The Transmissible Spongiform Encephalopathies of Livestock

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Abstract

Prion diseases or transmissible spongiform encephalopathies (TSEs) are fatal protein-misfolding neurodegenerative diseases. TSEs have been described in several species, including bovine spongiform encephalopathy (BSE) in cattle, scrapie in sheep and goats, chronic wasting disease (CWD) in cervids, transmissible mink encephalopathy (TME) in mink, and Kuru and Creutzfeldt-Jakob disease (CJD) in humans. These diseases are associated with the accumulation of a protease-resistant, disease-associated isoform of the prion protein (called PrPres) in the central nervous system and other tissues, depending on the host species. Typically, TSEs are acquired through exposure to infectious material, but inherited and spontaneous TSEs also occur. All TSEs share pathologic features and infectious mechanisms but have distinct differences in transmission and epidemiology due to host factors and strain differences encoded within the structure of the misfolded prion protein. The possibility that BSE can be transmitted to humans as the cause of variant Creutzfeldt-Jakob disease has brought attention to this family of diseases. This review is focused on the TSEs of livestock: bovine spongiform encephalopathy in cattle and scrapie in sheep and goats.

Key words: Bovine spongiform encephalopathy; cattle; goats; prion; protein misfolding; scrapie; sheep; transmissible spongiform encephalopathy

Prion Disease

Prion diseases are a family of infectious protein-misfolding disorders resulting from aberrant folding and accumulation of the prion protein that leads to neurodegeneration (Aguzzi et al. 2008; Prusiner 1991). The misfolded form of the prion protein is commonly denoted PrPres, as it was originally described as the disease causing agent of scrapie in sheep and goats (Prusiner et al. 1998). While normal cellular prion protein is in a predominantly alpha-helical structure (Prusiner 2001), misfolding of the prion protein results in a largely beta-sheet conformation (Pan et al. 1993). Misfolded prion molecules (PrPres) act as a template for the aberrant refolding of normal cellular prion protein (PrPc) (Caughey and Raymond 1991) (Figure 1). Molecules of PrPres form aggregates, become resistant to cellular digestion, and can accumulate in lymphoid and nervous tissues, particularly the central nervous system (CNS) (DeArmond et al. 1985). In addition to the resistance of PrPres to cellular digestion (Caughey et al. 1990), it is also resistant to digestion in vitro by proteinase K (PK) (McKinley et al. 1983; Oesch et al. 1985). Prion diseases result in accumulation of PrPres and spongiform change in the brain (Budka 2003; Jeffrey and Gonzalez 2004) (Figure 2). Prion diseases are transmissible from one susceptible host to another and are therefore referred to as transmissible spongiform encephalopathies (TSEs).

Strains of Prion Disease

Although all are caused by misfolded prion protein, TSEs within a species exist in multiple strains that may exhibit biochemical
differences, as well as different disease phenotype and pathogenesis (Cobb and Surewicz 2009; Sigurdson et al. 2007). In a given host, both PrP C and PrPSc are composed of identical amino acid sequences, so strain properties are maintained through conformational differences in PrPSc (Bessen and Marsh 1994; Legname et al. 2005). Strains can be differentiated by:

- differing clinical signs (Pattison and Millson 1961)
- incubation periods and lesion profiles in mouse models (Fraser and Dickinson 1968, 1973)
- cellular and neuroanatomical deposition patterns of PrPSc (Bruce et al. 1989; Gonzalez et al. 2002)
- molecular profile on Western blot:
  - electrophoretic mobility (Bessen and Marsh 1992)
  - reactivity to antibodies near the PK cleavage point (Langeveld et al. 2011)
  - glycoform ratios (Stack et al. 2002)

The reaction of different strain isolates to conformational antibodies (Safar et al. 1998) and stability assays (Vrentas et al. 2013) further supports the hypothesis that strains arise from differences in protein structure (Peretz et al. 2002). Strain diversity may arise from the inherent conformational flexibility of the prion protein, the presence of prion protein gene (PRNP) polymorphisms within a host species, or interspecies transmission events (Morales et al. 2007). Both bovine spongiform encephalopathy (BSE) and scrapie have strains classified as either classical or atypical. The classical strains have molecular, transmission, and phenotype characteristics consistent with the disease endemic within a population; and the atypical strains have molecular and epidemiologic characteristics that allow them to be differentiated from the classical strains (Benestad et al. 2003; Biacabe et al. 2004). The transmissibility and pathogenesis of a given TSE are dependent upon multiple factors including the PRNP genotype of the host animal, the strain of TSE, and the route of exposure.

Prion Diseases are Diseases of Protein Misfolding

Prion diseases share a common mechanism with other protein-misfolding disorders including Alzheimer’s disease, Parkinson’s disease, amyotrophic lateral sclerosis (ALS), and Huntington’s disease, in which misfolded disease-associated proteins catalyze a refolding of normal cellular protein that accumulates and is associated with neural degeneration (Fernandez-Borges et al. 2013; Hetz et al. 2003; Prusiner 2013; Soto 2003; Soto and Satani 2011). However, prion diseases are distinct from these other disorders in that they are readily transmissible to susceptible individuals via biologically significant routes of exposure.

Prion Diseases and Livestock

Besides having fatal consequences to infected animals, scrapie and BSE are particularly important because of economic consequences to the livestock industry and potential public health concerns (Williams 2002). Scrapie can spread within a flock and brings economic losses due to production loss, export loss, and increased cost for carcass disposal, which ranges from $10–$20 million USD annually. The national scrapie eradication program began in 1952, was revisited in 2001, and implemented new surveillance practices in 2002–2003 that have decreased the number of cull sheep that are scrapie positive at slaughter by 90% (U.S. Department of Agriculture 2015). A single case of classical BSE was diagnosed in the United States in 2003 in an animal imported from Canada (Richt et al. 2007). As a result, loss of beef exports and domestic consumption was estimated to cost U.S. producers $4.7 billion USD in 2004. Since the first case of BSE was diagnosed,
three additional atypical cases have been reported (Richt and Hall 2008; Richt et al. 2007; U.S. Department of Agriculture Animal and Plant Health Inspection Service 2012). The announcement of a single case of BSE in Canada in 2003 cost over $5.7 billion in total outputs (Fréchette 2005). A decade after the first reported case, beef exports from Canada were still approximately $1 billion less than they were in 2002. In the United Kingdom, where over 180,000 cattle were diagnosed with BSE and up to 3 million were likely to have been affected (Donnelly et al. 2002), the cost to the public was more than £5 billion. Loss of exports, consumer safeguards, compensation payments, and decrease in consumer demand all contribute to the costs incurred when additional cases of BSE are diagnosed.

Prion Disease and Human Health

There are a number of TSEs that have been described in humans. They can be categorized as sporadic (unknown etiology), genetic, or acquired (Sikorska and Liberski 2012). The acquired TSEs in humans include iatrogenic Creutzfeldt-Jakob disease (iCJD), Kuru, and variant CJD (vCJD). Kuru and iCJD are the result of transmission between humans (Will 2003), and vCJD is a zoonotic disease in which BSE from cattle infects humans that have ingested contaminated beef (Dormont 2002).

TSEs of Livestock

Experimentally, TSEs of livestock are of interest for several reasons. Firstly, interspecies transmission experiments are crucial for the food animal industry to ensure the safety of the food supply. Secondly, TSEs in ruminants provide a well-characterized large animal model of protein-misfolding neurodegenerative disorders. Here we will review scrapie in sheep and goats, and BSE in cattle, experiments to assess interspecies transmission, approaches to antemortem diagnosis of TSEs in sheep and cattle, and finally the relevance of scrapie and BSE to human health and disease.

Scrapie

Genetics of Susceptibility

The TSEs are diseases of misfolded prion protein, and disease susceptibility is modulated by the amino acid sequence of the host’s prion protein. The codons of PRNP that are especially
significant in determining susceptibility or resistance to scrapie in sheep include 136, 154, and 171. PRNP haplotypes correlated with increased susceptibility are 136 valine (V), 154 arginine (R), and 171 glutamine (Q) (VRQ), whereas 136 alanine (A) (Goldmann et al. 1996; Hunter et al. 1991), 154 histidine (H) (Laplancha et al. 1993), and 171 arginine (R) (AHR) (Goldmann et al. 1994; O’Rourke et al. 1997) are associated with resistance to natural scrapie. Codon 171 appears to have the most discernible influence, where sheep with 171QQ are susceptible and 171RR are resistant. Another amino acid, lysine (K), occurs at codon 171 in some breeds of sheep. Similarities in charge and structure suggest that K may behave similarly to R. Sheep with a single 171K allele have prolonged incubation times after intracranial inoculation (Greenlee, Zhang et al. 2012), but it has not been determined whether sheep homozygous for the 171K allele will be resistant to scrapie or have prolonged incubation times. A significant reduction in the number of scrapie cases has been achieved in many locations by instituting breeding programs to increase the ARR allele in the sheep population in conjunction with reduction in the number of scrapie cases has been achieved in many locations by instituting breeding programs to increase the ARR allele in the sheep population in conjunction with removal of affected animals. Polymorphisms that occur with lesser frequency, such as a substitution of methionine (M) by threonine (T) at codon 112 (Gonzalez et al. 2012; Laegreid et al. 2008) or leucine (L) by phenylalanine (F) at codon 141 (Gonzalez et al. 2012), also have been shown to increase resistance to scrapie.

Allelic variation in PRNP has been shown to modulate incubation times and susceptibility for scrapie in sheep. Scrapie in goats has been less well studied than in sheep, but numerous (at least 37) polymorphisms have been described in goat breeds worldwide (Acin et al. 2013). A subset of these potential amino acid substitutions appear to affect scrapie susceptibility: G127S (Goldmann et al. 2011), I142M (Goldmann et al. 1996), N146S/D (Papasavva-Stylianou et al. 2007), H154R (Barillet et al. 2009; Billinisy et al. 2002; Papasavva-Stylianou et al. 2011; Vaccari et al. 2006), Q211R (Barillet et al. 2009), and Q222K (Acitis et al. 2006; Barillet et al. 2009; Vaccari et al. 2006). Experimental studies suggest that M142 prolongs incubation period (Goldmann et al. 1996) but does not result in resistance to scrapie (Lacroux, Perrin-Chauveneau et al. 2014). However, H154, Q211, and K222 carriers are all resistant to scrapie after oral exposure (Lacroux, Perrin-Chauveneau et al. 2014). Cases have been reported in heterozygous K222 animals, but not in homozygous K222 (Barillet et al. 2009; Corbiere et al. 2013). The fact that a small number of K222 goats developed scrapie after intracranial inoculation confirms that resistance to scrapie is not absolute. Goats have been naturally infected with both atypical scrapie (Le Dur et al. 2005; Seuberlich 2007) and BSE (Elloï et al. 2005). Similar to sheep, the risk of developing atypical scrapie has been shown to be higher in K154 goats (Colussi et al. 2008). Results of studies using transgenic mice expressing K222 demonstrated resistance to bovine spongiform encephalopathy, further suggesting K222 would make a good candidate for selective breeding programs to enhance scrapie resistance in goats (Aguilar-Calvo et al. 2014).

Classical Scrapie in Sheep

Scrapie is the first described and most studied transmissible spongiform encephalopathy. Since the first description nearly 300 years ago in the United Kingdom and other western European countries, scrapie has been reported throughout the world with the notable exception of Australia and New Zealand. The transmissibility of scrapie was demonstrated by Cuillé and Chelle in 1936 (Schneider et al. 2008). It most likely spread through the export of subclinically affected animals (Dewiler and Baylis 2003). The first case of scrapie in the United States was reported in 1947 in imported sheep. This naturally occurring disease has a low incidence in affected flocks and now is rare due to eradication programs.

Transmission of scrapie from suspensions of brain and spinal cord from infected animals by intraocular, epidural, subcutaneous, and intracranial routes was demonstrated as early as 1936 (Schneider et al. 2008). Later, the accidental inclusion of tissues from a scrapie-infected flock in a loping-ill vaccine demonstrated that brain, spinal cord, and spleen contained infectious material; the agent was resistant to formalin; subcutaneous injection was sufficient for transmission; and the incubation period was 2 years or more (Gordon 1946). Natural transmission within flocks is thought to occur most commonly from the ewe to offspring of susceptible genotypes through the placenta and placental fluids, resulting in vertical transmission to susceptible offspring and the potential for horizontal exposure at the time of lambing (Touzeau et al. 2006). The demonstration of PrPSc in the placenta has been shown to be associated with placentomes of genetically susceptible offspring (Andreoletti et al. 2002) and in small amounts in the cotyledons of lambs of resistant genotypes that share the same uterine horn with a sibling of a susceptible genotype (Alverston et al. 2006). In addition to the presence of PrPSc in the cotyledons, infectivity of fetal tissues supports the possibility of utero transmission (Spiroupolous et al. 2014). Transmission by milk and colostrum represent an additional risk of transmission to susceptible offspring born to infected dams (Konold et al. 2008, 2013). Environmental contamination leading to oral exposure of the scrapie agent is a major route of entry (Andreoletti et al. 2000; Erdsal et al. 2005; Gough and Maddison 2010; Ryder et al. 2004; van Keulen et al. 2002). Testing of infectivity of semen from scrapie-infected rams suggests that artificial insemination or natural breeding represents a negligible risk for scrapie transmission (Sarradin et al. 2008). However, potential prion seeding activity has been demonstrated in semen from infected rams by in vitro assays, suggesting that this route of transmission may require further study (Rubenstein et al. 2012).

There is a long asymptomatic period lasting 2 to 7 years (depending on dose and genotype of sheep) between initial exposure to scrapie and the development of clinical signs. After ingestion, PrPSc crosses the intact intestinal barrier at the level of the enterocytes and passes rapidly into lymph and blood. These initial PrPSc steps are identical in susceptible and resistant sheep; however, only in susceptible sheep has PrPSc accumulation been shown to subsequently take place in lymphoid structures (particularly in association with follicular dendritic cells) (Heggo et al. 2000; Jeffrey et al. 2006). The first replication of the prion agent is within the lymphoreticular system, most likely the ileal Peyer’s patches (Andreoletti et al. 2000; Jeffrey et al. 2006). Early in the course of disease, PrPSc can be demonstrated in tonsil, spleen, and retropharyngeal and mesenteric lymph nodes (Muramatsu et al. 1994; Race et al. 1992). Regardless of the first site of replication, the abnormal prion spreads throughout lymphoid organs and is detectable for months prior to being detectable in the brain. Abnormal prion is detectable by immunohistochemistry in the lymphoid follicles, which allows for potential antemortem diagnosis using biopsies from the third eyelid (O’Rourke et al. 2000), tonsil (Schreuder et al. 1998), or rectal mucosa (Gonzalez, Horton et al. 2008). Immunoreactivity is most apparent at the center of the follicles and has been localized to both follicular dendritic cells and tingible body macrophages (Jeffrey et al. 2000). Oral uptake requires normal numbers of Peyer’s patches, but not enteric lymphocytes (Prinz et al. 2003), suggesting an important role of follicular dendritic cells (Mabott and Bruce 2002). One factor in the greater relative susceptibility of younger sheep as compared with older sheep may have to do with more
prominent gut-associated lymphoid tissues in young animals (Press et al. 2004). Because of widespread lymphoid accumulation of PrPC\textsuperscript{Sc} during the asymptomatic phase, infected sheep may serve as a source of prion contamination to the environment long before the onset of clinical signs. Despite the participation of the lymphoid system in the pathogenesis of scrapie, there is no specific humoral immune response (Porter et al. 1973) due to tolerance to host PrPC\textsuperscript{Sc} (Bendheim et al. 1992), which has the same amino acid sequence.

Atypical Scrapie

Atypical (Nor98) scrapie is a second TSE of sheep that was first described in Norway in 1998 (Benestad et al. 2003, 2008). These cases were defined as atypical based on several criteria: clinical presentation, molecular characteristics of the abnormal prion protein, distribution of PrPC\textsuperscript{Sc} within infected sheep, genotypes of affected sheep, and epidemiology. Since 1998, additional cases have been diagnosed throughout Europe (Arsac et al. 2007; Dagleish et al. 2008; De Bosschere et al. 2007; Fediaevsky et al. 2008; Gavier-Widen et al. 2004; Loiacono et al. 2009; Mitchell et al. 2010) and one case in New Zealand (Kittelberger et al. 2010). Progressive ataxia was the predominant clinical sign in the original report (Benestad et al. 2003), but most often these cases are detected during routine diagnostic screening of older cull animals (active surveillance) where neurologic findings are absent or ill defined. Cases of atypical scrapie are distinguished by a western blot profile that has a 5-band profile with a prominent lower band at approximately 12 kDa (or lower depending on conditions [Klingeborn et al. 2006]), whereas classical scrapie has the non-glycosylated band at approximately 19–21 kDa (Hayashi et al. 2005; Hope et al. 1999; Somerville and Ritchie 1990). Furthermore, the PrPC\textsuperscript{Sc} molecule of atypical scrapie is relatively sensitive to proteases (Klingeborn et al. 2006), resulting in discrepant diagnostic test results depending on the test method used (Baron, Biacabe et al. 2007; Buschmann et al. 2004). The spongiform change and PrPC\textsuperscript{Sc} deposition in atypical scrapie cases occur predominantly in the cortices of the cerebellum and the cerebrum, rather than the medulla oblongata as seen in classical sheep scrapie. This does not fit the model of oral infection of intestinal Peyer’s patches followed by ascension through the vagus nerve to the brainstem. The agent has not been described outside of the CNS: currently available methods have failed to demonstrate PrPC\textsuperscript{Sc} in the lymphoid tissues of sheep affected with atypical scrapie (Andreoletti et al. 2011; Benestad et al. 2003). Atypical scrapie cases are sometimes identified in sheep with genotypes considered resistant to classical scrapie: affected sheep in the original report carried at least one A136H154Q171 allele (Benestad et al. 2003), and subsequent studies have demonstrated an increased risk of atypical scrapie in sheep with the AHQ and AF154RQ haplotypes (Saunders et al. 2006). Sheep with the ARR haplotype do not appear to be protected against developing atypical scrapie (De Bosschere et al. 2007; Saunders et al. 2006). Because atypical scrapie is usually detected in only one sheep in a flock, does not appear to be contagious or transmits very poorly under natural conditions (Fedieaevsky et al. 2010), and is usually found in older animals, a sporadic etiology (i.e., spontaneous occurrence or de novo pathogenesis) has been suggested. Supporting the concept of a sporadic occurrence is the long-held categorization of certain forms of CJD having a sporadic etiology and recent evidence that abnormally-folded prions can arise de novo in experimental systems. Interestingly, atypical scrapie has been transmitted experimentally to AHQ sheep by the intracranial (Simmons et al. 2007) and oral (Simmons et al. 2011) routes, and infectivity has been demonstrated in tissues outside the CNS (ileum and spleen [Simmons et al. 2011]); skeletal muscle, lymphoid tissues, peripheral nerves [Andreoletti et al. 2011]) of affected individuals despite a failure to detect PrPC\textsuperscript{Sc} (Simmons et al. 2011). Evidence of infectivity outside of the CNS suggests that there is the possibility for natural transmission or spread through contaminated animal feed. Atypical scrapie has also been reported in goats (Seuberlich 2007), where the molecular profile on western blot is similar to atypical scrapie in sheep, but the distribution of lesions within the brain is more rostral (thalamus and midbrain) than that of atypical scrapie of sheep (Seuberlich 2007).

Classical Scrapie in Goats

Scrapie in goats was first described in 1939 after experimental transmission from sheep, and the first natural case was described shortly thereafter (Fast and Groscup 2013). Goats with scrapie are commonly in mixed herds with sheep, but scrapie can also occur in goats that have not had contact with sheep (Wood et al. 1992). Similar to what is observed in sheep with scrapie, assessment of goats with natural scrapie suggests widespread involvement of the lymphoreticular system, specifically retropharyngeal lymph node and palatine tonsil (Gonzalez et al. 2009). In contrast to what is commonly observed in sheep, accumulation of PrPC\textsuperscript{Sc} in the brain of goats with scrapie appears to be highly dependent upon genotype at codon 142. In a study of naturally infected goats, 75% of M142 carriers had PrPC\textsuperscript{Sc} in the brain as opposed to only 35% of isoleucine homozygotes (II142) (Gonzalez et al. 2010). Accumulation of PrPC\textsuperscript{Sc} in placentas from goats with scrapie has been reported to be relatively lower when compared with what has been observed in the placentas of scrapie-positive sheep (O’Rourke et al. 2011).

Atypical Scrapie in Goats

Atypical scrapie also has been described in goats (Le Dur et al. 2005; Seuberlich 2007). Similar to sheep, a histidine substitution at 154 is a risk factor for atypical scrapie in goats (Colussi et al. 2008), and PrPC\textsuperscript{Sc} has not been demonstrated in the lymphoid tissues of affected animals (Seuberlich 2007).

Bovine Spongiform Encephalopathy

What is now known as classical bovine spongiform encephalopathy (BSE; mad cow disease) was first diagnosed in the United Kingdom in 1985 (Wells et al. 1987). Cattle with BSE are symptomatic with neuropsychologic signs of incoordination and weight loss, and they demonstrate unusual aggression in some cases (Wilesmith 1988). The origin of BSE remains unknown. Early theories suggested that BSE was the result of passage of a scrapie-like disease into the cattle population (Wilesmith et al. 1988), potentially via the use of ruminant-derived meat and bone meal (MBM) as a food source. However, studies conducted in the United States and United Kingdom have shown that scrapie does not transmit to cattle by oral routes of inoculation (Cutilip et al. 2001) and, when it does transmit by intracranial inoculation, the resultant disease is distinguishable from BSE by clinical signs, molecular profile of PrPC\textsuperscript{Sc}, and PrPC\textsuperscript{Sc} deposition patterns in brain sections (Cutilip et al. 1994, 1997; Konold et al. 2006). Preliminary evidence from laboratory animal models suggests that, after multiple passages, H-type atypical BSE (defined below), of presumed sporadic origin, can assume a classical BSE phenotype (Bencsik et al. 2013). Whatever the initial infectious source, the widespread BSE outbreak in the United Kingdom was due, in large part, to the feeding of MBM to cattle. MBM contains central nervous system tissues and other tissues that are known to contain the abnormal prion protein.
Prior to the identification of the epidemic, many cattle with BSE were being slaughtered, with offal entering MBM, thus facilitating distribution of infectious material (Wilesmith et al. 1991). Elimination of BSE focuses on preventing tissues with highest risk of causing infection (called specified risk materials or SRM) from getting into the human or animal food supply. In the United States, SRM includes brain, skull, spinal cord, trigeminal and dorsal root ganglia, eyes, and vertebral columns of cattle greater than 30 months of age, and tonsil and distal ileum of all cattle (U.S. Department of Agriculture 2014). Surveillance efforts are focused on cattle most likely to have BSE: dead, diseased, immobile, or disabled with neurologic signs and those greater than 30 months of age (U.S. Department of Agriculture 2014).

BSE can be subdivided into at least three strains: classical, H-type, and L-type. The H- and L- designations are based on higher or lower apparent molecular mass profiles of the unglycosylated PrPSc band on a western blot (Figure 3), and are collectively referred to as atypical BSE (Wells 2007). The vast majority of BSE cases are the classical subtype that was associated with the UK BSE epizootic.

Classical BSE

Classical BSE was the first cattle TSE to be recognized (Wells et al. 1987). Classical BSE is acquired by consumption of contaminated foodstuffs. Epidemiological studies suggest dietary supplements, in particular MBM containing CNS tissues with PrPSc, are a source of the outbreak in the United Kingdom (Wells et al. 1991). Studies in Great Britain have not detected vertical (Wilesmith et al. 2009), the Netherlands (Biacabe et al. 2007), Poland (Biacabe et al. 2004), Germany (Buschmann et al. 2006), Japan (Sugiura et al. 2004), the Netherlands (Biacabe et al. 2007), Poland (Biacabe et al. 2004), Switzerland (Tester et al. 2009), the United Kingdom (Sohn et al. 2009), and the United States (Richt et al. 2007). The molecular phenotype of H-BSE cases on western blot (Figure 3) is characterized by (1) a higher molecular mass of the unglycosylated PrPSc isoform; (2) a strong labeling of all 3 PrPSc polypeptides (unglycosylated, monoglycosylated, and diglycosylated forms of the prion protein). Low-type BSE (BSE-L) in lane 1 is the lowest molecular weight compared with high-type BSE (BSE-H, lane 2) or classical BSE, which has an intermediate molecular weight (lane 3). There is no detectable PrPSc in the brain of the negative control animal (lane 4). The different molecular weights of BSE-L, BSE-H and classical BSE are due to differential cleavage of the n-terminal region of the abnormal prion protein.

Atypical BSE in Cattle

Several hypotheses have been proposed to explain the etiology of atypical BSE cases. At the forefront of these is the possibility that both H-type and L-type BSE may arise spontaneously in cattle. This is drawn from atypical BSE’s parallels to sporadic CJD in humans, specifically, it’s prevalence in older hosts and a comparable low incidence (Tramulis et al. 2011). The fact that the number of cattle affected by classical BSE has fallen precipitously in response to control measures, while the number of cattle affected by atypical BSE remains relatively stable over time, supports the hypothesis that these atypical cases occur spontaneously (Brown et al. 2006). Furthermore, prion gene promoter region polymorphisms that influence susceptibility to classical BSE do not appear to influence atypical BSE (Brunelle et al. 2007), and atypical BSE occurs as isolated cases in contrast to the clustering of cases observed for feedborne classical BSE. Finally, atypical BSE cases have occurred predominantly in cattle greater than 10 years of age, whereas classical cases have occurred in much younger animals (Biacabe et al. 2004; Casalone et al. 2004).

H-type BSE has been described in cattle from France (Biacabe et al. 2004), Germany (Buschmann et al. 2006), Japan (Sugiura et al. 2009), the Netherlands (Biacabe et al. 2007), Poland (Biacabe et al. 2007), Switzerland (Tester et al. 2009), the United Kingdom (Sohn et al. 2009), and the United States (Richt et al. 2007). The molecular phenotype of H-BSE cases on western blot (Figure 3) is characterized by (1) a higher molecular mass of the unglycosylated PrPSc isoform; (2) a strong labeling of all 3 PrPSc polypeptides (unglycosylated, monoglycosylated, and diglycosylated iso forms) with the PrP-specific monoclonal antibodies that bind to the N-terminus (such as mAb p4) (Biacabe et al. 2004); and (3) a second unglycosylated band at approximately 14 kDa when developed with antibodies that bind in the C-terminal region (amino acids 154–236), such as SAF 84 (Biacabe et al. 2007). Brains of animals inoculated with H-type BSE have similar spongiform lesions at the level of the obex, but have more vacuolation in the frontal cortex than occurs in the brains of cattle similarly challenged with classical BSE (Konold et al. 2012). Immunoreactivity for PrPSc in brains of cattle inoculated with BSE-H is distinct from that of classical BSE, as well as BSE-L, the other form of atypical BSE (Greenlee, Smith et al. 2012; Okada et al. 2011).

L-type BSE has been found in cattle in Italy (Casalone et al. 2004), Japan (Yamakawa et al. 2003), Germany (Buschmann et al. 2006), Belgium (De Bosschere et al. 2004), and Canada.
Experimental Interspecies Transmission

Prion diseases can be transmitted from one species to another. Experimental cross-species transmission of TSE agents provides valuable information about potential host ranges and differentiation from known TSEs and may lead to clues about their origins. The most common routes of inoculation to test for interspecies transmission are oral and intracranial (IC). Oral inoculation is generally effective in intraspecies transmission studies, is considered a natural route by which animals acquire TSEs, and best mimics natural disease pathogenesis. Intracranial inoculation is very efficient for testing whether a new host has any potential to acquire a TSE from a given species since it requires smaller dosage of inoculum and results in higher attack rates and reduced incubation periods than does oral inoculation. A species resistant to a TSE by IC inoculation would have negligible potential for successful oral transmission.

Unsuccessful attempts at interspecies transmission led to the concept of a species barrier. The species barrier phenomenon limits transmission of prions between different mammalian species and is influenced by mismatches between host and recipient prion amino acid sequence and the resulting structures and folding (Beringue et al. 2008; Collinge and Clarke 2007; Hill and Collinge 2001, 2004; Moore et al. 2005; Scott et al. 1993; Vanik et al. 2004). The species barrier, even in closely related species, can manifest as complete lack of susceptibility, incomplete attack rates, or prolonged incubation times. Primary passage is generally not efficient between species, and sequential passages are required for a TSE strain to stabilize in a new species (Hill and Collinge 2004). To fully assess potential risks to animal and human health, it is important to study interspecies transmission of scrapie and BSE in addition to the other known animal TSEs: chronic wasting disease (CWD) and transmissible mink encephalopathy (TME). Furthermore, species barrier can be studied through the use of transgenic mice expressing the PRNP of the species of interest or in vitro conversion assays (discussed below).

Chronic Wasting Disease (CWD)

Naturally occurring CWD has been documented in mule deer (Odocoileus hemionus hemionus), black-tailed deer (Odocoileus hemionus columbianus), white-tailed deer (Odocoileus virginianus), Rocky Mountain elk (Cervus elaphus nelsoni), and moose (Alces alces shirasii) (Baeten et al. 2007; Williams 2005). The disease was first recognized in a captive population of mule deer at the Colorado Division of Wildlife Foothills Wildlife Research Facility in Fort Collins in 1967, although identification of the disease as a TSE did not occur until 1978 (Williams and Young 1980). The origins of CWD are uncertain, but white-tailed deer appear to be readily susceptible to scrapie of North American origin (Greenlee et al. 2011).

Transmissible Mink Encephalopathy (TME)

Transmissible mink encephalopathy was identified in ranched mink (Neovison vison). It was first documented in Wisconsin, in 1947 (Eckroade et al. 1973), and the last reported outbreak in the United States was in 1985 (Marsh et al. 1991). TME is a foodborne disease that has been experimentally transmitted to a variety of animal species (Eckroade et al. 1970, 1973). The origin of TME is unknown, but has been associated with feeding downer cattle to farmed mink (Marsh and Hartsough 1988). Molecular similarities and results of rodent studies suggest a high degree of similarity between TME in cattle and BSE-L (Baron, Bencsik et al. 2007).

Experimental Transmission of Bovine and Cervid TSEs to Sheep and Goats

Numerous experimental studies have been done to better understand the potential transmission of BSE, CWD, and TME to sheep and goats. The likelihood that sheep were exposed to the same contaminated feedstuffs as cattle during the UK BSE epizootic and detection of a natural case of BSE in a goat (Elliott et al. 2005) heightened concern about the potential transmission of BSE to small ruminants. Sheep are readily infected with the agent of BSE by the oral route, with the highest susceptibility prior to weaning (Hunter et al. 2012), and resulting in infectivity through-out lymphoid tissues (Foster et al. 1993; Foster, Parnham, Hunter et al. 2001). In most cases, sheep have PrPSc accumulation in tissues similar to those infected with scrapie, but some sheep develop neuroinvasion without accumulation of PrPSc in lymphoid tissues (Jeffrey, Ryder et al. 2001). Maternal and contact transmission of BSE to sheep appears to be quite inefficient (Foster et al. 2004), although natural transmission to offspring born to experimentally infected ewes can occur (Bellworthy et al. 2005). Strain-specific processing of misfolded PrP within the nervous system and lymphoid tissues leads to differences in immunohistochemical staining patterns depending on the antibodies used, which allows for differentiation of BSE from scrapie in sheep (Jeffrey, Martin et al. 2001). Similarly, western blotting can be used to differentiate BSE from scrapie in sheep by molecular weight, glycoform ratio, and different apparent N terminal cleavage point after PK digestion using either brain (Stack et al. 2002) or lymph...
node samples (Langeveld et al. 2006). The incubation period of BSE in sheep is genotype dependent and, in contrast to sheep with scrapie, ARQ/ARQ and AHQ/AHQ sheep have the shortest incubation times (Foster, Parnham, Chong et al. 2001), with a V136 allele greatly increasing the incubation time (Goldmann et al. 1994). Sheep carrying a R171 allele are not entirely resistant to BSE, but do have prolonged incubation periods (Foster, Parnham, Chong et al. 2003). Only the M to T substitution at codon 112 has been shown to protect against challenge with BSE (Saunders et al. 2009).

Attempts to transmit the agent of CWD from mule deer to sheep by intracranial inoculation resulted in PrPSc accumulation in only two of eight inoculated lambs (Hamir, Kunkle, Cutlip et al. 2006). PrPSc was observed in the brain and tonsil of one ARQ/ARQ sheep euthanized at the end of the 72-month study without developing clinical signs. One ARQ/VRQ sheep developed clinical signs and was euthanized at 35 months post-inoculation with extensive spongiform lesions throughout the brain, and PrPSc was demonstrated throughout the nervous and lymphoid systems, suggesting that sheep with the V136 codon should be further investigated in their potential susceptibility to CWD (Hamir, Kunkle, Cutlip et al. 2006).

Transmission of the agent of transmissible mink encephalopathy to sheep and goats by intracranial inoculation has been documented (Hadlow et al. 1987; Marsh et al. 1969; Zlotnik and Barlow 1967). Incubation times ranged from 45 to 80 months in sheep and 31 to 40 months in goats (Hadlow et al. 1987). A mink bioassay (sheep tissues intracranially inoculated into a mink host) was used to demonstrate infectivity in lymph node and spleen material (Marsh et al. 1969). These studies were conducted prior to the ability to determine the PRNP genotype of sheep and goats, so no information on the effect of genotype on susceptibility or resistance is available for TME.

**Experimental Transmission of Sheep and Cervid TSEs to Cattle**

Interspecies transmission of TSEs to cattle has been extensively studied (Hamir et al. 2011). During the 1990s, the possibility that U.S. strains of sheep scrapie might cause BSE in cattle was assessed experimentally. Intracranial inoculations resulted in 100% transmission of scrapie to cattle between 14 and 18 months following inoculation (Cutlip et al. 1994). A separate study using multiple simultaneous routes of inoculation (including IC) found only 20%-40% transmission of scrapie to cattle depending on the source of inoculum and a longer incubation period of 24 to 48 months prior to affected cattle exhibiting anorexia, weight loss, leg and back stiffness, incoordination, and rear leg weakness, eventually leading to severe lethargy and ataxia (Clark et al. 1995). Following oral exposure to the scrapie agent, cattle did not develop symptoms of neurological disease, spongiform lesions, or PrPSc deposits in the CNS by the time of examination 8 years after inoculation (Cutlip et al. 2001). In contrast, oral inoculation of the agent of BSE into cattle is a highly efficient means of transmission (Wells et al. 2007). Scrapie in cattle can be differentiated from BSE microscopically: while there is no evidence of spongiform changes in the brains of scrapie-affected cattle, spongiform changes are usually observed in clinical BSE cases (Scott et al. 1990). Further, immunoreactivity for PrPSc in scrapie-affected cattle was observed predominantly in neuronal cell bodies with relatively little accumulation in the neuropil (Cutlip et al. 1994, 1997), in contrast to BSE where there is a diffuse distribution of PrPSc in the CNS (Scott et al. 1990). Despite the proposed linkage of the BSE epidemic to feed contamination by a scrapie-like agent (Wilesmith et al. 1988), scrapie isolates have only been transmitted to cattle by IC inoculation, and the pathology and clinical disease differed from both BSE in cattle and scrapie in sheep (Cutlip et al. 1994, 1997; Konold et al. 2006). Therefore, current experimental evidence from scrapie transmission studies into cattle does not support the hypothesis that the U.K. BSE epidemic originated from the feeding of scrapie PrPSc to cattle, however, only a limited number of scrapie isolates have been thoroughly tested in cattle.

The recognition of CWD (Williams 2002) in captive and free-ranging cervids in the United States raised questions about the possible transmissibility to other ruminant species that share the same pastures or range. Studies have been done to investigate the potential of cattle to serve as a host to the agent of CWD derived from mule deer, white-tailed deer, and elk sources. IC inoculation of cattle with the CWD agent from mule deer resulted in accumulation of PrPSc in the CNS, indicating that amplification of the abnormal CWD prion had occurred, although there was only subtle spongiform change in the brain (Hamir et al. 2001). The initial study demonstrated a low (38%) attack rate of mule deer CWD upon first passage in cattle (Hamir et al. 2005), but a second IC passage of mule deer CWD transmitted the disease to all inoculated cattle within 16.5 months after inoculation (Hamir, Kunkle, Miller, Greenlee et al. 2006). IC inoculation of the agent of CWD from white-tailed deer into cattle showed that the white-tailed deer inoculum had a higher attack rate (86%) in cattle than the mule deer CWD inoculum used previously, however, spongiform change in the brain was not observed (Hamir et al. 2007). None of the cattle given the same inoculum orally (50 g of pooled brain/animal) has shown any evidence of prion disease up to 9 years after inoculation (Williams 2005), however, calves were not inoculated until 4 months of age. IC inoculation of cattle with the CWD agent from elk also resulted in a low rate of transmission: clinical signs (poor appetite, weight loss, circling, and bruxism) occurred in 2 out of 16 cattle at 16 and 17 months post-inoculation with PrPSc, but no spongiform lesions were detected in the CNS (Greenlee, Nicholson et al. 2012). Regardless of CWD inoculum source, deposition of PrPSc appears to be similar and clearly distinguishable from BSE or scrapie in cattle: immunoreactivity is prominent in midbrain, brainstem, and hippocampus, with lesser immunoreactivity in the cerebral spinal cord that occurs in multifocal and distinct aggregates confined to glial cells and associated neuropil. Additional studies are required to fully assess the potential for cattle to develop CWD through a more natural route of exposure, but low attack rates after IC inoculation and failure of transmission following oral exposure suggests that the risk of transmission through routes other than IC is low.

Inoculation of cattle with the agent of TME from three different sources led to clinical disease and severe spongiform encephalopathy (Robinson et al. 1995), confirming earlier reports of TME transmission to cattle (Marsh et al. 1991). Subsequent IC inoculation of cattle with first and second passage cattle-adapted TME confirmed the earlier findings and also described for the first time the immunohistochemical and western blot characteristics (lower molecular weight of cattle-adapted TME vs. C-type BSE by western blot) of the accumulated PrPSc, which is dissimilar from experimental scrapie and experimental CWD in cattle (Hamir, Kunkle, Miller, Bartz et al. 2006). Studies conducted in ovinized transgenic mice demonstrated that cattle-passaged TME presented with the same phenotypic characteristics as atypical C-type BSE (Baron, Bencsik et al. 2007). However, experimental studies investigating the oral route of transmission of TME to cattle have not been reported.
Transgenic mice that have been engineered to express either the bovine PRNP sequence (Buschmann and Groschup 2005; Castilla et al. 2003) or ovine PRNP sequences (Kupfer et al. 2007; Vilotte et al. 2001) have greatly enriched interspecies transmission studies of scrapie and BSE. While a thorough discussion of the body of work using transgenic mice is outside the scope of this review, it is worth making note that transgenic mice have been a useful tool in understanding the pathogenesis of scrapie and BSE. Examples include the use of ovinized mice to characterize atypical scrapie (Griffiths et al. 2010) or to resolve scrapie cases with a BSE-like molecular profile (Beck et al. 2012) and the use of bovinized mice to assess potential infectivity of tissues from animals with BSE (Fast et al. 2013) or the potential of multiple passages of atypical BSE to promote conversion to a classical BSE phenotype (Torres et al. 2011).

TSEs of Livestock And Human Health

Our understanding of the risks of animal TSEs to human health comes from several lines of evidence. In addition to epidemiological studies, the potential for an animal TSE to be transmitted to humans has been experimentally evaluated in two different ways. The first is via the experimental use of “humanized: transgenic mice, where the mouse prion protein gene (PRNP) has been knocked out, and the mice instead express the human PRNP gene (Kitamoto et al. 1996). The second approach that has been used to evaluate the potential of an animal prion disease to affect humans is the in vitro conversion assay protein-misfolding cyclic amplification (PMCA, see below), where human brain homogenate is used as a substrate (Jones et al. 2007; Levavasseur et al. 2014).

Scrapie

Considerable evidence suggests that scrapie does not pose a risk to human health. Despite the presence of scrapie throughout much of the world for nearly 300 years, there is no evidence that scrapie has ever been transmitted to humans. In former times, it was a widespread management practice to slaughter sheep with clinical signs for human consumption (Schneider et al. 2008), but no increase in neurologic disease has been associated with consuming products from sheep. A number of studies have demonstrated that transgenic mice expressing the human prion are not susceptible to classical (Gombojav et al. 2003) or atypical scrapie (Wadsworth et al. 2013; Wilson et al. 2012). Further, using human brain or mouse brain expressing human PRNP as substrate in PMCA conversion assays fails to demonstrate replication of scrapie prions (Jones et al. 2007). However, recent results demonstrated transmission of classical scrapie strains to mice overexpressing the human prion protein, resulting in a molecular phenotype with similarities to human sporadic Creutzfeldt-Jakob disease (Cassard et al. 2014), indicating the need for continued research in this area to clarify any potential risks of scrapie to human health.

Bovine Spongiform Encephalopathy

Unlike scrapie, BSE is a known risk to human health. Due to conservation of molecular phenotype upon transmission of BSE to multiple species including mice expressing the human prion protein it is accepted that variant Creutzfeldt-Jakob disease (vCJD), which emerged in the United Kingdom in the mid-1990s, is caused by the ingestion of contaminated beef from cattle with BSE (Bruce et al. 1997; Collinge et al. 1996; Hill et al. 1997; Will et al. 1996). In vitro conversion assays demonstrate that BSE can convert PrPSc to PrPSc in human substrate much more efficiently than other animal TSEs (Barria et al. 2014). Susceptibility to vCJD is greatly affected by a polymorphism at codon 129 in the human PRNP, which can be either methionine or valine (Collinge et al. 1991). All diagnosed cases of vCJD have been homozygous MM at codon 129 (Saba and Booth 2013). Experiments in humanized mice further support the susceptibility conferred by this polymorphism (Wadsworth et al. 2004). Quite interestingly, a recent large-scale survey of human appendices removed in the United Kingdom identified abnormal PrPSc in 16 of 32,441 samples. The genotypes of the positive specimens were a high proportion VV at codon 129 (Gill et al. 2013) raising the possibility that the 129 genotype affects incubation time, rather than absolute susceptibility. Classical foodborne BSE appears to be the primary risk to human health, as atypical forms of BSE are not transmissible to humanized mice (Kong et al. 2008; Wilson et al. 2012).

Diagnosis of TSEs in Livestock

Definitive diagnosis of prion disease depends upon detection of misfolded prion protein (PrPSc) in the diseased animal. There is a significant effort to develop antemortem tissue collection and detection techniques; however, the current gold standard for diagnosis is the postmortem detection of PrPSc in brain tissue (usually brain stem at the level of the obex) by enzyme-linked immunosorbent assay (ELISA), immunohistochemistry, or western blot (NCBA 2014). Current diagnostics that are based on detection of PrPSc perform well on clinically affected animals for all described strains of BSE (Gray et al. 2012). The major diagnostic challenge is detecting infected animals in the preclinical stage.

Emphasis for the past decade has been on the development and optimization of techniques to amplify misfolded PrPSc in a diagnostic sample so that it can be detected ( Orru et al. 2012). Two basic approaches include PMCA (Saa et al. 2006), which uses brain homogenate as a substrate that is “seeded” by the diagnostic sample, and Quaking-Induced Conversion (QuIC), which uses recombinant protein as the substrate to be seeded by misfolded protein in a diagnostic sample (Atarashi et al. 2008). PMCA and QuIC have been used to detect PrPSc in biological samples including blood (Lacroix, Comoy et al. 2014), urine (Moda et al. 2014), and saliva (Okada et al. 2012).

Scrapie

Sheep and goats infected with scrapie accumulate PrPSc in the lymphoreticular system allowing the opportunity for antemortem sampling and testing by immunohistochemistry (Figure 4) or ELISA. Biopsy of rectal mucosa (Gonzalez, Dagleish et al. 2008), tonsil (Schreuder et al. 1998), or third eyelid (O’Rourke et al. 2000) holds promise for antemortem detection of infected animals (Dennis et al. 2009; Monleon et al. 2011). The sensitivity of immunohistochemistry to detect PrPSc in rectal mucosal biopsies from field samples of preclinically affected sheep has been reported to be as high as 89.4% (Dennis et al. 2009), although others report a more modest result of 48% (Monleon et al. 2011). A study of goats with natural scrapie reported that only 42% of preclinical scrapie positive goats had positive rectal mucosal biopsies (Gonzalez et al. 2009), suggesting that rectal biopsy of goats may be of lower sensitivity than for sheep.

Bovine Spongiform Encephalopathy

There is much less accumulation of PrPSc in the periphery of cattle infected with BSE when compared with sheep and goats with
scrapie. Thus, techniques for early detection of scrapie-infected sheep and goats (i.e., rectal biopsy) cannot be applied to preclinical detection of cattle infected with BSE. In studies of cattle orally challenged with BSE, immunoreactivity for PrPSc has been reported in the ileal Peyer’s patches 4 to 6 months post challenge (Terry et al. 2003) and in the palatine tonsil starting at 10 months after the challenge (Wells et al. 2005). This is much earlier than the first detection of PrPSc immunoreactivity in the brainstem at 27 months post challenge (Espinosa et al. 2007). During the clinical phase of BSE in cattle, however, there is a centrifugal spread of abnormal prions to the periphery (Buschmann and Groschup 2005; Franz et al. 2012), which suggests peripheral tissues may be useful for diagnosis of clinically ill animals.

Various studies have used samples from cattle experimentally challenged with BSE to further define the tissue distribution of prions in infected cattle (Bannach et al. 2013; Murayama et al. 2010). However, there is not, as yet, a validated antemortem test that can be used to screen cattle for BSE. A careful behavioral assessment of experimentally inoculated cattle demonstrates that numerous nonspecific behavioral changes can be observed prior to the clinical phase of BSE. Difficulty rising is the most consistent feature of BSE-challenged animals (Konold et al. 2012). While often observed months prior to the end stage of the disease, this behavioral feature did not have a consistent time course or onset (Konold et al. 2012).

Our group has described changes in retinal function and morphology in cattle experimentally inoculated with TSEs (Greenlee, Smith et al. 2012; Smith and Greenlee 2014; Smith et al. 2008; Smith, Greenlee, Hamir, Greenlee 2009; Smith, Greenlee, Hamir, Richt et al. 2009). The retina is a part of the CNS, and accumulates PrPSc in experimentally and naturally occurring cases of prion disease in sheep and cattle (Greenlee et al. 2006; Hortells et al. 2006; Okada et al. 2011; Smith, Greenlee, Hamir, Richt et al. 2009). Using electroretinography (ERG) and optical coherence tomography (OCT) (Figure 5) we have demonstrated that changes in retinal function and retinal thickness are detectable from 5 to 19 months prior to BSE-inoculated cattle displaying clinical signs of disease (West Greenlee et al. 2015).

To understand the impact that detection of PrPSc accumulation may have on public health, Arnold and colleagues estimated how the time course of PrPSc detection in cattle related to the incubation period in cattle experimentally challenged with BSE. Based on their data, they estimate that, in cattle orally challenged with 100 g of infectious material, PrPSc accumulation is detectable in the brainstem 9.6 months prior to clinical disease. In cattle orally challenged with 1 g of infectious material, however, it was estimated that PrPSc would be detectable in the brainstem 1.7 months prior to the clinical onset of disease (Arnold et al. 2007).

Figure 4 Biopsy of rectal mucosa in sheep and goats can detect PrPSc accumulation in live animals prior to onset of clinical illness. Mucosa-associated lymphoid follicles are strongly immunoreactive for PrPSc (red reaction product) in a sheep with scrapie. Scale bar = 100 µm.

Animal Models of TSEs

The TSEs are a family of fatal, infectious neurodegenerative human and animal diseases that share a common mechanism of misfolding of the prion protein. The strong similarities between human and animal prion diseases offer advantages to biomedical research. Whereas human prion diseases are often diagnosed late in the course of disease after the onset of clinical signs and beyond the point of any possibility of productive intervention, prion diseases in animals have defined incubation periods, progression of lesions, and genotype susceptibilities that can be used to design prospective studies that may lead to methods of prevention or cure for prion diseases. There is a high likelihood that results of animal studies will translate to human medicine through improved diagnostics, prevention strategies, and policies to improve public health.

Research to understand the animal prion diseases and the potential for interspecies transmission has lead to major policy changes to protect public health. Understanding the pathogenesis of BSE, including the tissue distribution of BSE prions, is critical to identifying SRM, which allows for removal of those tissues from the human and animal food supplies. Interspecies transmission studies in ruminants or using ruminant-derived prions has helped identify prion isolates with zoonotic risk and others that deserve further study to fully understand the potential zoonotic risk (Beringue et al. 2008; Hamir et al. 2011).

Ruminants have been used extensively to study the transmission and differentiation of various TSEs. The identification of the E211K PRNP polymorphism in cattle and it’s association with H-type BSE (Greenlee, Smith et al. 2012) may be another another animal model that can contribute to our understanding of hereditary forms of TSEs. Besides the potential to study the onset of spontaneous disease late in life, inoculation of E211K cattle with H-type BSE leads to clinical signs in approximately 10
months, which greatly reduces the duration normally required to conduct TSE studies in large animals.

Prion diseases are related to other neurodegenerative protein misfolding disorders including Parkinson’s disease, Alzheimer’s disease, ALS, and Huntington’s disease (Aguzzi and O’Connor 2010; Prusiner 2012). Common features like self-propagation of misfolded protein and spread throughout neuroanatomic regions of the brain suggest that TSE models (rodent and ruminant) of disease and successful TSE treatment strategies may translate to related neurodegenerative disorders.

Conclusions

Our understanding of TSEs has advanced tremendously since prions were first reported to be the disease-causing agent of scrapie in sheep and goats. Misfolding of the prion protein results in similar diseases in cattle, cervids, humans, and other mammalian species. TSEs are transmissible within a species by exposure to or ingestion of infectious material, but the likelihood of transmission between species varies with the TSE. For example, scrapie does not appear to be transmissible to humans, but consumption of BSE contaminated beef can result in developing vCJD.

This review highlighted scrapie and BSE, the natural TSEs of small ruminants (sheep and goats) and cattle, respectively. A major difference between scrapie and BSE is host-related differences in distribution of PrPSc accumulation within an animal. Sheep and goats have widespread accumulation of PrPSc throughout the lymphoreticular system, while cattle have very limited accumulation of PrPSc outside of the CNS. For example, scrapie does not appear to be transmissible to humans, but consumption of BSE contaminated beef can result in developing vCJD.

Although much has been learned about scrapie and BSE, there is still a role for livestock in the study of TSEs moving forward. As new strains of these TSEs emerge, work to determine their interspecies transmission will be necessary to continuously reevaluate their risk to the food supply. In addition, TSEs of livestock are a natural, large-animal model of a protein-misfolding disease. Thus, they can provide a valuable step between mouse and human studies on intervention strategies for a whole family of protein misfolding disorders.

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