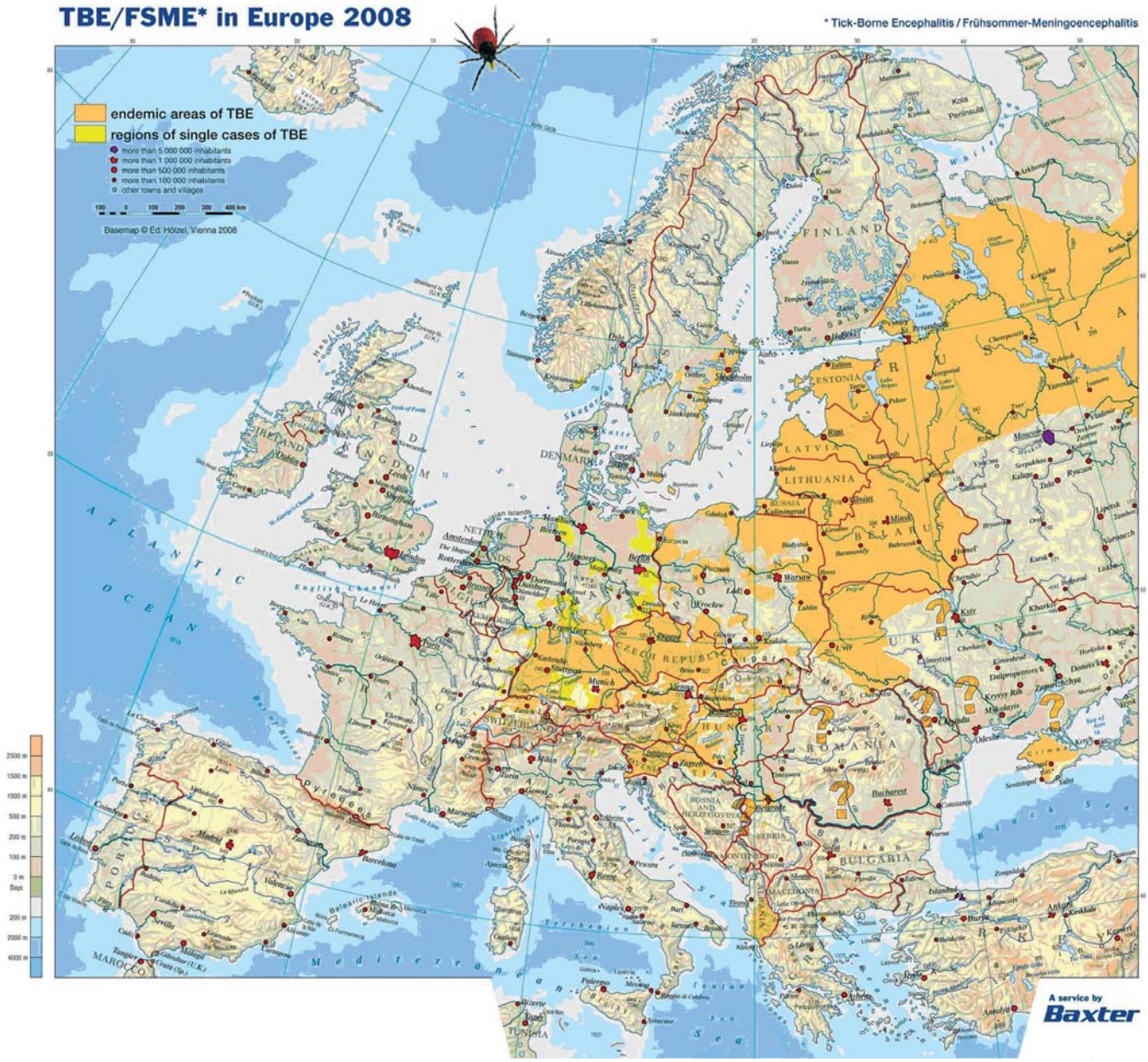

Figure 2. TBE in Europe: Established endemic areas in 2008 (From ISW-TBE 2008, with permission from Baxter/Hölzel Verlag).

TICK-BORNE ENCEPHALITIS IN HUMANS

Exposure

Humans usually become infected with TBEV through bites from *I. ricinus* larvae, nymphs or adults. In affected countries, people contract TBE during spring and summer through working outdoors (e.g. farmers, forest workers, military personnel) (Haglund, 2002; Donoso Mantke *et al.*, 2008a) or through leisure activities such as sports, hunting, fishing, rambling/hiking, berry/mushroom/wood collecting, or through the consumption of raw (unpasteurized) goat, cow and sheep milk (Süss, 2003/2008a; Heinz, 2008). The risk of human infection after a single tick bite in an endemic area varies from 1/200 to 1/1000 (Süss, 2003/2008a).

Clinical course

TBEV is neurotropic and clinical signs vary from mild fever to meningitis, encephalitis or myelitis (Haglund *et al.*, 2003). Between one-third and two-thirds of the patients experience a typical biphasic course, starting with non-specific influenza-like symptoms and a brief viremia (1-4 days), followed by an asymptomatic interval (Lešničar *et al.*, 1997; Kaiser, 1999/2008). This is followed by a second febrile stage around two to four weeks post-infection, during which patients will develop one of four possible clinical manifestations: meningitis, meningo-encephalitis, meningo-encephalomyelitis or meningo-radiculoneuritis (Kaiser, 1999/2008; Donoso Mantke *et al.*, 2008a). This is usually when medical advice is sought and the first diagnostic investigations take place (Holzmann,

2003). Natural infection results in lifelong immunity (Mickiené, 2008). A mortality rate of 0-3.9% has been reported in Europe (Haglund; 2002; Donoso Mantke *et al.*, 2008a; Süss, 2008a), but morbidity and mortality rates increase with increasing age of the patient (Günther *et al.*, 1997; Lešničar *et al.*, 1997; Mickiené *et al.*, 2002).

Prognosis

Regardless of the clinical course, symptomatic patients need hospitalization for weeks to months, and between 35% and 58% of them are left with permanent sequelae called "post-encephalitic syndrome" (Haglund *et al.*, 1996; Günther *et al.*, 1997; Kaiser, 1999/ 2008). Patients who develop this syndrome require many years of costly treatment and rehabilitation due to long-term neurological symptoms, cognitive and psychiatric dysfunctions, social distress, inability to work (94% of cases) and loss of life quality. TBE comes at a very high cost to society and the health care system due to this high morbidity (Mickiené *et al.*, 2002; Kunz, 2008; Baumhackl, 2009).

Diagnosis

TBE can only be diagnosed by laboratory testing, usually by means of ELISA, which detects specific IgM and IgG antibodies in serum or cerebrospinal fluid (CSF) (Holzmann, 2003; Mickiené, 2008). Due to possible cross-reactions in the case of flavivirus vaccination or infection, positive tests need to be confirmed by seroneutralization (SN), hemagglutination inhibition (HI), indirect immuno-fluorescence (IIF) or immunoblot tests such as western blot (WB) (Niedrig *et al.*, 2001; Holzmann, 2003; Mickiené, 2008). SN assays are the most specific confirmation tests for TBEV and are especially indicated in the case of suspected ELISA-positive patients in areas where TBEV has not been found before. Since SN assays are expensive and technically complex, they are only performed by specialized reference laboratories (Holzmann, 2003; Mickiené, 2008).

Demonstration of viral RNA in blood by RT-PCR is only possible during viremia, which lasts one to four days after the onset of clinical signs and has usually ceased by the time of presentation and clinical investigation. Therefore RT-PCR techniques are less useful for TBE diagnosis in patients (Puchhammer-Stöckl *et al.*, 1995; Ducoffre, 2007).

Differential diagnoses

TBE has to be differentiated from many other infectious encephalitides, including Lyme borreliosis, human granulocytic anaplasmosis (HGA), measles, herpes simplex and West Nile virus encephalitis (Donoso Mantke *et al.*, 2008b; Mickiené, 2008). Co-infection with *Borrelia burgdorferi sensu lato* (Lyme borreliosis) occurs in 15% of patients, while *Anaplasma phagocytophilum* (HGA) can be demonstrated occasionally (Kaiser, 2008).

Treatment and prevention

As opposed to Lyme borreliosis, which can be managed by antibiotic therapy, TBE can only be treated symptomatically through maintenance of hydration and caloric intake, analgesics, antipyretics and vitamins. Strict rest and physiotherapy are also paramount to avoid complications (Kunz, 2008; Kunze, 2008). TBE can easily be prevented by vaccination, which is strongly advised for people of all ages who live or travel in endemic areas (Kunz, 2008; Mutz, 2008; Steffen, 2009). General prevention also includes avoidance of tick-infested areas, wearing light-colored clothing covering as much skin as possible, application of diethyltoluamide (DEET) or permethrin, and performing a daily tick check and removal (ITG, 2008; Mutz, 2008; Luyasu, 2009).

SURVEILLANCE OF TBEV IN HUMANS, TICKS AND SENTINELS

Surveillance in humans

In endemic areas, human clinical cases are registered and geographically mapped, while tick-exposed persons are serologically screened (Donoso Mantke *et al.*, 2008a; Süss, 2008a; RKI, 2009). Human seroprevalence in endemic areas ranges from 4-22% (Günther *et al.*, 1997; Süss, 2008a). It is known that TBE endemicity outside known foci can be underestimated, mostly due to not considering TBE in the differential diagnosis of CNS infections (Haglund, 2002; Donoso Mantke, 2008b). Additional systematic surveillance of tick numbers, tick TBEV prevalence and sentinel animal seroprevalence is therefore necessary to determine the true epidemiological situation in and outside natural foci (Süss, 2008a; Donoso Mantke *et al.*, 2008a; RKI, 2009).

Surveillance in ticks

As opposed to humans and animals, ticks are lifelong TBEV carriers and the virus usually remains detectable (Süss *et al.*, 1999; Süss, 2003). RT–PCR techniques have proven their value for detecting TBEV-RNA from ticks in epidemiological studies and surveillance programs, thereby replacing virus isolation and mouse inoculation (Puchhammer-Stöckl *et al.*, 1995; Süss *et al.*, 1997b; Schrader and Süss, 1999).

Ticks are collected from vegetation by the flagging technique or from dogs, mice and wildlife in the region (Skarphédinsson *et al.*, 2005; Van Der Poel *et al.*, 2005; Skotarczak *et al.*, 2008). They are identified with standard taxonomic keys (Hamer *et al.*, 2009) and TBEV-RNA is detected through sensitive nested nRT-PCR and confirmed through sequencing or Southern Blot (SB) hybridization (Süss *et al.*, 1996/1999; Schrader and Süss, 1999; Van Der Poel *et al.*, 2005).

Average tick TBEV prevalence in affected countries varies spatially and temporally from 0.1-5% (Süss *et al.*, 1999; Leschnik *et al.*, 2002), with local extremes

of up to 26.6% (Latvia) (Süss, 2008a). In areas with low incidence/prevalence, infected ticks are often not detected (Süss, 2003). However, tick surveillance remains essential for understanding tick ecology and TBE epidemiology (Stjernberg *et al.*, 2008).

Surveillance in sentinels

Pets as well as farm animals and wildlife can be used as sentinels for disease detection (Wobeser, 2006; Gubernot *et al.*, 2008), offering a practical index for public health surveillance and risk assessment of zoonotic diseases (Schrader and Süss, 1999; Leighton *et al.*, 2001; Wobeser, 2006). TBEV seropositivity has been found in many domestic and wild animals (Rieger *et al.*, 1999; Björsdorff, 2002). Dogs (*Canis vulgaris*), cattle (*Bos taurus*), goats (*Capra hircus*), wild boar (*Sus scrofa*), roe deer (*Capreolus capreolus*), red foxes (*Vulpes vulpes*) and wild rodents such as the yellow-necked field mouse (*Apodemus flavicollis*), the wood mouse (*Apodemus sylvaticus*), the common shrew (*Sorex araneus*) and the common vole (*Microtus arvalis*) have proven to be good TBEV sentinels (Björsdorff, 2002; Leschnik *et al.*, 2002) and have been used in several surveillance studies (Gerth *et al.*, 1995; Csàngo *et al.*, 2004; Skarphédinsson *et al.*, 2005; Van Der Poel *et al.*, 2005; Herpe *et al.*, 2007; Hamer *et al.*, 2009). In some studies high antibody prevalences were found, for example in the yellow-necked field mouse 47.9%, the red fox 18.0%, roe deer 83%, dogs 24%, goats 44% and cattle 91% (Leschnik *et al.*, 2002; Süss, 2008a).

In European sentinel studies in dogs, IgG seroprevalences were in the range of 0.97-24% (Leschnik *et al.*, 2002) and were usually higher than for humans, most likely due to the more frequent contact of dogs with *I. ricinus* (Klimeš *et al.*, 2001; Björsdorff, 2002). Sentinel dogs can also reveal that TBEV is more common than expected in certain geographical areas (Björsdorff, 2002). In Norway, 16.4% of screened dogs tested seropositive for TBEV in an area considered non-endemic, which was unanticipated. The study facilitated the discovery of a new focus and highlighted the need for better TBEV monitoring in Norway (Csàngo *et al.*, 2004; Skarpaas *et al.*, 2004).

Game animals are often infested with ticks and were screened for anti-TBEV-IgG by ELISA, HI and SN in several European countries (Rieger *et al.*, 1999; Süss, 2003). A Danish study in roe deer measured 8.7% seropositivity and revealed a marked shift in Danish TBEV distribution (Skarphédinsson *et al.*, 2005). A Dutch study found 0.5% of foxes, 7% of wild boar and 3% of cattle ELISA-positive, but this was not confirmed by the follow-up SN and HI tests. It was concluded that TBEV prevalence is currently either absent or very low (≤0.3%) in the Netherlands. Nevertheless, a future westwards expansion of TBEV cannot be excluded (Gould *et al.*, 2004; Van Der Poel *et al.*, 2005).

Regular investigation of rodents and their ticks can lead to TBE viral strain identification (Herpe *et al.*, 2007) and could reveal dynamic pathogen distribution in foci (Heyman *et al.*, 2002). TBEV prevalence in vertebrate hosts from endemic areas can be much higher than in the local tick or human populations, and it correlates better to the true TBEV endemicity of the area (Rieger *et al.*, 1999; Leschnik *et al.*, 2002; Süss, 2008a).

DISCUSSION

Belgian climate, vegetation and ticks

According to the FAO Global Ecological Zoning Report (2001), Belgium falls within the European "Temperate Oceanic Forest Ecological Zone" (TeDo-EZ), in which climate is "influenced by the Gulf Stream and by proximity to the ocean". Average annual temperatures vary from 7-13°C and annual rainfall from 600-1700mm (FAO, 2001; KMI, 2009). Vegetation in this EZ is dominated by deciduous and mixed deciduous/coniferous forests, cultivated land and different types of wetlands (FAO, 2001; AGIV, 2009; RW, 2009), offering many suitable habitats to *I. ricinus* ticks, which are widely present in Belgium (Misonne *et al.*, 1998; E. Claerebout, personal communication, 2009).

To the authors' knowledge, it is currently unknown whether *I. ricinus* co-feeding occurs in Belgium and therefore whether TBEV foci could potentially develop and persist. In the 1990s, exotic *R. sanguineus* and *D. reticulatus* ticks were introduced by traveling dogs and small endemic foci were established in Belgium and the Netherlands (Losson *et al.*, 1999; Matjila *et al.*, 2005; Beugnet and Marié, 2009; Heyman, 2009).

Tick-borne diseases (TBDs) in Belgium

Two tick-borne human pathogens present in Belgium are *B. burgdorferi* and *A. phagocytophilum,* which cause Lyme disease and human granulocytic anaplasmosis (HGA), respectively. In recent years, the reported incidences of these diseases have been rising, most likely through a combination of genuine pathogen emergence and increased awareness and diagnosis (Randolph, 2001; Ducoffre, 2008; Heyman, 2009). Lyme disease risk areas now include several areas in Northern and Southern Belgium, and the bulk of human HGA cases have been diagnosed in Flanders (Northern Belgium) (Ducoffre, 2008). TBE is still considered to be an exotic disease in Belgium and, so far, no autochthonous cases have been reported (Donoso Mantke *et al.*, 2008a; Süss, 2008b).

Canine TBD distribution has now also extended towards the lowland countries with the appearance of the first autochthonous cases of canine babesiosis (*Babesia canis*) and Lyme disease in dogs (McKenna *et al.*, 1995; Losson *et al.*, 1999; Matjila *et al.*, 2005; Hovius and Houwers, 2007; Van de Maele *et al.*, 2008). TBE has not yet been reported in Belgian dogs. Nonetheless, in Germany and France, TBEV endemic areas have shown a clear western expansion, increasing the risk for local and traveling Western European dogs (Kirtz *et al.*, 2001; Leschnik *et al.*, 2002).

Table 1. Risk factors increasing incidence, prevalence and distribution of vector-borne diseases / tick-borne encephalitis.

Risk Factor	Literature reference
Increased human population density	Randolph (2001); Süss (2008a)
Increased urbanization and abandonment of countryside	Beltrame *et al.* (2006); Süss (2008a)
Human migration towards suburban areas	Merino *et al.* (2000); Randolph (2001); Süss (2003); Linard *et al.* (2007); Heinz (2008); Süss (2008a); Beugnet and Marié (2009); Linard and Vanwambeke (2009)
Increased travel of humans and dogs	Haglund (2002); Leschnik *et al.* (2002); Süss (2003); Rendi-Wagner P. (2004); Donoso Mantke (2008a); Kunz (2008); Beugnet and Marié (2009); BSAVA (2009); Heyman (2009); Luyasu (2009)
Change in leisure and outdoor activities	Randolph and Rogers (2000); Lindgren and Gustafson (2001); Randolph (2001); Haglund (2002); Süss (2003); Beltrame *et al.* (2006); Donoso Mantke (2008a); Heinz (2008); Kunz (2008); Randolph *et al.* (2008); Süss (2008a); Beugnet and Marié (2009)
Displacement due to conflict/war	Süss (2008a)
Exotic disease introduction	Süss (2008a); Beugnet and Marié (2009); BSAVA (2009)
Social/political/economic factors and change	Merino *et al.* (2000); Randolph and Rogers (2000); Randolph (2001); Süss (2003); Beltrame *et al.* (2006); Linard *et al.* (2007); Donoso Mantke (2008a); Heinz (2008); Randolph *et al.* (2008); Beugnet and Marié (2009); Linard and Vanwambeke (2009)
Change in agricultural practices	Randolph (2001); Süss (2003); Süss (2008a)
Increased consumption of raw milk	Randolph and Rogers (2000); Randolph (2001); Süss (2003); Heinz (2008);
Increased transport of goods and domestic/ wild animals	Beugnet and Marié (2009)
Occupational exposure: agriculture, military, forestry, hunters, laboratory	Randolph (2001); Süss (2003); Süss (2008a); Donoso Mantke (2008a)
Human impact on landscape, land use change	Randolph (2001); Beltrame *et al.* (2006); Randolph *et al.* (2008); Beugnet and Marié (2009); Linard and Vanwambeke (2009)
Increased reforestation/conservation of tick habitat leading to increasing host and tick populations	Lindgren and Gustafson (2001); Randolph (2001); Haglund (2002); Randolph *et al.* (2008); Stjernberg *et al.* (2008); Süss (2008a); Beugnet and Marié (2009); Linard and Vanwambeke (2009)
Ecological factor: climate change, global warming	Randolph and Rogers (2000); Lindgren and Gustafson (2001); Randolph (2001); Haglund (2002); Donoso Mantke (2008a); Heinz (2008); Stjernberg *et al.* (2008); Beugnet and Marié (2009); Heyman (2009)
Environmental factor: biotope, vegetation	Randolph (2001); Haglund (2002); Linard *et al.* (2007); Heinz (2008); Stjernberg *et al.* (2008); Beugnet and Marié (2009)
Higher awareness in the medical community and among the population in general	Lindgren and Gustafson (2001); Randolph (2001); Haglund (2002); Beltrame *et al.* (2006); Donoso Mantke (2008a) ; Stjernberg *et al.* (2008)
Improved diagnostic procedures	Randolph (2001); Haglund (2002); Donoso Mantke (2008a); Stjernberg *et al.* (2008)
Increased surveillance	Randolph (2001); Beltrame *et al.* (2006); Stjernberg *et al.* (2008)

Present TBD/TBE surveillance in Belgium

Though Belgium introduced systematic reporting of Lyme borreliosis during the 1980s and of HGA from 2004 onwards (Randolph, 2001; Ducoffre, 2008), there is no mandatory screening or notification for TBE (Donoso Mantke *et al.*, 2008a; Süss, 2008b). An international follow-up study of human cases with presumed viral CNS infection of unknown etiology included four patients from Southern Belgium. IgG ELISA of serum and CSF revealed one borderline and two TBE seropositive samples. None were confirmed by SN and it was concluded that these were false positives due to unspecific or cross-reactivity (Haglund *et al.*, 2003).

National TBE surveillance is currently limited to case-by-case diagnostics in humans at the Reference Laboratory of Vector-borne Diseases in the Queen Astrid Military Hospital in Brussels. PCR is available for acutely viremic patients, though ELISA is the test of choice (Ducoffre, 2007b; P. Heyman, personal communication, 2009). Although several patients with "viral encephalitis" who had not traveled recently tested positive for TBEV in ELISA, confirmation in these cases by SN tests was unsuccessful (P. Heyman, personal communication, 2009).

A recent analysis showed that in Europe between 30% and 80% of human viral meningo-encephalitis cases remain unexplained. This is mainly due to a lack of standardization in surveillance, diagnostic and notification procedures, and to the fact that occasionally a lack of effort or cooperation leads to insufficient reporting (Donoso Mantke *et al.*, 2008b).

Necessity of veterinary TBE surveillance in Belgium

It has become evident that the epidemiology of arthropod-borne diseases is changing in Europe and worldwide. According to Randolph (2001) and Otranto and Wall (2008), this necessitates "continued and new measures to survey their epidemiology", as well as the development of "new alert and control strategies". TBEV is undoubtedly the most important arbovirus in

Europe, causing considerable morbidity and non-negligible mortality (Kunz, 1992; Klimeš *et al.*, 2001), and leading to high costs for the healthcare systems and for society in general (Günther *et al.*, 1997; Kaiser, 1999; Haglund, 2002). The recently observed increases of TBE incidence in affected countries and the discovery of new foci in other countries are prompting governments to establish increased surveillance of the endemicity of TBEV in Europe (Haglund, 2002; Donoso Mantke *et al.*, 2008a).

Belgian citizens are increasingly traveling to Eastern European countries with known TBE endemic areas (Luyasu, 2009). It has been estimated through risk analysis that 1/10,000 tourists will acquire TBE during a 4-week-long stay in an endemic area (Rendi-Wagner, 2004; Luyasu, 2009). Since general TBE awareness among travelers and their physicians is low, usually no systematic efforts at prevention, diagnosis or treatment are initiated in these people (Rendi-Wagner, 2004; Donoso Mantke *et al.*, 2008a). An increasing number of travel related human TBE cases may be expected in the future (Rendi-Wagner, 2004; Luyasu, 2009). In the Netherlands, two TBE cases in Dutch travelers returning from abroad have been confirmed in the laboratory (Aendenkerk *et al.*, 1996; Kessels *et al.*, 1999). Moreover, TBEV antibodies have been detected in Dutch forestry workers (no confirmatory SN assay) (Moll van Charante *et al.*, 1998) and tick-bite patients (2.4%). Therefore, a number of infections have probably already occurred in the Netherlands (Groen, 1993; Van Der Poel *et al.*, 2005).

TBE is apparently emerging in the canine population in Western Europe and, given the dramatic increase in human cases, it is likely to be more frequently diagnosed in dogs in the future (Leschnik *et al.*, 2002; Beugnet and Marié, 2009). TBE is also of relevance to food chain management due to the possible transmission of TBEV through raw milk of viremic ruminants (Gritsun *et al.*, 2003; Kerbo *et al.*, 2005). All of the above observations would justify increased veterinary research and sentinel surveillance for TBEV in Belgium.

Veterinary surveillance possibilities

For the management of TBE in humans, systematic identification of endemic areas is necessary. This can be accomplished through the assessment of tick numbers and TBEV prevalence in ticks and sentinel animals (Süss, 2003/2008a; Donoso Mantke *et al.*, 2008a; RKI, 2009). However, in areas with low TBE incidence and only rare transmission to humans, infected ticks are often not detected (Süss, 2003). In other areas, where mass vaccination has decreased human incidence (e.g. Austria), surveillance in humans alone becomes insufficient for risk assessment (RKI, 2009). In these cases the use of sentinel surveys is very useful, since animals (particularly wildlife) are still exposed, and since their seropositivity reflects the true TBE endemicity of the area better than human incidence (Rieger *et al.*, 1999; Merino *et al.*, 2000). Disease investigation, especially in wildlife, often benefits from cost-effective, risk-based surveillance with sampling targeted towards high-risk populations (Hadorn and Stärk, 2008). Consequently, sentinel surveillance both in domestic and wild animals would significantly increase the chance of detecting possible TBEV endemic foci in Belgium.

CONCLUSION

"Even though TBE was described as early as 1931, this dangerous form of encephalitis has been underestimated for a long time" (Kunz, 2008). The total absence of reported human cases in Belgium is perhaps questionable since Belgian citizens and their dogs regularly travel to known endemic areas (Heyman, 2009; Luyasu, 2009). Furthermore, Belgium has climatic and environmental conditions similar to those of other European countries where TBEV is endemic (FAO, 2001; AGIV, 2009; RW, 2009) and suitable vectors and hosts are present and abundant (Misonne *et al.* 1998; Verkem *et al.*, 2003; E. Claerebout, personal communication, 2009).

However, presently clinicians do not routinely test for TBE and notification of clinical cases is not mandatory (Randolph, 2001; Donoso Mantke *et al.*, 2008a/b; Süss, 2008b). Until now, TBE surveillance in Belgium has been almost non-existent and human or canine cases could currently remain undiagnosed. The establishment of endemic foci with low prevalence could easily be missed, as was the case in Scandinavia (Haglund, 2002). Just as increased awareness and testing have probably contributed to the rise in Belgian cases of Lyme disease (Ducoffre, 2008), so too may increased veterinary and human surveillance demonstrate the arrival or circulation of TBEV in Belgium.

ACKNOWLEDGMENTS

This work is a part of the Wildsurv Project and was funded by a grant from the Belgian Federal Government Department of Public Health. The authors would like to thank Prof. E. Claerebout and Prof. S. Randolph gratefully for their personal communications on the subjects of *I. ricinus* and TBEV.

SELECTED REFERENCES

The extensive reference list can be requested from the authors.

Donoso Mantke O., Schädler R., Niedrig M. (2008a). A survey on cases of tick-borne encephalitis in European countries. *Eurosurveillance 13(4–6).* www.eurosurveillance.org

Haglund M. (2002). Occurrence of TBE in areas previously considered being non-endemic: Scandinavian data generate an international study by the International Scientific Working Group for TBE (ISW-TBE). Mini review. *International Journal of Medical Microbiology 291, Suppl.33*, 50-54.

ISW-TBE. International Scientific Working Group on Tick-

Borne Encephalitis. Baxter. (2008). Map TBE/FSME* in Europe 2008. Copyright Baxter – Hölzel Verlag. Downloaded on 25/9/09. http://www.isw-tbe.info/upload/medialibrary/FSME_inet_klein_e2009.jpg

Lindgren E., Gustafson R. (2001). Tick-borne encephalitis in Sweden and climate change. *Lancet 358,* 16-18.

Luyasu V. (2009). How to protect the tourist: preventive measures for Lyme borreliosis and vaccine against tick-borne encephalitis. In: *Proceedings of the International Conference: Climate change impact on ticks and tick-borne diseases,* Brussels, Belgium 2/2/2009.

Randolph S.E., Rogers D.J. (2000). Fragile transmission cycles of tick-borne encephalitis virus may be disrupted by predicted climate change. In: *Proceedings of the Royal Society of London B 267,* 1741-1744.

Randolph S.E. (2001). The shifting landscape of tick-borne zoonoses: tick-borne encephalitis and Lyme borreliosis in Europe. *Philosophical Transactions of the Royal Society of London B 356,* 1045-1046.

Schrader C., Süss J. (1999). A nested RT-PCR for the detection of tick-borne encephalitis virus (TBEV) in ticks in natural foci. *Zentralblatt für Bakteriologie 289,* 319-328.

Süss J., ISW-TBE Study Group (2008a). Epidemiology. In: *Compendium of Tick-borne encephalitis (TBE, FSME). Monograph, Baxter, ISW-TBE*, pp. 15-20. http://www.isw-tbe.info/tbe.aspx_param_target_is_51410_and_1_is_2.v.aspx

Van Der Poel W.H.M., Van Der Heide R., Bakker D., De Looff M., De Jong J., Van Manen N., Gaasenbeck C.P.H., Borgsteede F.H.M. (2005). Attempt to detect evidence for tick-borne encephalitis virus in ticks and mammalian wildlife in the Netherlands. *Vector-Borne and Zoonotic Diseases 5(1),* 58-64.

Uit het verleden

VECHTENDE HONDEN UIT ELKAAR HALEN

Vechtende honden uit elkaar halen is niet zonder risico. Wat daarbij kan gebeuren verhaalt ons niemand minder dan Jan Frans Willems (1797 - 1846) in een ellenlang gedicht *De twee Honden*. De ene met de sprekende naam Rapax, een vreselijk beest *tuk op roof en moorden,* haalt het van *onzen Hylas bij alle buren om zijn vriendelijkheid vermaard*. Hylas kan nauwelijks meer doen dan ellendig janken. Maar zie, een *jongling*

Krijgt met Hylas mededoogen,
grijpt den overwonnen hond,
rukt hem los, ontslaekt zijne ooren.
Maer helaes! Ter zelver stond
wordt hij in den hand gebeten.
En ... 't was Hylas die hem wondt!

Uit: Van Duyse, P., *Dicht- en Toneelkundige Nalatenschap van J. F. Willems*, De Busscher, Gent, 1856, p. 28 – 32.

L. Devriese