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# Assessment of Change in Risk for Release of *Ehrlichia ruminantium* (Heartwater) into the Continental United States following Discontinuation of the Tropical Bont Tick (*Amblyomma variegatum*) Eradication Program on St. Croix, U.S. Virgin Islands

**Centers for Epidemiology and Animal Health**

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**Acronyms and Abbreviations**

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|---|---|
| *A.* | *Amblyomma* |
| APHIS | Animal and Plant Health Inspection Service |
| CEAH | Centers for Epidemiology and Animal Health |
| CITES | Convention on International Trade in Endangered Species |
| *E.* | *Ehrlichia* |
| NESOI | Not elsewhere specified or indicated |
| NAHPP | National Animal Health Policy and Programs |
| OIE | World Organization for Animal Health, Office International des Epizooties, Paris, France |
| Spp. | Species |
| TBT | Tropical bont tick |
| USDA | United States Department of Agriculture |
| USFWS | United States Fish and Wildlife Service |
| USGS | United States Geological Survey |
| USVIDOA | United States Virgin Islands Department of Agriculture |
| VS | Veterinary Services |

## Executive Summary

The United States Department of Agriculture (USDA) Animal and Plant Health Inspection Service (APHIS) Veterinary Services (VS) National Animal Health Policy and Programs requested VS' Centers for Epidemiology and Animal Health (CEAH) to assess the change in risk associated with release (introduction) of *Ehrlichia ruminantium* into ruminant livestock and wildlife in the continental United States, if the current *Amblyomma variegatum* (referred to as tropical bont tick or TBT) eradication effort is discontinued on St. Croix, U.S. Virgin Islands. The risk that other Caribbean island countries contribute to the release of *E. ruminantium* via the TBT to the continental United States was not assessed; however, it is of note that other Caribbean islands are known to be reservoirs of infection and *A. variegatum* remains in several St. Croix herds. Consequences of the disease remain the same whether from St. Croix or elsewhere. Thus, change in risk may be defined in terms of the increase in likelihood of heartwater occurring in the mainland United States.

The hazard identified in this release assessment is the release of *E. ruminantium*, the causal agent of the disease known as heartwater, into domestic ruminant livestock and ruminant wildlife in the continental United States. *Ehrlichia ruminantium* is a gram-negative intracellular rickettsia. Ticks in the genus *Amblyomma* are only important to the identified hazard to the extent that these arthropods serve as the vector capable of transmitting *E. ruminantium*. As a vector, *Amblyomma* spp. ticks are required for transmission of these organisms from an infected ruminant to an uninfected, but susceptible ruminant in order to sustain a disease outbreak. The TBT, *A. variegatum*, is the most important and only tick in the Western Hemisphere that has been associated as a vector for *E. ruminantium* in outbreaks, and is the focus of this release assessment as it relates to releasing this rickettsia into the continental United States.

A release assessment describes the pathway(s) necessary for an activity to introduce a pathogenic agent into a particular environment and estimates the probability of that occurring in quantitative or qualitative terms. Five pathways were identified and subsequently evaluated (using government and public data, expert opinion, and literature review) for their feasibility to release *E. ruminantium* into the continental United States. A qualitative assessment was also made using terms such as 'minimal,' 'low,' or 'high,' to describe the likelihood of <u>increasing</u> the risk of an adverse event within the context of each release pathway due to the proposed discontinuation of the TBT eradication program on St. Croix. The term 'minimal' assumes that an increase in risk is possible but would be difficult or impossible to detect or demonstrate. A summary of the pathways follows with detailed facts and figures in the remainder of the document.

### Release Pathway 1: Importation of *E. ruminantium*-infected animal species

Legal interstate movement of domestic ruminants from St. Croix is a feasible pathway for release of *E. ruminantium* into domestic ruminant livestock and ruminant wildlife in the continental United States. However, historically, only a small number of animals have been shipped to a small number of States. In contrast, based on USDA data, legal interstate meovement of ruminant wildlife species from St. Croix currently does not appear to be occurring. Likewise, there is no evidence of illegal trade of domestic and wild ruminants originating from St. Croix.

If the TBT eradication program is eliminated on St. Croix, the change in risk of release is minimal for *E. ruminantium* and its associated vector tick (*A. variegatum*) into the continental United States from domestic ruminants or ruminant wildlife species originating from St. Croix provided that certain mitigations are applied prior to entry. Currently, as per Veterinary Services Memorandum No. 556.1, animals (including domestic ruminant livestock) infested with exotic ticks (including *A. variegatum*), animals exposed to such infestations, and animals moving interstate from areas placed under Federal quarantine must meet the requirements of part 71 and part 72, Title 9, Code of Federal Regulations for interstate movement purposes. These requirements include treatment of animals with an acaricide, documentation of treatment, physical inspection of animals for presence of ticks, issuance of a permit for interstate movement, and issuance of a health certificate or certificate of veterinary inspection. In addition to acaricide treatment, animal health officials in any State considering receiving St. Croix-origin ruminants should require additional mitigations to ensure that animals are not infected with *E. ruminantium*. Since no Federal regulations currently address mitigations for States to adopt regarding identification of *E. ruminantium*-infected animals prior to interstate movement, the World Organization for Animal Health (OIE) provides reasonable guidelines on this subject for adoption:

- Veterinary attestation that animals showed no clinical signs of heartwater on the day of shipment; and
- Animals are subjected to a diagnostic test (of high sensitivity) for heartwater (*E. ruminantium*) with negative results during the 15 days prior to shipment.

**Release Pathway 2: Mechanical transport of *E. ruminantium*-infected *A. variegatum* ticks by migrating cattle egrets**

The migration of cattle egrets from the Caribbean to the continental United States, some of which may be infested with *E. ruminantium*-infected *A. variegatum* ticks, is a feasible pathway for the release of *E. ruminantium* into susceptible domestic ruminant livestock and ruminant wildlife in the continental United States. However, cattle egrets will continue to have a presence on and around St. Croix and their ability to transport these ticks mechanically is unchanged even if the TBT eradication program in its present form is discontinued. The change in risk from egret migration is minimal because there is a reservoir of infection and ticks in other Caribbean locations that contribute to the population of affected egrets in St. Croix and travel to the continental United States with or without the current eradication program.

**Release Pathway 3: Mechanical transport of *E. ruminantium*-infected *A. variegatum* ticks by humans and imported animals, reptiles, and other birds**

Although a feasible pathway, the risk of introducing *E. ruminantium* and its tick vector into the continental United States is independent of whether or not the TBT eradication program is terminated in St. Croix, inasmuch as airline and cruise ship passenger traffic, as well as that of reptiles, birds (other than poultry), poultry, primates, horses, NESOI animals, and other mammals, originates from islands other than St. Croix on a recurring basis.

**Release Pathway 4: Mechanical transport of *E. ruminantium*-infected ticks by fomites**

Although a feasible pathway, the risk for release of these ticks on the U.S. mainland is minimal, provided mitigations are followed per Federal regulations. These regulations require treating/destroying litter and manure from carriers and premises of tick-infested animals and vehicles. The change in risk of humans inadvertently releasing these ticks into the continental United States is independent of whether or not the TBT eradication program is discontinued in St. Croix inasmuch as human traffic in volume from Caribbean countries other than St. Croix is taking place on a recurring basis.

**Release Pathway 5: Smuggling of live *E. ruminantium***

Although this is a feasible pathway, the risk of introducing *E. ruminantium* into the continental United States is independent of whether or not the TBT eradication program is terminated in St. Croix, inasmuch as stocks of this agent are not known to be stored on St. Croix and sources of infected animals are equally available on other islands.

The continental United States is vulnerable to the release of heartwater from at least five different pathways. Discontinuing the TBT eradication program in its current form (i.e., focused on eradicating the *A. variegatum* ticks on livestock premises) has the potential to increase the risk of releasing *E. ruminantium* onto the U.S. mainland through pathways involving uncontrolled trade in ruminant animals. However, if the aforementioned mitigations are followed, this risk (above that currently present) is minimal. The other pathways for release of *E. ruminantium* onto the U.S. mainland are independent of whether or not the TBT eradication program on St. Croix is discontinued and represent minimal change in risk.

Due to the reservoir of infection in neighboring islands and the potential for transport of infected ticks between islands, it is unlikely that a localized eradication program can succeed. Alternatives would include a broader eradication/control plan encompassing the majority of the Caribbean basin or intensive management and parasite control on St. Croix animals on the part of stock owners. The former would undoubtedly be expensive and fraught with political challenges between the various governments. The latter would seem to be a more reasonable and effective solution.

## Section 1: Introduction

The U.S. Virgin Islands Department of Agriculture (USVIDOA) began an aggressive eradication effort shortly after the TBT was found on St. Croix in 1967. The effort was successful in eradicating *A. variegatum* ticks from the island (54,000 acres; 84 square miles) in 1972. However, in 1987, the tick returned to the island. By September 2003, the TBT had infested 8 farms (Gilsdorf et al. 2006). In an attempt to eradicate the TBT for a second time from this island, VS entered into a cooperative agreement with USVIDOA beginning in 2004 and running through December 2007. As of July 2006, 11 premises in the western one-third (affected zone) of St. Croix and one premises in the buffer zone were under quarantine—a metric demonstrating little success of the program.

The TBT is a vector of *Ehrlichia ruminantium*, the causal agent of the disease known as heartwater in ruminant livestock and wildlife. This disease has not been diagnosed in St. Croix, but the presence of the tick vector on the island restricts interstate and international trade of heartwater-susceptible ruminant livestock, particularly Senepol cattle and St. Croix white hairless sheep.

VS NAHPP requested CEAH to assess the change in risk associated with release (introduction) of *E. ruminantium* into ruminant livestock and wildlife in the continental United States, if the TBT eradication effort is discontinued on St. Croix.

The risk that other Caribbean island countries contribute to the release of *E. ruminantium* via the TBT into the continental United States was not assessed. However, it is noted that other Caribbean islands are known to be reservoirs of infection for *E. ruminantium* and *A. variegatum* ticks.

## Section 2: Hazard Identification

The hazard identified in this release assessment is the release of *E. ruminantium*, the causal agent of the disease known as heartwater, into domestic ruminant livestock and ruminant wildlife in the continental United States. *Ehrlichia ruminantium* is a gram-negative intracellular rickettsia (Cowdry 1925, Dardiri et al. 1987, Camus et al. 1996, Deem 1998, Peter et al. 1998). Ticks in the genus *Amblyomma* are only important to the identified hazard to the extent that these arthropods serve as the only known vector capable of transmitting *E. ruminantium* (Camus et al. 1996). As vectors, *Amblyomma* spp. ticks are required for transmission of these rickettsial organisms from an infected ruminant to an uninfected but susceptible ruminant in order to sustain a disease outbreak. The TBT, *A. variegatum*, the most important tick vector for *E. ruminantium*, is the focus of this release assessment as it relates to releasing this rickettsia into the continental United States.

## Section 3: Release Assessment

A release assessment describes the pathway(s) necessary for an import activity (e.g., live animals or their products) to introduce a pathogenic agent (the hazard) into a particular environment and

estimates the probability of that occurring (World Organization for Animal Health (OIE) Web site 2007a).

### *3.1. Host, Agent, and Environmental Interactions Important to Pathways for Release of E. ruminantium into Domestic and Wild Ruminants in the United States*

#### 3.1.1. Arthropod vectors and disease agent interactions

Only ticks of the genus *Amblyomma* are presently known to be capable of transmitting *E. ruminantium*, either naturally or experimentally (Mackenzie et al. 1980, Norval et al. 1981, Uilenberg 1982, Barré et al. 1987, Walker 1987, Camus et al. 1996, Mahan et al. 2000, Peter et al. 2000, Wesonga et al. 2001). Natural tick vectors are those *Amblyomma* spp. ticks that have transmitted *E. ruminantium* under field conditions to susceptible animal species and subsequently caused clinical signs of disease in these animals (Mahan et al. 2000). Five species of African-origin *Amblyomma* ticks (*A. variegatum, A. hebraeum, A. lepidum, A. astrion,* and *A. pomposum*) are considered natural vectors and have been implicated in field outbreaks of heartwater (Walker 1987). Seven other species of *Amblyomma* ticks (*A. cohaerens, A. gemma, A. tholloni, A. sparsum, A. marmoreum, A. maculatum* and *A. cajennense*) are capable of transmitting heartwater experimentally. None have been implicated in field outbreaks of heartwater (Walker 1987).

This release assessment is focused solely on *A. variegatum* ticks. The life cycle of these ticks may take from 5 months to 4 years to complete (Petney et al. 1987, Maré 1998). Regarding the Caribbean islands, the maximum longevity of *A. variegatum* in Guadeloupe is 11 months for larvae, 15 months for nymphs, and 20 months for adults (Camus et al. 1996, Corn et al. 1996). Because larvae or nymphs may become infected and then transmit *E. ruminantium* in their next stages of development, infection can endure within a tick for a very long time (Barré et al. 1987, Maré 1998). In fact, due to the hardiness and longevity of *Amblyomma* spp. ticks, *E. ruminantium* has been shown to persist in them for up to 15 months (Ilemobade 1976).

*Amblyomma* spp. ticks are classified as three-host ticks. They feed on a wide variety of livestock, wild ungulates, ground birds, small mammals, and reptiles (Horak et al. 1987, Oberem et al. 1987, Petney et al. 1987, Camus et al. 1996, Maré 1998, Peter et al. 2002). The life cycle of *Amblyomma* spp. ticks increases the probability that these arthropods survive a journey of several weeks of transit. Their survival and development from larvae to nymph to adult stages require attaching to a living host (Barré et al. 1987, Horak et al. 1987, Petney et al. 1987, Camus et al. 1996, Maré 1998). However, the molting period, which is spent off the host, can range from 11 to 31 days for nymphal premolt (larva molts to nymph) and 15 to 35 days for adult premolt (nymph molts to adult), depending on the species of *Amblyomma* tick (Petney et al. 1987). The length of the molting period is sufficient, in most cases, to allow the survival of a tick off the host and on fomites during transport to the United States—a period of hours or days in the case of air travel and two to three weeks if traveling by oceangoing vessel, depending upon the origin of a shipment.

*Amblyomma variegatum* is considered to be the most important vector of *E. ruminantium* (Cumming 1998). Because *E. ruminantium* cannot persist outside a host for more than a few

hours, the principal mode of releasing *E. ruminantium* into a new geographic location is via infected ticks or carrier animals (Maré 1998). Consequently, the host range of these ticks parallels the host range of *E. ruminantium*. Trans-stadial transmission[a] of *E. ruminantium* is the primary means by which *A. variegatum* ticks transmit heartwater (Camus et al. 1987). All immature stages of *A. variegatum* infest cattle, sheep, and goats (Petney et al. 1987, Cumming 1998).

Infection rates of *A. variegatum* ticks vary widely. According to a study performed in Guadeloupe, only 1–4 percent of adult *A. variegatum* ticks were infected (Camus et al. 1987). Another study demonstrated higher infection (0–91 percent) rates (Mahan et al. 1995). Further research on nymphs of *A. variegatum* yielded experimental infection rates of 43.5 percent (Mahan et al. 2000).

In heartwater-endemic areas, subclinical carriers are more abundant than clinically reacting hosts (Andrew et al. 1989). In these animals, it appears that *E. ruminantium* organisms reproduce in capillary cells and are intermittently released into the bloodstream; thus, ticks can only acquire the infection at certain times. Consequently, the level of rickettsemia in carriers is generally lower than in clinically reacting animals. As a result, ticks feeding on carriers have lower infection rates (Deem et al. 1996, Peter et al. 1999).

3.1.2. Host interactions

3.1.2.1. Animal species susceptible to *E. ruminantium* infection

Domestic ruminants and ruminant wildlife are target species in an epizootic of heartwater. This disease has also been observed clinically in non-ruminant species such as the domestic ferret, the four-striped grass mouse, and the southern multi-mammate mouse. Although the latter two rodent species are susceptible to infection with *E. ruminantium*, they are not hosts of the ticks and are not believed to play a role in the epizootiology of heartwater (Petney et al. 1987, Maré 1998, Peter et al. 2002).

3.1.2.2. Species that are resistant to *E. ruminantium* infection but serve as transport hosts of *A. variegatum*

3.1.2.2.1. Reptiles

Of the known *Amblyomma* spp. that are capable of transmitting *E. ruminantium*, either naturally or experimentally, only some have immature stages that infest reptiles. *Amblyomma variegatum* nymphs and larvae have been found to infest varanid lizards, vipers, and chameleons (Cumming 1998).

3.1.2.2.2. Ground-dwelling birds

3.1.2.2.2.1. Ostriches

[a] Trans-stadial transmission means passage of a disease agent from an infected immature stage to the next immature stage or to the adult stage of an arthropod vector (Public Health and Medical Entomology 2007).

Ostriches (*Struthio camelus*) are common hosts for several *Amblyomma* spp. ticks, including *A. variegatum* (Mertins et al. 1991).

3.1.2.2.2.2. Other birds

Ground-dwelling birds such as francolin (*Francolinus* genus), quail (Phasianidae family), and hornbills (Bucerotidae family) have been infested with several different *Amblyomma* spp., including *A. variegatum* (Petney et al. 1987, Wesonga et al. 2001, Cumming 1998).

3.1.2.2.3. Migratory birds

3.1.2.2.3.1. Cattle egrets (*Bubulcus ibis*)

The cattle egret (*Bubulcus ibis*) is native to Africa, the western Mediterranean, humid Asian tropics from India to Japan, and Northern Australia. Cattle egrets colonized South America some time in the later half of the 19$^{th}$ century with initial sightings in British Guiana and Suriname in 1877. This bird species continued to expand its range and was established in all of South America by the 1970s. Cattle egrets initially became established in the eastern Caribbean with reports as early as 1951 (Arendt 1988) and then, during this same decade, expanded northward into Florida and the coastal United States; however, sightings of cattle egrets are reported in Florida as early as 1941 (Arendt, 1988, Corn et al. 1993, Telfair 2006). An estimated 750,000 to 1.5 million birds inhabit North America (U.S. Fish and Wildlife Service Web site 2006a). Figure 1 shows the known breeding range of cattle egrets in the Americas based on data from Nature Serve (Ridgely et al. 2003).

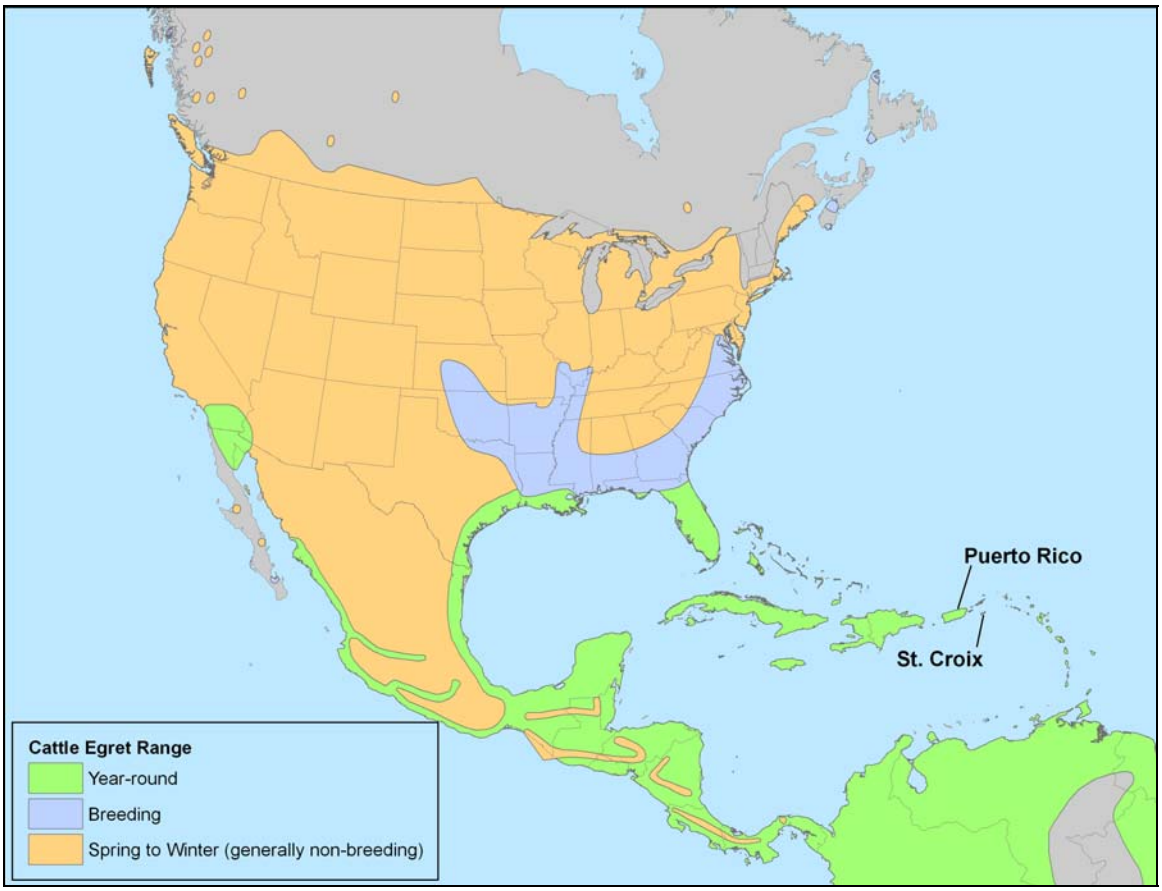


Figure 1. Known breeding range of cattle egrets (*Bubulcus ibis*) in the Americas.

Breeding cattle egret colonies occur in every State in the continental United States except Montana, New Hampshire, Washington, and West Virginia (Telfair 2006). However, these birds continue to colonize new areas, primarily in response to clearing of native grassland vegetation, which is often used for cattle grazing. Observations of nesting and young in Florida indicate that these birds may breed year-round in south Florida (Anderson 2003). In Canada, the birds breed in Ontario and Saskatchewan (Telfair 2006).

Cattle egrets use three distinct spring and fall migration corridors in North America: (1) an eastern route from Newfoundland along the Atlantic Coast States to Florida and Mississippi, then through the Greater Caribbean Basin, lower portion of Mexico, and south through Central America and into Colombia; (2) a central route from Kansas east to Alabama then south to Panama; and (3) a Pacific Coast route from southern Arkansas to west-central Mexico (Telfair 2006). However, distinguishing between migration and dispersal is difficult in this species, as individuals tend to wander extensively, exhibiting repeated incursions, temporary colonization, retreats, and establishments, depending on environmental conditions (Hancock et al. 1984). Juveniles are known to rapidly disperse great distances from natal colonies, up to 5,000 km from their birthplace (Van den Bosch et al. 1992). Banding records from the U.S. Geological Survey (USGS) Bird Banding Laboratory have documented dispersals greater than 3,100 km in North

American populations (USGS Web site 2007). McKilligan et al. (1993) reported tagged birds moving 2,500 km in New Zealand.

Cattle egrets' feeding biology is adapted almost exclusively to ungulate hosts. Egrets generally forage in close association with grazing cattle or other livestock, but may glean elsewhere, especially around margins of aquatic areas and fields where fire, tractors, or cutting/mowing machinery are used and disturb prey (Grubb 1976, Thompson et al. 1982, Heatwole 1965, Dinsmore 1973). Cattle egrets are often seen perched or riding on the backs of host animals and may provide a mechanical transport of ticks from host to host. Egrets often synchronize resting and feeding periods with those of cattle and in some cases attempt to stir resting cattle by making restless flights near them (Singh et al. 1988, Dawn 1959). These birds are voracious, active foragers that usually feed together in loose aggregations of small to large flocks (10–200 individuals), but may forage in smaller groups or individually (Mora et al. 1998). Cattle egrets have a highly varied diet depending on conditions; however, major prey includes active insects disturbed by grazing of host animals, primarily grasshoppers and crickets, and host-associative flies (Guillén et al. 1994).

Due to cattle egrets' association with pastureland habitat and cattle in the Caribbean islands, these birds are susceptible to infestation by *A. variegatum* ticks (Arendt 1988, Corn et al. 1993, Telfair 2006). Cattle egrets are believed to be partly responsible for the dissemination of *A. variegatum* ticks between countries in the Caribbean region (Pegram et al. 1996, Pegram et al. 2004). The feeding periods for *A. variegatum* larvae and nymphs on cattle egrets are 6–13 days and 5–10 days, respectively. Thus, these immature stages could be expected to survive for several days during the dispersal or migration of cattle egrets (Garris 1984). To date adult *A. variegatum* ticks have not been found on cattle egrets although infestation with larval and nymph stages occurred (Barré et al. 1988, Southeastern Cooperative Wildlife Disease Study Web site 2006, Corn et al. 1993). However, these studies have been limited to small localized surveys of cattle egrets on a few Caribbean islands. More extensive surveys of cattle egrets throughout the range of *A. variegatum* in the Caribbean basin are needed.

3.1.2.2.3.2. Passerine and near passerine birds

Small passerine birds (larks, canaries, and thrushes of the Passeriformes order) and near passerine birds (doves, pigeons of the Columbidae family) have been reported to carry *Amblyomma* spp. ticks, including *A. variegatum* (Cumming 1998, Burridge 2001). As a result, they may serve to disseminate *Amblyomma* ticks, some of which may be infected with *E. ruminantium*, during flight from one area to another.

3.1.2.2.4. Humans

Although humans are believed to be resistant to *E. ruminantium* infection (Maré 1998), they can act as transport hosts of *Amblyomma* spp. ticks, including *A. variegatum* (Burridge et al. 2002, Estrada-Peña et al. 1999).

3.1.3. Environmental interactions

3.1.3.1. Heartwater-endemic Caribbean island countries

The first isolate of *E. ruminantium* from ruminant livestock in the Caribbean occurred in 1980 from a goat living on the French Antilles island of Guadeloupe (Perreau et al. 1980) and then on the nearby island of Marie-Galante (Uilenberg et al. 1984). This rickettsial agent has also been isolated from *A. variegatum* ticks inhabiting Antigua (Birnie et al. 1985). In addition, antibodies to *E. ruminantium*, indicating exposure to this rickettsial organism, have been found in cattle inhabiting Dominica, Grenada, Martinique, Montserrat, St. Kitts, St. Lucia, St. Martin/St. Maarten, and St. Vincent (Kobold et al. 1992).

According to the latest OIE information, Guadeloupe was the only member country from the Caribbean islands that reported either clinical cases or laboratory evidence of heartwater (e.g., serum antibody, isolation of rickettsia) in susceptible species (World Organization for Animal Health (OIE) Web site 2007b). St. Kitts and Nevis were suspected to have heartwater but its presence was not confirmed. The OIE did not list several other countries in the Caribbean Island chain, namely Antigua and Marie-Galante, known to have had heartwater in prior years. Consequently, their heartwater status is unknown, although they are assumed to still be infected with the disease.

As of December 2007, susceptible ruminant livestock species inhabiting St. Croix have not exhibited clinical signs of heartwater nor been shown to have laboratory evidence of heartwater (e.g., serum antibody, isolation of the rickettsia).

3.1.3.2. Social and political factors

The Centers for Disease Control and Prevention has not classified *E. ruminantium* as a human select agent (U.S. National Archives and Records Administration Web site 2007a). In contrast, this rickettsia is listed as a select agent of concern for livestock (U.S. National Archives and Records Administration Web site 2007b).

*3.2. Pathways for Release of Ehrlichia ruminantium into the United States*

Considering the aforementioned agent, host, and environmental interactions, several potential pathways exist in which susceptible domestic ruminant livestock and ruminant wildlife in the continental United States can be exposed to *E. ruminantium*. These pathways are discussed in greater detail below.

3.2.1. Pathway 1: Importation of *E. ruminantium*-infected animal species

3.2.1.1. Background information

Legal and illegal importation of domestic ruminant livestock, ruminant wildlife, or other animals infected with *E. ruminantium* is a potential source of the disease agent for susceptible domestic ruminant livestock and ruminant wildlife species in the continental United States. In 2006, a total of 2,878,687 head of domestic ruminant livestock inhabited the Caribbean islands (FAOSTAT, Food and Agriculture Organization of the United Nations Web site 2007). The U.S.

Virgin Islands, including St. Croix, contributed a very small number of ruminant livestock (5,150 head; 0.18 percent) toward this total. More specific to St. Croix, as of June 30, 2007, 1,808 head of cattle, 3,965 head of sheep, and 3,814 head of goats were present on the island.[b]

The volume of illegal trade in ruminant wildlife is difficult to estimate. By its very nature, illegal trade is undocumented with the exception of information available for seized shipments that is sometimes reported in the media and/or Convention on International Trade in Endangered Species (CITES) trade data. Whereas live birds, ornamental fish, reptiles, and amphibians are commonly traded, live wild ruminants are not (Roe et al. 2002). This may be due, in part, to the size of the animal (not easy to conceal) and also the demand for the animals.

To date, St. Croix has not reported evidence of heartwater in domestic ruminant livestock.

3.2.1.2. Analysis of data

Although the time frame analyzed for this release assessment is for 2000-2006, records of livestock shipments from St. Croix to the continental United States have been acquired only up through 2004.[b] In 2002, 24 head of Senepol cattle were shipped to Texas. In 2003, 40 and 52 head of Senepol cattle were shipped to Florida and Louisiana (through Florida), respectively. Thirty-nine head of Senepol cattle were shipped to Florida in 2004. In 2004, 4 sheep were also shipped to Virginia (through Pennsylvania). No ruminant wildlife species that are susceptible to heartwater (*E. ruminantium*) were exported to the continental United States from St. Croix.

USFWS wildlife inspectors stationed at 32 ports and border crossings processed more than 123,000, 136,000, and 155,000 wildlife shipments for the years 2002–2004, respectively (U.S. Fish and Wildlife Service Web site 2006b, U.S. Fish and Wildlife Service Web site 2006c, U.S. Fish and Wildlife Service Web site. 2006d). No illegal shipments of wild ruminants were seized from any Caribbean countries, including St. Croix.

3.2.1.3. Conclusions

Legal interstate movement of domestic ruminants from St. Croix is a feasible pathway for release of *E. ruminantium* into domestic ruminant livestock and ruminant wildlife in the continental United States, given that, historically, these animals have been shipped to a few States. In contrast, legal interstate movement of ruminant wildlife species from St. Croix does not appear to be occurring. Likewise, illegal trade of domestic and wild ruminants originating from St. Croix does not appear to be occurring.

If the TBT eradication program is eliminated on St. Croix, the change in risk of release is minimal for *E. ruminantium* and its associated vector tick (*A. variegatum*) into the continental United States from domestic ruminants or ruminant wildlife species originating from St. Croix provided that certain mitigations are applied prior to entry. Currently, as per Veterinary Services Memorandum No. 556.1 (USDA:APHIS Web site 2007), animals (including domestic ruminant livestock) infested with exotic ticks (including *A. variegatum*) animals exposed to such

[b] Office of USDA-APHIS-VS Area Veterinarian In Charge - Puerto Rico and USVI, Hato Rey, PR. 2007. Personal communication

infestations, and animals moving interstate from areas placed under Federal quarantine must meet the requirements of part 71 (U.S. National Archives and Records Administration Web site 2007c) and part 72 (U.S. National Archives and Records Administration Web site. 2007d), Title 9, Code of Federal Regulations for interstate movement purposes. These requirements include treatment of animals with an acaricide, documentation of treatment, physical inspection of animals for presence of ticks, issuance of a permit for interstate movement, and issuance of a health certificate or certificate of veterinary inspection. In addition to acaricide treatment, animal health officials in any State considering receiving St. Croix-origin ruminants should require additional mitigations to ensure that animals are not infected with *E. ruminantium*. Since no Federal regulations currently address mitigations for States to adopt regarding identification of *E. ruminantium*-infected animals prior to interstate movement, the World Organization for Animal Health (OIE) provides reasonable guidelines on this subject for adoption (World Organization for Animal Health (OIE) web site 2007c):

- Veterinary attestation that animals showed no clinical signs of heartwater on the day of shipment; and
- Animals are subjected to a diagnostic test (of high sensitivity) for heartwater (*E. ruminantium*) with negative results during the 15 days prior to shipment.

To some observers, these mitigations may appear costly. However, given the small numbers of domestic ruminant livestock on St. Croix and even smaller numbers historically exported to the continental United States, the cost of these mitigations on a per-head basis for any future exported animals is substantially less than the current cost of the TBT eradication program and still allows for trade to occur.

3.2.2. Pathway 2: Mechanical transport of *E. ruminantium*-infected *A. variegatum* ticks by migrating cattle egrets (*Bubulcus ibis*)

3.2.2.1. Background information

A census of the number of cattle egrets living on St. Croix or other islands has not been done and would be extremely challenging logistically. Similarly, the precise number of cattle egrets with established migration routes to and from the U.S. mainland and islands in the Caribbean is not known. However, a portion of the cattle egret population in Florida reportedly leaves the State during the winter, migrating to various Caribbean islands before returning to the State in the spring (Florida Fish and Wildlife Conservation Commission Web site 2006). In addition, the migration of cattle egrets from other Gulf Coast States and the eastern seaboard of the United States to Central America, South America, and the Caribbean has been documented by the bird banding record, but the magnitude (number of individuals) of this migration has not been established.

*Amblyomma variegatum* larvae and nymphs do infest cattle egrets (Barré et al. 1988, Southeastern Cooperative Wildlife Disease Study Web site 2006, Corn et al. 1993). Circumstantial evidence strongly links the increase in populations of the cattle egret with increased colonization of new islands by *A. variegatum* ticks. These ticks have infested as many

as 16 of the 27 islands of the Lesser Antilles (Alderink et al. 1988, Pegram et al. 1996, Deem 1998, Bram et al. 2002).

3.2.2.2. Analysis of data

Bird band[c] recovery data are available at a continental scale and are distributed by the U.S. Department of the Interior USGS Patuxent Wildlife Research Center Bird Banding Laboratory, Laurel, MD. Bird banding is a mark-recapture technique for studying the movement, survival, and behavior of birds. This technique documents both the origin of each banded bird and its recovery location. Recovery of bands is reported in 10-minute blocks of degrees longitude and latitude, which is approximately 100 square miles. Cattle egrets are generally banded while at their breeding colonies and then recovered in most cases as a result of a mortality event (i.e., shot, found dead, killed by farm machinery, etc). Cattle egrets are not hunted in the Americas by sportsmen resulting in relatively low recovery rates.

For the purpose of this analysis, the entire banding record for cattle egrets was used. Between 1956 and 2002, 64,608 cattle egrets were banded in North America, Central America, and the Caribbean. Only 599 cattle egrets were recovered resulting in a recovery rate of 0.93 percent. Much of the banding effort (35 percent) over this time span was performed in central Texas. The data were constrained to only cattle egrets banded along the Gulf Coast, the southeastern seaboard, and Caribbean islands, resulting in 396 recoveries. The data were constrained to better approximate the movement dynamics of egret dispersal along the Caribbean. Over one-half (210) of the recovered egrets traveled greater then 600 km (360 mi) from their banding location with the greatest movement being 3,143 km (1,886 mi). Figure 2 shows a histogram representing the percentage of cattle egrets recovered by distance traveled.

[c] The North American Bird Banding Program is jointly administered by the U.S. Department of Interior and the Canadian Wildlife Service. Their respective banding offices have similar functions and policies and use the same bands, reporting forms, and data formats.

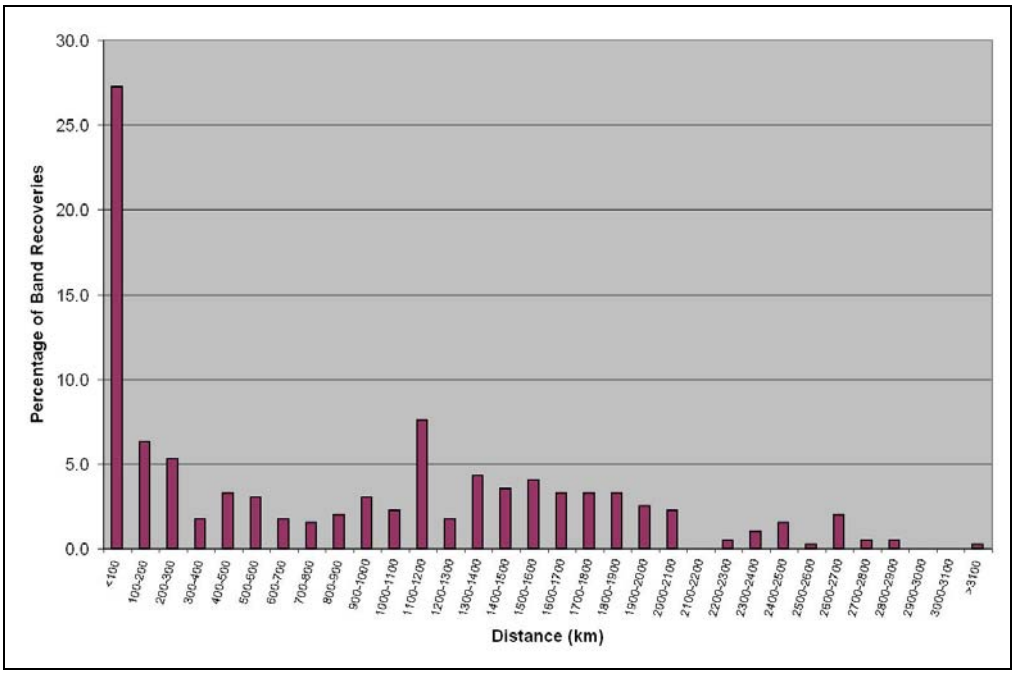


Figure 2. Cattle egret band recoveries by distance (n = 396).

Data on movement distances for banded cattle egrets were used to assign movement probabilities to 100 km (60 mi) distance categories from 0 to 3,200 km (0 to 1,920 mi). Using standard statistical software, the constrained data for cattle egrets (n = 396) were used to assign movement probabilities by fitting several density functions to those data. A Weibull probability function was selected as the best fit to the true distribution of movement probabilities for cattle egrets. Figure 3 shows a histogram of movement distances taken from the band recovery data and the associated Weibull probability of movement distances. From these data it is clear that few limitations exist on potential movement distances. Approximately 53 percent of the probability mass encompasses movement distances greater than 600 km (360 mi).

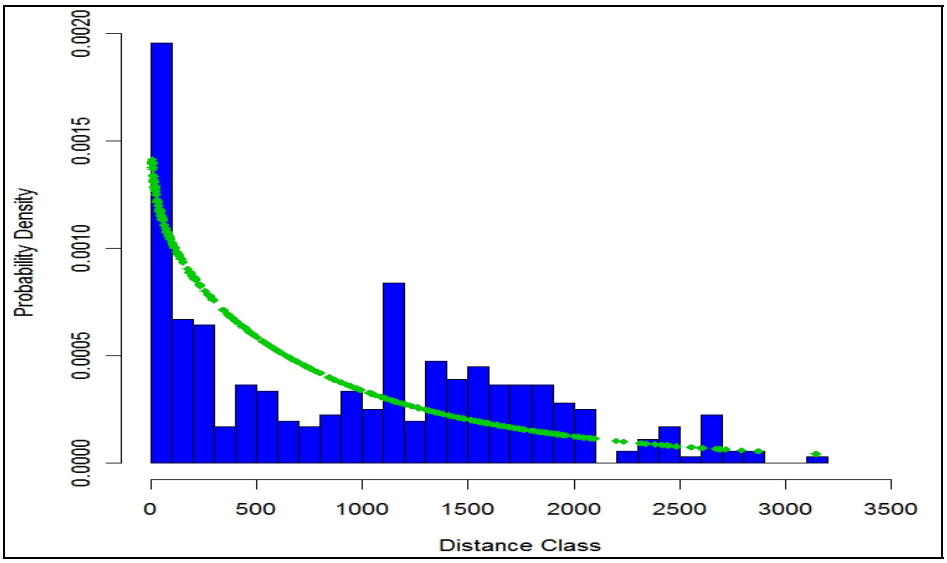


Figure 3. Cattle egret dispersal distance (in km) probabilities.

These resulting data on potential cattle egret movement were compared with the distance between islands in the Caribbean to determine islands that are within the potential dispersal range of cattle egrets. Table 1 presents the linear distance between major islands in the eastern Caribbean. It is important to note that all islands in the Caribbean basin and Florida are within the dispersal range of cattle egrets.

Table 1. Distance (km) between selected Caribbean island countries.

| | **Antigua[a,b]** | **Guadeloupe[a,b]** | **Saint Croix[b]** | **Vieques[c]** | **Puerto Rico[c]** | **Florida** |
|---|---|---|---|---|---|---|
| **Antigua[a,b]** | - | 60 | 300 | 382 | 416 | 2,100 |
| **Guadeloupe[a,b]** | 60 | - | 339 | 427 | 466 | 2,150 |
| **Saint Croix[b]** | 300 | 339 | - | 57 | 91 | 1,800 |
| **Vieques[c]** | 382 | 427 | 57 | - | 12 | 1,700 |
| **Puerto Rico[c]** | 416 | 466 | 91 | 12 | - | 1,540 |
| **Florida** | 2,100 | 2,150 | 1,800 | 1,700 | 1,540 | - |

[a]Islands with persistent *Ehrlichia ruminantium* infection.
[b] Islands with current infestations of *Amblyomma variegatum.*
[c] Islands with historical infestations of *Amblyomma variegatum.*

In addition, the potential movement of cattle egrets was mapped for Antigua and Guadeloupe, which currently have *E. ruminantium* circulating within *A. variegatum* tick populations. Figure 4 shows this potential dispersal of cattle egrets from these two islands. Note that these birds have the potential to reach the continental United States, bringing *A. variegatum* ticks that may be infected with *E. ruminantium*.

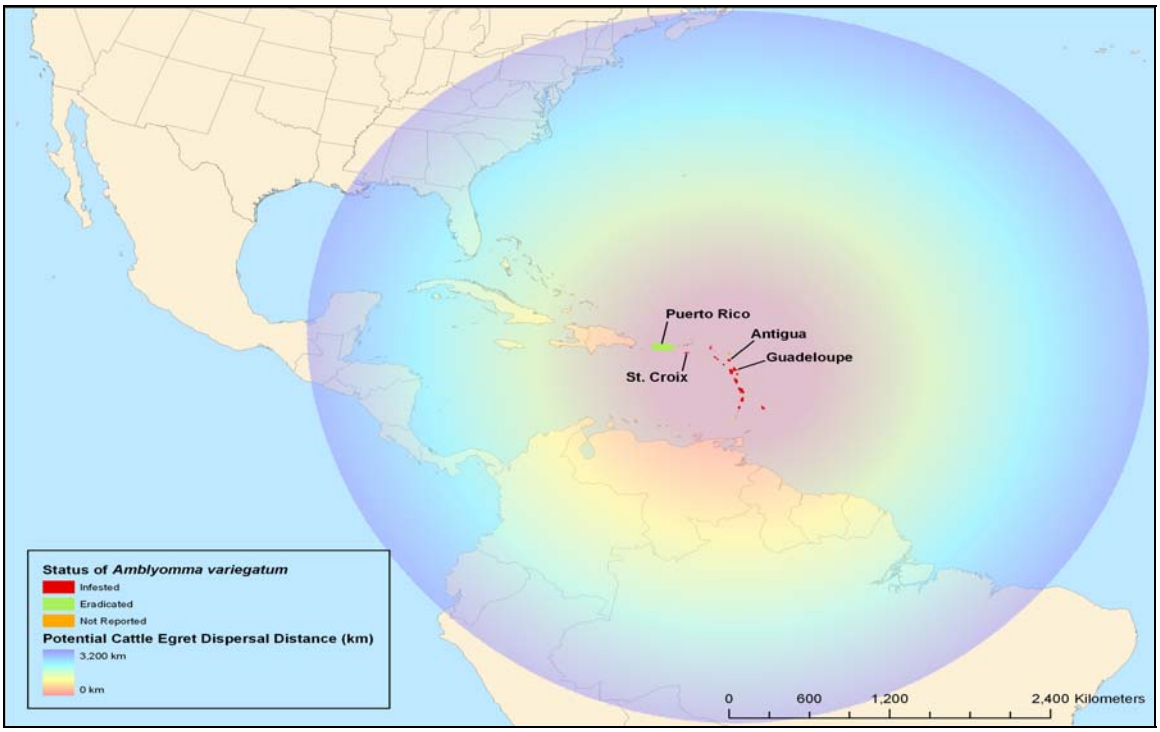


Figure 4. Potential dispersal distances of cattle egrets from Antigua and Guadeloupe.

This analysis provides a good foundation for understanding potential cattle egret movements. However, it is important to note that the movement of cattle egrets in the Caribbean basin is largely unknown and may differ from movement of cattle egrets along the Gulf Coast or the eastern seaboard of the continental United States. Cattle egrets have been documented to move from Guadeloupe to Florida (Corn et al. 1993). Cattle egrets have also been reported to frequently move between the Caribbean basin and Florida (Browder 1973). However these movements, both locally and long range, require further investigation to fully understand the frequency with which cattle egrets move from the Caribbean basin to the continental United States. A study of cattle egret movements from St. Croix to the continental United States has not been performed.

3.2.2.3. Conclusions

The migration of cattle egrets from the Caribbean to the continental United States, some of which may be infested with *E. ruminantium*-infected *A. variegatum* ticks, is a feasible pathway for the release of *E. ruminantium* into susceptible domestic ruminant livestock and ruminant wildlife in the continental United States. However, cattle egrets will continue to have a presence on and around St. Croix and their ability to transport these ticks mechanically is unchanged even if the TBT eradication program in its present form is discontinued on this island. Although the exact number of egrets migrating from St. Croix is difficult to estimate, it is clear from the information above that St. Croix represents a very small portion of the total egret population that may migrate to the mainland of the United States. Further, the TBT program on St. Croix has little, if any, effect on the reservoir of egrets moving between islands. Thus, it is evident that the TBT program has minimal effect on this pathway.

3.2.3. Pathway 3: Mechanical transport of *E. ruminantium*-infected *A. variegatum* ticks by humans and imported animals, reptiles, and other birds

3.2.3.1. Background information

*Amblyomma variegatum* ticks have been imported inadvertently into the United States on a number of occasions over the past 35 years (Wilson et al. 1998). This tick species was typically found attached to mammals or birds. However, reptiles such as monitor lizards, chameleons, and species from the Viperidae family can be infested (Cumming 1998, Burridge 2001). *Amblyomma variegatum* ticks have been recovered from ostriches imported into the United States from Africa and Europe (Mertins et al. 1991). No mention of whether these ticks were tested for *E. ruminantium* was made.

3.2.3.2. Analysis of data

3.2.3.2.1. Humans

Tremendous numbers of people travel between countries in the Caribbean island and the continental United States each year. For example, 11,907,736 airline passengers traveled between 21 Caribbean countries and the continental United States in 2006.[d] People arriving from the U.S. Virgin Islands accounted for 3.37 percent (400,973) of the passengers.

Five major cruise lines service the Caribbean islands and have a U.S. port of call (U.S. Department of Transportation Maritime Administration Web site 2007). Together, in 2006, these five cruise lines conducted 1,328 cruises and carried 3,332,109 passengers. St. Croix is visited by some of these cruise lines. Although no data were found that summarized the number of arriving passengers that had ticks removed from their person, there is little evidence to support a conclusion that tick-infected cruise passengers are a major pathway of risk.

3.2.3.2.2. Animals, reptiles, and birds

Between 2000–2006, the United States imported 106,298 reptiles, 4 poultry, 35 birds (other than poultry), 1,087 primates, 358 horses, 57 other mammals, and 182 NESOI animals from 14 Caribbean countries.[e,f] St. Croix only exported a few horses to the continental United States. In 2002, one horse was shipped to Florida. Three horses were shipped to Washington (through Florida) in 2003 and again (one horse) in 2004.[b]

No data were found detailing interceptions of smuggled mammals, birds, and reptiles from St. Croix at ports.

3.2.3.3. Conclusions

[d] U.S. Department of Transportation, Bureau of Transportation Statistics database.
[e] U.S. Department of Commerce, Bureau of Census, World Trade Atlas.
[f] USDA:APHIS:VS:CEAH import tracking system.

Passengers aboard aircraft and cruise ships that travel between Caribbean countries, including St. Croix, and the continental United States is a feasible although unlikely pathway for introducing *A. variegatum* ticks onto the U.S. mainland. However, the risk of introducing *E. ruminantium* and the tick vector into the continental United States is independent of whether or not the TBT eradication program is terminated in St. Croix, inasmuch as passenger traffic originates from islands other than St. Croix on a recurring basis.

Reptiles, poultry birds (other than poultry), poultry, primates, horses, NESOI animals, and other mammals originating from the Caribbean islands are a feasible pathway for introducing *A. variegatum* ticks, some of which may be infected with *E. ruminantium*, into the continental United States. The risk of introducing *E. ruminantium* and its tick vector into the continental United States is independent of whether or not the TBT eradication program is terminated in St. Croix, inasmuch as this mammal, bird, and reptile traffic is taking place on a recurring basis in greater volume from Caribbean island countries other than St. Croix.

3.2.4. Pathway 4: Mechanical transport of *E. ruminantium*-infected *A. variegatum* ticks by fomites

3.2.4.1. Background information

Historically, *Amblyomma* spp. ticks have been found on fomites such as chamedor (palm) leaves (Becklund 1968) and other plant material imported into the United States. [g]

Through Veterinary Services Memorandum No. 556.1 (USDA:APHIS Web site 2007), 9 CFR part 72.24 ( U.S. National Archives and Records Administration Web site 2007d), which requires litter and manure to be treated and destroyed from carriers and premises of animals infested with *Boophilus* spp. ticks, will also apply to other exotic ticks, including *A. variegatum*. Cars or other vehicles having carried (*Boophilus* spp.) infested or exposed animals as well as premises exposed to these ticks shall also be cleaned and treated as described in 9 CFR part 72.23 ( U.S. National Archives and Records Administration Web site 2007d). Through Veterinary Services Memorandum No. 556.1, this regulation is extended to apply to other exotic ticks such as *A. variegatum*,

3.2.4.2. Analysis of data

During the time period 2000–2006, out of 17 interceptions of ticks associated with fomites, *Amblyomma* spp. ticks were found on only one occasion; however, this incident did not involve St. Croix.[h] No ticks were reported recovered from bedding, etc., associated with the shipments of livestock from St. Croix to the continental United States during the time period analyzed by this release assessment (see Pathway 1).

Analysis of data for Caribbean-origin airline passenger luggage and other personal items inspected for illegal plant and animal products did not yield any tick interceptions.[h] Inspectors (APHIS Plant Protection and Quarantine) at Miami and Fort Lauderdale ports of call in Florida

[g] Pest ID Database, USDA:APHIS:PPQ
[h] Work Accomplishment Data System (WADS), USDA:APHIS:PPQ

inspected the baggage of 1 percent of cruise ship passengers arriving from foreign locations (USDA Office of Inspector General Web site 2007). No tick interceptions were reported.

3.2.4.3. Conclusions

The importation of *A. variegatum* ticks on bedding or the conveyances originating from St. Croix is a feasible pathway inasmuch as ruminant livestock have been exported to the continental United States, albeit on an infrequent basis. However, even if the TBT eradication program is terminated in St. Croix, the change in risk for release of these ticks on the U.S. mainland is minimal, provided, per Federal regulations, mitigations of treating/destroying litter and manure from carriers and premises of tick-infested animals and cars or vehicles that carried these animals are followed. The risk of humans inadvertently releasing these ticks into the continental United States is independent of whether or not the TBT eradication program is discontinued in St. Croix as substantial human traffic is taking place from Caribbean countries other than St. Croix on a recurring basis.

3.2.5. Pathway 5: Smuggling of live *E. ruminantium*

3.2.5.1. Background information and analysis of data

Besides the United States, principal laboratories conducting research on *E. ruminantium* are located in France, Kenya, Netherlands, Senegal, South Africa, United Kingdom, and Zimbabwe (Camus et al. 1996). Stocks of this rickettsial agent are not known to be stored on St. Croix.

3.2.5.2. Conclusions

This pathway should be considered a feasible one. Consequently, the risk is not "zero" for introducing *E. ruminantium* into the continental United States by this pathway. The uncertainty surrounding the degree of importance to place on this pathway stems, in part, from a current absence of an international system that tracks the number of laboratories that maintain stocks of *E. ruminantium*, the quantities of *E. ruminantium* available, and their movement. This uncertainty also reflects an inability to predict illicit activities, and a lack of access to national intelligence information. Nonetheless, the risk of introducing *E. ruminantium* into the continental United States is independent of whether or not the TBT eradication program is terminated in St. Croix, as stocks of this agent are not known to be stored on St. Croix. Further, multiple sources of *E. ruminantium* in livestock populations occur in neighboring islands. Because of the presence of disease in many other locations, the pathway of introduction by intentional release of this agent results in minimal change from the otherwise occurring risk.

**Section 4: Summary of Risk Associated with Each Release Pathway**

Certain deficiencies of data precluded our ability to complete a quantitative risk assessment with the output for risk estimation expressed numerically. Instead, preparation of this document was qualitative in nature whereby the outputs for the likelihood of the outcomes are expressed in qualitative terms such as 'minimal,' 'low,' or 'high' with respect to increasing the risk of an adverse event due to the proposed change within the context of each release pathway. The term

'minimal' assumes that an increase in risk is possible but would be difficult or impossible to detect or demonstrate.

Based upon present information, the overall risks associated with heartwater occurring in domestic ruminant livestock and ruminant wildlife in the continental United States as a result of discontinuing the TBT eradication program in St. Croix, U.S. Virgin Islands, are:

*4.1. Release Pathway 1: Importation of E. ruminantium-Infected Animal Species*

The change in risk of release of *E. ruminantium*, and associated vector, *A. variegatum*, into the continental United States from St. Croix-origin domestic ruminants or ruminant wildlife species is minimal, provided certain mitigations prior to entry are directed toward these animals that focus both on the tick vector and the defined hazard (*E. ruminantium*).

*4.2. Release Pathway 2: Mechanical Transport of E. ruminantium-Infected A. variegatum Ticks by Migrating Cattle Egrets (Bubulcus ibis)*

The migration of cattle egrets from the Caribbean to the continental United States, some of which may be infested with *E. ruminantium*-infected *A. variegatum* ticks, is a feasible pathway for the release of *E. ruminantium* into susceptible domestic ruminant livestock and ruminant wildlife in the continental United States. However, cattle egrets will continue to have a presence on and around St. Croix and their ability to transport these ticks mechanically is unchanged even if the TBT eradication program in its present form is discontinued on this island. The change in risk is therefore minimal.

*4.3. Release Pathway 3: Mechanical Transport of E. ruminantium-Infected A. variegatum Ticks by Humans and Imported Animals, Reptiles, and Other Birds*

The risk of introducing *E. ruminantium* and its tick vector into the continental United States is independent of whether or not the TBT eradication program is terminated in St. Croix, given that airline and cruise ship passenger traffic, as well as that of reptiles, birds (other than poultry), poultry, primates, horses, NESOI animals, and other mammals are originating on a recurring basis from islands other than St. Croix. Change in risk is minimal.

*4.4. Release Pathway 4: Mechanical Transport of E. ruminantium-Infected A. variegatum Ticks by Fomites*

The risk is minimal for release of these ticks on the U.S. mainland, provided mitigations of treating/destroying litter and manure from carriers and premises of tick-infested animals and vehicles are followed per Federal regulations. The risk of humans inadvertently releasing these ticks into the continental United States is independent of whether or not the TBT eradication program is discontinued in St. Croix inasmuch as human traffic is taking place on a recurring basis from Caribbean countries other than St. Croix. Therefore, the change in risk from discontinuing the program is minimal.

*4.5. Release Pathway 5: Smuggling of Live E. ruminantium*

The risk of introducing *E. ruminantium* into the continental United States is independent of whether or not the TBT eradication program is terminated in St. Croix, as stocks of this agent are not known to be stored on St. Croix and infected animals or ticks are readily available in other locations. Therefore, the change in risk is minimal following the cessation of the TBT program.

**Closing Comments**

The continental United States is vulnerable to the release of heartwater from at least five different pathways. The TBT eradication program in its current form, i.e., a focus on eradicating the *A. variegatum* ticks on livestock premises, has the potential to increase the risk of releasing *E. ruminantium* onto the U.S. mainland through pathways involving uncontrolled trade in ruminant animals. However, if the aforementioned mitigations are followed, this risk (above that currently present) is minimal. The other pathways for release of *E. ruminantium* onto the U.S. mainland are independent of whether or not the TBT eradication program on St. Croix is discontinued and represent a minimal change in risk.

Due to the reservoir of infection is neighboring islands, and the potential for transport of infected ticks between islands, it is unlikely that a localized eradication program can succeed. Alternatives would include an eradication/control plan on a broader scale that would encompass the majority of the Caribbean basin or intensive management and parasite control on St. Croix animals by stock owners. The former would undoubtedly be expensive and fraught with political challenges between the various governments. The latter would seem to be a more reasonable and effective solution.

**References**


1. Alderink, F. and E. McCauley. 1988. The probability of the spread of *Amblyomma variegatum* in the Caribbean. Preventive Veterinary Medicine 6: 285–294.

2. Anderson, B. 2003. Cattle egret: *Bubulcus ibis*. Florida Fish and Wildlife Conservation Commission. January 2003. Florida's breeding bird atlas: A collaborative study of Florida's birdlife. Available at: wld.fwc.state.fl.us/bba/caeg.htm.

3. Andrew, H. and R. Norval. 1989. The carrier status of sheep, cattle, and African buffalo recovered from heartwater. Veterinary Parasitology 34: 261–266.

4. Arendt, W. 1988. Range expansion of the cattle egret (*Bubulcus ibis*) in the Greater Caribbean Basin. Colonial Waterbirds 11 (2): 252–262.

5. Barré, N., G. Uilenberg, P. Morel, E. Camus. 1987. Danger of introducing Heartwater onto the American mainland: potential role of indigenous and exotic *Amblyomma* ticks. Onderstepoort Journal of Veterinary Research 54: 405-417.

6. Barré, N., G. Garris, G. Borel, E. Camus. 1988. Hosts and populations dynamics of *Amblyomma* (Acari: Ixodidae) on Guadeloupe, French West Indies. Journal of Medical Entomology 25(2a): 111–115.

7. Becklund, W. 1968. Ticks of veterinary significance found in imports in the U.S. The Journal of Parasitology 54(3): 622–628.

8. Birnie, E.F., M.J. Burridge, E. Camus, N. Barre. 1985. Heartwater in the Caribbean: isolation of *Cowdria ruminantium* from Antigua. The Veterinary Record 116: 121–123.

9. Bram, R., J. George, R. Reichard, W. Tabachnick. 2002. Threat of foreign arthropod-borne pathogens to livestock in the U.S. Journal of Medical Entomology 39(3): 405–416.

10. Browder, J.A. 1973. Long-distance movements of Cattle Egrets. Bird-Banding 44: 158-170.

11. Burridge, M. 2001. Ticks (Acari: Ixodidae) spread by the international trade in reptiles and their potential roles in dissemination of diseases. Bulletin of Entomological Research 91: 3–23.

12. Burridge, M., L. Simmons, B. Simbi, S. Mahan, P. Fourneir, D. Raoult. 2002. Introduction of the exotic tick *Amblyomma hebraeum* into Florida on a human host. Journal of Parasitology 88 (4): 800–801.

13. Camus, E. and N. Barré. 1987. Epidemiology of Heartwater in Guadeloupe and in the Caribbean. Onderstepoort Journal of Veterinary Research 54 (3): 419–426.

14. Camus, E., N. Barré, D. Martínez, G. Uilenberg. 1996. Heartwater (Cowdriosis). A review, 2nd Edition. Office International des Epizooties, Paris, 177 pp.

15. Corn, J., N. Barré, B. Thiebot, T. Creekmore, G. Garris, V. Nettles. 1993. Potential role of cattle egrets, *Bubulcus ibis* (Ciconiformes: Ardeidae), in the dissemination of *Amblyomma variegatum* (Acari: Ixodidae) in the Eastern Caribbean. Journal of Medical Entomology 30 (6): 1029–1037.

16. Corn, J., N. Barré, G. Garris, V. Nettles. 1996. Potential impact of wildlife on the Tropical Bont Tick eradication program in the Caribbean. Annals of the New York Academy of Sciences 791: 77–84.

17. Cowdry, E. 1925. Studies on the etiology of heartwater. I. *Rickettsia ruminantium* (n. sp.) in the tissues of infected animals. Journal of Experimental Medicine 42: 231–252.

18. Cumming, G. 1998. Host preferences in African ticks (Acari: Ixodidae): A quantitative data set. Bulletin of Entomological Research 88: 379–406.

19. Dardiri, A., L. Logan, C. Mebus. 1987. Susceptibility of white-tailed deer to experimental heartwater infections. Journal of Wildlife Diseases 23 (2): 215–219.

20. Dawn, W. 1959. Cattle egrets provoke cattle to move and pick flies off bulls. The Auk 76: 97–98.

21. Deem, S., R. Norval, T. Yonow, T. Peter, S. Mahan, M. Burridge. 1996. The epidemiology of heartwater: establishment and maintenance of endemic stability. Parasitology Today 12 (10): 402–405.

22. Deem, S. 1998. A review of heartwater and the threat of introduction of *Cowdria ruminantium* and *Amblyomma* spp. ticks to the American mainland. Journal of Zoo and Wildlife Medicine 29 (2): 109–113.

23. Dinsmore, J.J. 1973. Foraging success of Cattle egrets, *Bubulcus ibis*. The American Midland Naturalist 89: 242–246.

24. Estrada-Peña, A. and F. Jongejan. 1999. Ticks feeding on humans: a review of records on human-biting Ixodoidea with special reference to pathogen transmission. Experimental and Applied Acarology 23: 685–715.

25. FAOSTAT, Food and Agriculture Organization of the United Nations, Web site. 2007. Cattle, sheep, and goat inventories in Caribbean for 2006. Available at: faostat.fao.org/site/573/DesktopDefault.aspx?PageID=573.

26. Florida Fish and Wildlife Conservation Commission Web site. 2006. Florida's breeding bird atlas: cattle egret (Bubulcus ibis). Available at: www.floridaconservation.org/bba/caeg.htm.

27. Garris, G. 1984. Colonization and life cycle of *Amblyomma variegatum* (Acari: Ixodidae) in the laboratory in Puerto Rico. Journal of Medical Entomology 21: 86–90.

28. Gilsdorf, M., J. Bentz-Blanco, M.S. Camacho, M.A. Borri-Diaz. 2006. Briefing paper on the *Amblyomma variegatum* Eradication Program in St. Croix, USVI. USDA:APHIS:VS.

29. Grubb, T.C. 1976. Adaptiveness of foraging in the Cattle Egret. Wilson Bulletin 88: 145–148.

30. Guillén, A., J. Prosper, J.L. Echevarrias. 1994. Estimacion de la dieta de la Garcilla Bueyera a partir del analisis de regurgitaciones de pollos: problemas debidos a la digestion diferencial de las presas. Doñana, Acta Vertebrata 21: 204–212.

31. Hancock, J and J. Kushlan. 1984. Cattle egrets (*Bubulcus ibis*). The Herons Handbook. Harper and Row, New York. 288 p.

32. Heatwole, H. 1965. Some aspects of the association of Cattle egrets with cattle [in Puerto Rico]. Animal Behavior 13: 79–83.

33. Horak, I., K. de F. MacIvor, T. Petney, V. de Vos. 1987. Some avian and mammalian hosts of *Amblyomma hebraeum* and *Amblyomma marmoreum* (Acari: Ixodidae). Onderstepoort Journal of Veterinary Research 54: 397–403.

34. Ilemobade, A. 1976. Study on heartwater and the causative agent *Cowdria ruminantium* (Cowdry, 1925) in Nigeria. PhD Thesis, Ahmadu Bello University, Zaria, Nigeria, 299 pp.

35. Kobold, A., D. Martinez, E. Camus, F. Jongejan. 1992. Distribution of heartwater in the Caribbean determined on the basis of detection of antibodies to the conserved 32-kiloDalton protein of *Cowdria ruminantium*. Journal of Clinical Microbiology 30 (7): 1870–1873.

36. Mackenzie, P. and R. Norval. 1980. The transmission of *Cowdria ruminantium* by *Amblyomma tholloni*. Veterinary Parasitology 7: 265–268.

37. Mahan, S., T. Peter, S. Semu, B. Simbi, R. Norval, A. Barbet. 1995. Laboratory reared *Amblyomma hebraeum* and *Amblyomma variegatum* ticks differ in their susceptibility to infection with *Cowdria ruminantium*. Epidemiology and Infection (Cambridge, Eng) 115: 345-353.

38. Mahan, S.M., T.F. Peter, B.H. Simbi, K. Kocan, E. Camus, A.F. Barbet, M.J. Burridge. 2000. Comparison of efficacy of American and African *Amblyomma* ticks as vectors of heartwater (*Cowdria ruminantium*) infection by molecular analyses and transmission trials. Journal of Parasitology 86: 44-49.

39. Maré, C. 1998. Heartwater (Cowdriosis). Foreign Animal Diseases: "The Gray Book." US Animal Health Association. Available at: http://www.vet.uga.edu/vpp/gray_book/pdf/HEA.htm.

40. McKilligan, N.G., D.S. Reimer, D.H.C. Seton, D.H.C. Davidson, J.T. Willows. 1993. Survival and seasonal movements in the Cattle Egret in Eastern Australia. Emu 93: 79–87.

41. Mertins, J. and J. Schlater. 1991. Exotic ectoparasites of ostriches recently imported into the U.S. Journal of Wildlife Diseases 27(1): 180–182.

42. Mora, M.A. and J.M. Miller. 1998. Foraging flights, reproductive success and organochlorine contaminants in cattle egrets nesting in a residential area in Bryan, Texas. Texas Journal of Science 50: 205–214.

43. Norval, R. and P. Mackenzie. 1981. The transmission of *Cowdria ruminantium* by *Amblyomma sparsum.* Veterinary Parasitology 8: 189–191.

44. Oberem, P. and J. Bezuidenhout. 1987. Heartwater in hosts other than domestic ruminants. Onderstepoort Journal of Veterinary Research 54: 271–275.

45. Pegram, R., A. Rota, R. Onkelinx, D. Wilson, P. Bartlette, B. Nisbett, G. Swanston, P. Vanterpool, J. de Castro. 1996. Eradicating the tropical bont tick from the Caribbean. World Animal Review 87.

46. Pegram, R., L. Indar, C. Eddi, J. George. 2004. The Caribbean *Amblyomma* Program: some ecologic factors affecting its success. Annals of the New York Academy of Sciences 1026: 302–311.

47. Perreau, P., P.C. Morel, N. Barre, P. Durand. 1980. Existence de la cowdriose (heartwater) a *Cowdria ruminantium* chez les ruminants des Antilles Francaises (La Guadeloupe) et des Mascareignes (La Reunion et Ile Maurice). Revue d Elevage et de Medecine Veterinaire des Pays Tropicaux (Paris) Trop 33: 21–22.

48. Peter, T., E. Anderson, M. Burridge, S. Mahan. 1998. Demonstration of a Carrier State for *Cowdria ruminantium* in Wild Ruminants from Africa. Journal of Wildlife Diseases 34 (3): 567–575.

49. Peter, T., N. Bryson, B. Perry, C. O'Callaghan, G. Medley, G. Smith, G. Mlambo, I. Horak, M. Burridge, S. Mahan. 1999. *Cowdria ruminantium* infection in ticks in the Kruger National Park. The Veterinary Record 145: 304–307.

50. Peter, T., M. Burridge, S. Mahan. 2000. Competence of the African Tortoise tick, *Amblyomma marmoreum* (Acari: Ixodidae) as a vector of the agent of heartwater (*Cowdria ruminantium*). Journal of Parasitology 86 (3): 438–441.

51. Peter, T.F., M.J. Burridge, S.M. Mahan. 2002. *Ehrlichia ruminantium* infection (heartwater) in wild animals. Trends in Parasitology 2002 May;18 (5): 214–218.

52. Petney, T.N., I.G. Horak, Y. Rechav. 1987. The ecology of the African vectors of heartwater, with particular reference to *Amblyomma hebraeum* and *Amblyomma variegatum*. Onderstepoort Journal of Veterinary Research 54: 381–395.

53. Public Health and Medical Entomology Web site. 2007. Trans-stadial transmission definition. Available at: www.entm.purdue.edu/publichealth/glossary.html#t.

54. Ridgely, R.S., T.F. Allnutt, T. Brooks, D.K. McNicol, D.W. Mehlman, B.E. Young, J.R. Zook. 2003. Digital Distribution Maps of the Birds of the Western Hemisphere, version 1.0. NatureServe, Arlington, Virginia, USA.

55. Roe, D., T. Mulliken, S. Milledge, J. Mremi. 2002. Making a killing or making a living: Wildlife trade, trade controls and rural livelihoods. Biodiversity and Livelihoods Issue paper 06. International Institute for Environment and Development (IIED), London, UK. 128 pp.

56. Singh, N., N. Sodhi, S. Khera. 1988. Biology of the Cattle Egret *Bubulcus ibis cormandus* (Boddaert). Records of the Zoological Survey of India 104: 1–143.

57. Southeastern Cooperative Wildlife Disease Study Web site. 2006. Cattle Egrets and the Tropical Bont Tick. SCWDS Briefs. July 1992; 8.2. Available at: www.uga.edu/scwds/topic_index/1992/CATTLE~1.pdf.

58. Telfair, R. 2006. Cattle egret (*Bubulcus ibis*). The birds of North America Online (A. Poole, Ed.). Ithaca: Cornell Laboratory of Ornithology. Birds of North American Online database. Available at: bna.birds.cornell.edu/BNA/account/Cattle_Egret/.

59. Thompson, C.F., S.M. Lanyon, K.M. Thompson. 1982. The influence of foraging benefits on association of cattle egrets (*Bubulcus ibis*) with cattle. Oceologia 52: 167–170.

60. Uilenberg, G. 1982. Experimental transmission of *Cowdria ruminantium* by the Gulf Coast tick *Amblyomma maculatum*: Danger of introducing heartwater and benign African theileriasis onto the American mainland. American Journal of Veterinary Research 43 (7): 1279–1282.

61. Uilenberg, G., N. Barre, E. Camus, M.J. Burridge, G.I. Garris. 1984. Heartwater in the Caribbean. Preventive Veterinary Medicine 2: 255–267.

62. USDA:APHIS Web site. 2007. Veterinary Services Memorandum No. 556.1. Ticks – Dipping and Treatment Procedures. Available at: inside.aphis.usda.gov/vs/downloads/memos/556_1.pdf.

63. USDA Office of Inspector General Web site. 2007. Activities in Florida, Audit Report No. 3304-1-At. March 2000. APHIS PPQ. Available at: www.usda.gov/oig/webdocs/Fullrpt.pdf.

64. U.S. Department of Transportation, Maritime Administration Web site. 2007. Cruise Statistics: Summary Tables 2003–2007. Available at: www.marad.dot.gov/Marad_Statistics/2007%20STATISTICS/2007%20CRUISE%20STATS/cruise%20data,%202003-2007(1)%20(summary).xls.

65. U.S. Fish and Wildlife Service Web site. 2006a. Cattle egret (*Bubulcus ibis ibis*). Waterbirds. Available at: www.fws.gov/birds/waterbirds/MANEM/PDFs/CAEG6.pdf.

66. U.S. Fish and Wildlife Service Web site. 2006b. Annual report FY 2004. U.S. Fish and Wildlife Service, Department of the Interior. Office of Law Enforcement. August 2005. Available at: www.fws.gov/le/pdffiles/AnnualReportFY2004.pdf.

67. U.S. Fish and Wildlife Service Web site. 2006c. U.S. CITES Biennial Report, 2000–2001. U.S. Fish and Wildlife Service. Department of the Interior. Fall 2002. Available at: library.fws.gov/IA_Pubs/CITES_Biennialreport00-01.pdf.

68. U.S. Fish and Wildlife Service Web site. 2006d. U.S. CITES Biennial Report, 2002–2003. U.S. Fish and Wildlife Service. Department of the Interior. October 2004. Available at: www.fws.gov/international/pdf/CITESBiennialrpt0203.pdf

69. U.S. Geological Survey Web site. 2007. Patuxent Wildlife Research Center Bird Banding Laboratory. Available at: www.pwrc.usgs.gov/bbl/homepage/.

70. U.S. National Archives and Records Administration Web site. 2007a. Code of Federal Regulations. Title 42: Public Health. Chapter I: Public Health Service, Department of Health and Human Services. Part 73: Select Agents and Toxins. Available at: a257.g.akamaitech.net/7/257/2422/16nov20071500/edocket.access.gpo.gov/cfr_2007/octqtr/42cfr73.3.htm.

71. U.S. National Archives and Records Administration Web site. 2007b. Code of Federal Regulations. Title 9: animals and animal products. Chapter I: Animal and Plant Health Inspection Service, Department of Agriculture. Part 121: Possession, Use, and Transfer of Select Agents and Toxins. Available at: a257.g.akamaitech.net/7/257/2422/14feb20071500/edocket.access.gpo.gov/cfr_2007/janqtr/9cfr121.3.htm.

72. U.S. National Archives and Records Administration Web site. 2007c. Code of Federal Regulations. Title 9: animals and animal products. Chapter I: Animal and Plant Health Inspection Service, Department of Agriculture. Part 71: General Provisions. Available at: www.access.gpo.gov/nara/cfr/waisidx_07/9cfr71_07.html.

73. U.S. National Archives and Records Administration Web site. 2007d. Code of Federal Regulations. Title 9: animals and animal products. Chapter I: Animal and Plant Health Inspection Service, Department of Agriculture. Part 72: Texas (Splenetic) Fever in Cattle. Available at: www.access.gpo.gov/nara/cfr/waisidx_07/9cfr72_07.html.

74. Van den Bosch, F., R. Hengeveld, J.A.J. Metz. 1992. Analysing the Velocity of Animal Range Expansion. Journal of Biogeography 19: 135–150.

75. Walker, J. 1987. The tick vectors of *Cowdria ruminantium* (Ixodoidea, Ixodidae, genus *Amblyomma*) and their distribution. Onderstepoort Journal of Veterinary Research 54: 353–379.

76. Wesonga, F., S. Mukolwe, J. Grootenhuis. 2001. Transmission of *Cowdria ruminantium* by *Amblyomma gemma* from Infected African Buffalo (*Syncerus caffer*) and Eland (*Taurotragus oryx*) to sheep. Tropical Animal Health and Production 33: 372–390.

77. Wilson, D. and R. Bram. 1998. Foreign pests and vectors of arthropod-borne diseases. Foreign Animal Diseases. United States Animal Health Association. Available at: www.vet.uga.edu/vpp/gray_book/FAD/fpv.htm.

78. World Organization for Animal Health (OIE) Web site. 2007a. Guidelines for Import Risk Analysis. Chapter 1.3.2. Terrestrial Animal Health Code 2005. Office International des Epizooties (OIE), Paris, France. Available at: www.oie.int/eng/normes/mcode/en_chapitre_1.3.2.htm.

79. World Organization for Animal Health (OIE) Web site. 2007b. World Animal Health Information Database (WAHID). List of countries by disease situation. Heartwater. Available at: www.oie.int/wahidprod/public.php?page=disease_status_lists&disease_id=20.

80. World Organization for Animal Health (OIE) web site. 2007c. Terrestrial Animal Health Code. Chapter 2.2.7. Heartwater. Available at: http://www.oie.int/eng/normes/MCode/en_chapitre_2.2.7.htm.