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Transmissible Spongiform Encephalopathies

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Edited by

Ray Bradley, CBE
Central Veterinary Laboratory,
Weybridge, United Kingdom

and

Brian Marchant
Commission of the European Communities
Brussels, Belgium

SPONGIFORM ENCEPHALOPATHY IN CAPTIVE WILD ANIMALS IN BRITAIN: EPIDEMIOLOGICAL OBSERVATIONS

James K Kirkwood & Andrew A Cunningham

Institute of Zoology,
Regent's Park,
London NW1 4RY, UK.

ABSTRACT

Since 1986, scrapie-like spongiform encephalopathy SE has been diagnosed in 18 captive wild animals of eight species at or from seven zoological collections in the British Isles. The affected animals included members of the subfamily bovinae: one nyala *Tragelaphus angasi*, four eland *Taurotragus oryx*, six greater kudu *Tragelaphus strepsiceros*; members of the subfamily hippotraginae: one gemsbok *Oryx gazella*, one Arabian oryx *Oryx leucoryx*, one scimitar-horned oryx *Oryx dammah*; and members of the family felidae: three cheetah *Acinonyx jubatus* and one puma *Felis concolor*. In addition to these, three cases of an SE of unknown aetiology have been reported in ostriches *Struthio camellus* from two zoos in North West Germany. Three features suggest that some of these cases may have been caused by the BSE agent. First, they have been temporally and geographically coincident with the BSE epidemic. Second, in all the ungulate cases for which details are available, it is possible that either the affected animal itself, or the herd into which it was born or moved, had been exposed to proprietary feeds containing ruminant-derived protein (RDP) or other potentially contaminated material, and in all the carnivore cases there was a history of feeding parts from cattle carcases judged unfit for human consumption. Third, the results of mouse inoculation tests using a homogenate of fixed brain tissue from the nyala and from one greater kudu were similar to results of BSE-brain inoculations into mice. All the kudu cases occurred in the small herd kept by the Zoological Society of London. The pattern of incidence in these animals suggests that the disease may have entered the herd through feed containing RDP but that, in contrast to the epidemiology of BSE in cattle, transmission between animals occurred subsequently.

INTRODUCTION

Prior to 1980, naturally occurring scrapie-like subacute spongiform encephalopathies (SE) were known in 6 species. These diseases included: scrapie itself in domestic sheep and goats (Dickinson, 1976), kuru and other spongiform encephalopathies in man (Prusiner and Hadlow, 1979), transmissible mink encephalopathy in mink *Mustela vison* (Hartsough and Burger, 1965), and chronic wasting disease in mule deer *Odocoileus hemionus* (Williams and Young, 1980) and Rocky Mountain elk *Cervus elaphus* (Williams and Young 1982). Since 1980, spongiform encephalopathies have been reported in a variety of other species, especially of wild animals maintained in zoos and wildlife parks in Great Britain (Wells & McGill, 1992; Bradley & Matthews, 1992).

Here we present information on all the recent cases of SE in captive wild animals of which we are aware. These total 18 cases in eight species at or from seven zoological collections in the British Isles. An SE has also been reported in ostriches in Germany (Schoon et al, 1991). In view of the temporal and geographic coincidence of some these zoo- animal cases with the (BSE) epidemic, the possibility that at least some of these could have been caused by the agent of bovine spongiform encephalopathy BSE has been considered (Kirkwood et al, 1990; 1993a, Wells & McGill, 1992; Bradley & Matthews, 1992). However other explanations for this cluster of cases could be envisaged. For example, increased vigilance as a result of the BSE epidemic could have lead to diagnosis of SE's in species or groups in which the disease may have been present but previously undetected. In this context, however, it is pertinent to note that the diagnosis of the first SE case in a zoo animal (Jeffrey & Wells, 1988) predated the diagnosis of BSE in cattle.

In this paper we review some epidemiological and other features of all the cases in captive wild animals in or from collections in Britain that have been reported to us. Accounts of some of these cases have been

published previously, but we also include information on some which have not as yet been described in the literature. Pathological findings have been presented in many of the original reports and in some reviews (Wells & McGill, 1992) and we have not discussed these here. We conclude with some considerations on the control of the disease in wildlife collections.

We have not included in this review, the cases of spongiform encephalopathy that were reported in white tigers that had died at the Bristol Zoo between 1970 and 1977 (Kelly and others, 1980). It is thought that these cases differed from the scrapie-like spongiform encephalopathies that were subsequently recognised in other species because of differences in the histopathological lesions and because transmission to mice was unsuccessful (Kelly, personal communication). Nor have we included here details of cases of SE that have been diagnosed and recently reported in mouflon *Ovis musimon* in British zoos (Wood et al, 1992). The mouflon is an ancestor of domestic sheep and these authors assumed that the SE was scrapie.

METHOD

Information has been collected on epidemiological aspects of 18 cases of SE in captive wild animals in the British Isles and three from Germany, both from the literature and from discussion with veterinarians and curators involved with the cases. Data collected included dates of birth and death, history of feeding practices, and details of any movements between collections and contact with animals of the same or other species. Brief details of the cases are presented in Table 1. In all these cases, SE had been diagnosed on the basis of the histopathology and, in some cases, by the detection of scrapie-associated fibrils SAF and/or the detection of the disease-specific isoform of the glycoprotein PrP (Wells & McGill, 1992), and by experimental transmission to mice (Table 2)(Jeffrey et al, 1992; H Fraser, personal communication).

THE CASES

Nyala *Tragelaphus angasi*

The diagnosis of the scrapie-like spongiform encephalopathy in the Nyala (Tables 1 and 2) pre-dated the diagnosis of the first cases of BSE in cattle (Jeffrey & Wells, 1988). Although the clinical and pathological findings of this case have been described previously (Jeffrey & Wells 1988), little information relevant to enquiry into the epidemiology of the disease have hitherto been available. Notes on the clinical signs are listed in Table 3.

This animal was hand-reared on cow or goat milk and weaned onto a diet of which part was a proprietary concentrate pellet. This pellet was not thought to contain RDP either prior to March 1986 or after March 1987. If this was so, the nyala was only exposed to RDP during the 3 months preceding her death. It seems very unlikely that the incubation period could be this short. Perhaps the diet had unwittingly been contaminated with RDP prior to March 1986, or perhaps the animal acquired the disease from another source. No other source is, however, apparent. The nyala herd which numbered 16 animals at the time of this female's birth, was not in direct contact with any other ungulates. Sheep were kept at the premises where the animal was hand-reared but there had been no cases of scrapie in these. The results of bioassay of brain material from the nyala suggested the agent was like that of BSE (see below).

Table 1 Cases of scrapie-like spongiform encephalopathy in captive wild animals in Britain

Species	Animal	DOB	DOD	Age months	Sex	Source
Nyala		4.9.83	18.6.86	33	F	Jeffrey & Wells 1988, P. Bircher, pers comm
Gemsbok		18.6.83	8.6.87	48	F	Jeffrey & Wells 1988, P. Bircher, pers comm
Eland	1	6.4.87	20.12.89	32	M	Fleetwood & Furley 1990
Eland	Molly	14.1.89	9.4.91	27	F	M. Hosegood pers comm
Eland	Neddy	10.1.89	25.5.91	28	M	M. Hosegood pers comm
Eland	Electra	12.1.90	23.1.92	24	F	M. Hosegood pers comm
Arabian oryx		12.1.86	24.3.89	38	F	Kirkwood *et al* 1990
Greater kudu	Linda	11.2.87	18.8.89	30	F	Kirkwood *et al* 1990
Greater kudu	Karla	19.4.89	13.11.90	19	F	Kirkwood *et al* 1992
Greater kudu	Kaz	14.5.88	6.6.91	37	M	Kirkwood *et al* 1993a
Greater kudu	Bambi	29.10.88	24.10.91	36*	M	Kirkwood *et al* 1993a
Greater kudu	346/90	26.8.90	26.2.92	18*	M	Kirkwood *et al* 1993a
Greater kudu	324/90	5.8.89	22.11.92	39	F	Kirkwood *et al* 1993b
Scimitar-horned oryx		12.7.90	29.1.93	30	F	D. Lyon pers comm
Puma		8.3.86	16.5.91	62	F	Willoughby *et al* 1992
Cheetah	1	16.6.86	7.1.92	55	M	Peet & Curran 1992 P. Bircher pers comm
Cheetah	Duke	3.9.84	27.10.92	96	M	J. Lewis, pers com, P. Bircher, pers comm
Cheetah	Saki	23.2.86	4.5.93	86	F	S. McKewan pers comm

* these animals showed no clinical signs at time of euthanasia

Table 2 Features on which diagnosis of SE was based in captive wild animal cases

Species	Animal	Histopath	SAF	PrP	Transmission	Source
Nyala		+			+	Jeffrey & Wells 1988; H. Fraser, pers comm
Gemsbok		+				Jeffrey & Wells, 1988
Eland	1	+				Fleetwood & Furley, 1990
Eland	Molly	+				M. Hosegood, pers comm
Eland	Neddy	+				M. Hosegood, pers comm
Eland	Electra	+	+			M. Hosegood, pers comm
Arabian oryx		+				Kirkwood *et al* 1990
Greater kudu	Linda	+			+	Kirkwood *et al* 1990, 1992; H. Fraser, pers comm
Greater kudu	Karla	+	+	+		Kirkwood *et al* 1992
Greater kudu	Kaz	+		+		Cunningham *et al* 1993, G.A.H. Wells, pers comm
Greater kudu	Bambi	+	+	+		"
Greater kudu	346/90	+	+	+		"
Greater kudu	324/90	+	+			Kirkwood *et al* 1993b, G.A.H. Wells, pers comm
Scimitar oryx		+		+		D. Lyon, pers comm
Puma		+		+		Willoughby *et al* 1992
Cheetah	1	+	+			Peet & Curran 1992
Cheetah	Duke	+	+			J.C.M. Lewis, pers comm
Cheetah	Saki	+				S. McKewan, pers comm
Ostrich	1, 2 & 3	+				Schoon *et al* 1991

(+ indicates positive)

Table 3 Brief notes on the clinical signs observed in cases of spongiform encephalopathy in zoo animals

Animal	Clinical Signs	Duration of clinical signs (days)	Source
Nyala	Hind-limb ataxia, abnormal head posture, persistent nibbling of tail base and rump leading to mutilation and ulceration, frequent micturition.	21	Jeffrey & Wells, 1988
Gemsbok	Sudden onset, frequent, episodic collapsing, good body condition.	*c*7	P Bircher, pers comm
Eland 1	High-stepping of hind limbs, fine muscle tremors of head and neck, loss of weight. Later: circling, head-pressing, dullness, drooling and clear nasal discharge.	8	Fleetwood & Furley, 1990
Eland 2, 3 & 4	Weight loss, drooling, standing apart from others, slight twitching of flank and intercostal muscles. Ataxia not a prominent feature, but one showed slight high-stepping.	14-21	M Hosegood, pers comm
Arabian oryx	Weight loss, muscle tremors, and later ataxia and dullness.	22	Kirkwood *et al*, 1990
Kudu *Linda*	Ataxia, head tilt, drooling, excessive lip-licking and nose-twitching, some weight loss	3	Kirkwood *et al*, 1990
Kudu *Karla*	Ataxia, forelimb crossing, hindlimb hypermetria, head tilt, depression	1	Kirkwood *et al*, 1992
Kudu *Kaz*	Similar to *Diana* and *Linda*	1	
Kudu *Bambi*	No clinical signs at time of euthanasia for management reasons	0	Cunningham *et al*, 1993, Kirkwood *et al*, 1993a
Kudu 346/90	No clinical signs at time of euthanasia for management reasons	0	Cunningham *et al*, 1993, Kirkwood *et al*, 1993a
Kudu 324/90	Initially intermittent head-tilt, mild head tremor, excessive lip movements, hindlimb muscle tremors and hunched posture. Later, decreased abnormal head and ear carriage, hypermetria, possible hyperaesthesia.	56	Kirkwood *et al*, 1993b
Scimitar oryx	Nasal discharge, cough, weight loss. Later collapsed.	18	D. Lyon, pers comm.
Puma	Ataxia, difficulty maintaining balance, fine whole body tremor, "looking upwards and around in an unusual way".	6	Willoughby *et al*, 1992
Cheetah 1	Ataxia, apparent disorientation, falling, and locomotory weakness	28	Peet & Curran, 1992
Cheetah *Duke*	Ataxia, hyperaesthesia. Later, weight loss	30	J C M Lewis, pers comm
Cheetah *Saki*	Ataxia	*c*42	S McKewan, pers comm
Ostriches	Ataxia, disturbance of balance, "discoordinated feeding behaviour".	'protracted'	Schoon *et al*, 1991

Although both of this animal's parents are now dead, the dam had 8 further calves between 1985 and 1991, and no cases of SE have been detected among these. The SE case was 33 months old when she died and had not bred.

In this, and in the case of the kudu *Linda* (see below), experimental transmission to mice was attempted by inoculation of homogenates of fixed brain tissue, and in both cases transmission was successful (Dr Hugh Fraser, pers comm; Jeffrey et al, 1992). The incubation periods observed after intracerebral and intraperitoneal inoculations in 5 strains of mice (Fraser et al, 1992) are shown in Table 4. The pattern of variation in the incubation periods between strains of mice was the same for both the nyala and kudu (RIII < C57BL < VM < IM < C57BL x VM (F, cross)) (Fraser and McConnell, personal communication), and this ranking is also the same as has been found in BSE transmissions (Fraser and others 1992).

Table 4 **Incubation periods measured following experimental inoculation of 6 strains of mice with brain homogenates from a nyala and a greater kudu in which scrapie-like spongiform encephalopathy had been diagnosed on the basis of histopathological signs (Fraser and McConnell, pers comm).** The methodology was as described by Fraser and others 1992.

Mouse strain	Incubation periods in days mean ± sem			
	Nyala		Kudu	
RIII	378	+/- 8	339	+/- 5
C57BL	528	+/- 11	455	+/- 9
VM	548	+/- 16	536	+/- 10
IM	614	+/- 11	560	+/- 12
C57BL x VM	772	+/- 3	751	+/- 24

Gemsbok *Oryx gazella*

The gemsbok case involved a four year old female at the same premises as the nyala. This animal was exposed to the same diet as the nyala and, likewise its diet was thought to contain RDP only from March 1986 to March 1987 - the period three to 15 months before she died.

One of the interesting features of this case was that the animal was in good bodily condition at the time of death. The symptoms were of episodes of sudden collapse lasting for a few minutes, followed by periods of relatively normal behaviour (Table 3).

There were 13 animals in the gemsbok herd at the time of this animal's death. This herd was isolated from other animals but in contact with some Congo buffalo *Synceros caffer*. There had been no contact with the nyala. No further SE cases have been observed in any animals on the premises.

The affected gemsbok had given birth to one calf, a male born on the 6th January 1987, five months before she died. This male calf died the day it was born. The dam of the SE case gave birth to a further six calves between 1984 and 1991 and was euthanased as a result of old age.

Arabian oryx *Oryx leucoryx*

There has been one case in an Arabian oryx (Tables 1,2 & 3). This female was born at Zurich in January 1986 and was imported to Regent's Park in October that year. The herd at Regent's Park was fed on proprietary cattle feed that was quite likely to have contained ruminant-derived protein (RDP) and it was considered by Kirkwood and others (1990) that this animal may have acquired the SE agent from the feed. Assuming that she acquired the infection after arriving in Britain, the incubation period was a maximum of 29 months.

Greater kudu *Tragelaphus strepsiceros*

The incidence of SE in the small herd of greater kudu maintained by the Zoological Society of London has been described in some detail (Kirkwood et al, 1990, 1992, 1993a, 1993b, Cunningham et al, 1993). The temporal and familial relationships of the six confirmed cases are shown in Fig 1. The disease was first confirmed in a female (*Linda*) that died on the 18th August 1989. In this case the diagnosis was confirmed by transmission to mice (Table 2). The ranking of incubation periods in five strains of mice after experimental inoculation with homogenised brain, was the same as found for the Nyala (see above and Table 4) and the same as that found in BSE transmissions (Fraser & McConnell, pers comm).

Since then 5 further cases have been diagnosed at post mortem (Tables 1 & 2). The number of cases in this small herd suggested that the animals were highly susceptible - the disease has been diagnosed in five out of eight born at Regent's Park since 1987 (Kirkwood and others 1993a). Furthermore, although the initial case may have acquired the infection from feed containing RDP, the pattern of incidence suggested that the disease may subsequently have spread by transmission between animals (Kirkwood and others 1992, 1993a, 1993b, Cunningham et al, 1993).

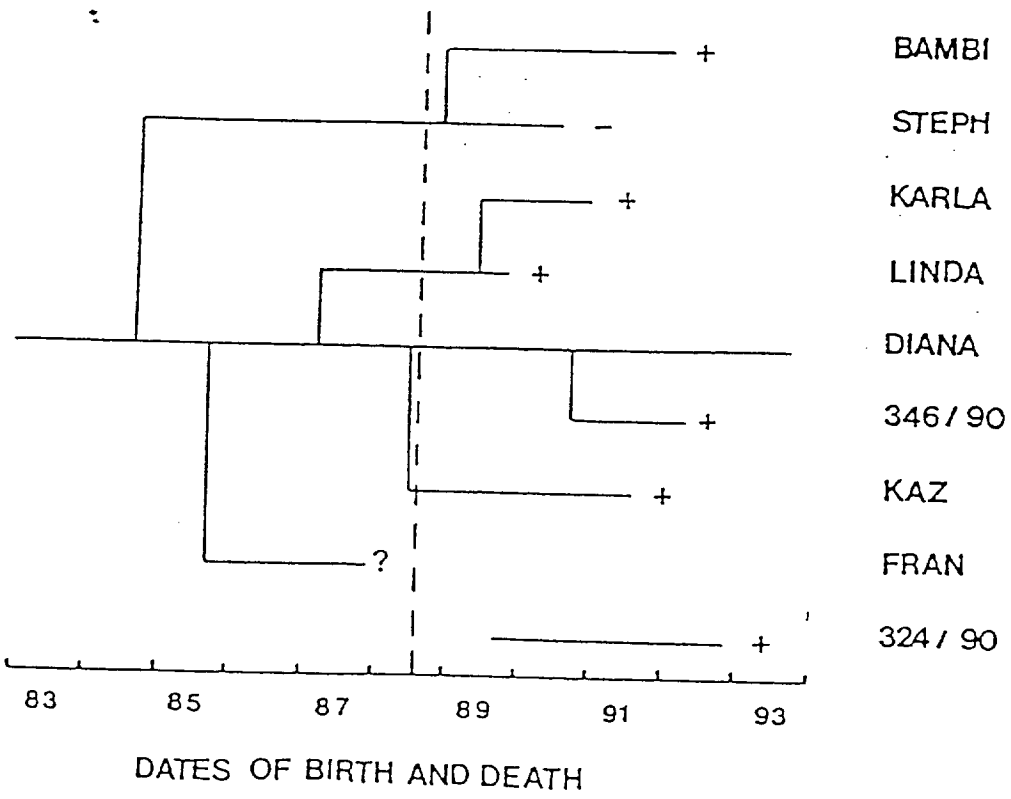
The diet fed to the kudu at the Zoological Society of London did not contain RDP after February 1987 although proprietary pellets that may have contained RDP were present on the premises. It is very unlikely that the kudu would inadvertently have been given the latter and furthermore, the birth dates of three of the affected animals born at Regent's Park were three, nine and 25 months after the ban on inclusion of RDP in ruminant feeds (Table 1). Feed storage practices were such that feed turnover times were not more than a few weeks. In the absence of other potential sources of the disease, we hypothesised that transmission may have occurred between animals.

The most recent case was in a kudu (No. *324/90*) born 13 months after July 1988 (see Tables 1,2 and 3). This animal was born at another zoo in England where feeds had not contained RDP since March 1987, and was moved to Regent's Park on 14th August 1990, 27 months before she died (Kirkwood et al, 1993b). Since the incubation period can be as little as 19 months in kudu (see details on '*Karla*' in Table 1) and there had been

no cases of SE in kudu at the zoo where this animal was born, nor the likelihood of exposure to RDP, we considered that this animal may have acquired the disease by transmission from affected animals after arriving at Regent's Park (Kirkwood et al, 1993b).

Figure 1. **The temporal and familial relationships of the cases of scrapie-like spongiform encephalopathy in greater kudu at the Zoological Society of London.**

Diana remains alive at time of writing, + indicates those animals in which SE was diagnosed, - indicates those which did not have the disease and, ? indicates an animal which, in retrospect, on the basis of clinical signs may have had the disease. The dotted line indicates the date of the July 1988 ban on the inclusion of ruminant-derived protein in ruminant feeds.



Eland *Taurotragus oryx*

Four confirmed cases of SE have occurred in eland (Tables 1, 2 & 3) (Bradley and Matthews, 1992), but only one of these has previously been reported in any detail in the literature. The first case was that described by Fleetwood and Furley (1990) in a 32 month-old male eland. This animal had been fed on a proprietary ungulate ration which contained RDP from calfhood to June 1988, and the authors suggested that it was possible

that this exposed the animal to the causal agent of BSE or scrapie.

The three other confirmed cases occurred at another collection in England, and involved animals born six, six and 18 months after the July 1998 ban on inclusion of RDP in ruminant feeds (Order 1988). Here, the eland herd had been maintained at a size of 14 to 26 animals and no new animals had been imported since before 1982. The diet included a commercial equine pellet which did not contain RDP but proprietary cattle feeds were used for other species kept on the premises. However, because of the rate of turnover of feed on the premises, it was considered very unlikely that potentially contaminated feeds were present at the time of the birth of these calves.

It seems unlikely that the calves could have acquired the disease by maternal transmission since the dams of all three remain alive at time of writing (July 1993). An adult female, *Daphne*, which died on 18th August 1990, prior to these three cases, had shown similar clinical signs and, in retrospect, it is suspected that she may have had SE. This cannot be confirmed because no brain tissue was preserved. This animal was born on 18th April 1988 and could therefore have been exposed to feeds which contained RDP. It is therefore conceivable that she acquired the disease from feed and that she transmitted it, through contact in some way, to the three subsequent cases. However, there are other possible explanations.

The eland herd where these three cases occurred is in contact with asian elephants *Elephas maximus*, white rhino *Ceratotherium simum*, Ankole cattle *Bos taurus*, and zebra *Equus burchelli*, but no cases have been diagnosed in these species. Lions *Panthera leo* and tigers *Panthera tigris* are also kept on the premises and have been fed heads and other pieces of cattle from a knacker yard. These heads used to be fed whole but, since August 1988, they have been split and have had the central nervous tissue removed. The delivery of these to the lions and tigers was via a route through the eland's enclosure and, although it is thought unlikely, it is possible that some contamination of this enclosure could have occurred as a result of material dripping from the meat vehicle. It is also possible, in view of the proximity of the lion and tiger enclosures, that scavenging wild birds might have carried contaminated tissues into the eland enclosure.

In this context it is interesting that two cases of feline spongiform encephalopathy (FSE) have been diagnosed (by histopathology) in semi-feral domestic cats that were known to have scavenged in the meat room where the food for the lions and tigers was stored and prepared. These cats were euthanased in January 1991 and January 1992 (Hosegood, pers comm) after developing progressive ataxia and other signs consistent with FSE (Leggett et al, 1990; Wyatt et al, 1990).

In view of the rarity of FSE (at the time of writing a total of 41 cases have been diagnosed in domestic cats in Britain - Bradley, personal communication), it is reasonable to consider if these two cases could have been related to the three SE cases in the eland. There are three ways in which they could be related. First, the cats could have infected the eland. Second, the eland could have infected the cats. Third, they could have been infected by exposure to a common source.

There is no evidence that FSE is transmissible and it is not thought that the cats would ever have been in contact with the eland, but the possibility that they could have foraged in their paddock cannot be ruled out. Conversely, when eland deaths occurred, the carcases were incinerated so it is unlikely that the cats could ever

have eaten eland tissues. Perhaps exposure to a common source, such as knacker cattle heads or dripped fluids from them, is the most likely. It is interesting to note that to date no cases have occurred in the lions or tigers.

Scimitar-horned oryx *Oryx dammah*

Scrapie-like spongiform encephalopathy has been diagnosed on the basis of histopathological observations in one female scimitar-horned oryx (Dr Sandra Scholes and Mr Gerald Wells, personal communications)(Tables 1 and 2). No cases of SE have been reported at the zoo where this animal was born on 12th July 1990. She was moved on 25th April 1991 to another zoo, where although no cases have been recognised in ungulates, SE was diagnosed in a puma (see below).

Her death occurred 21 months after moving from the zoo where she was born. We do not know the incubation period of the disease in scimitar-horned oryx, but observations in other species suggest that it could be greater or less than 21 months (see Table 1) and we are unable therefore to know whether she acquired the infection before or after moving from her birth place.

The animal's diet at the premises where she was born included a proprietary dairy cattle feed, oats, flaked maize and proprietary vitamin pellets. This animal was born 24 months after the ban on inclusion of RDP in ruminant feeds and the manufacturers consider it extremely unlikely that these feeds could have contained RDP. Her diet at the zoo she was moved to included a proprietary concentrate feed but the manufacturers ceased including RDP in July 1988 and are sure that all stocks of pre-July feed were used before November 1988. At the premises where she was born, in addition to contact with the 15 or so other scimitar horned oryx in the herd, the animal had been in contact with common zebra *Equus burchelli* and red lechwe *Kobus leche*. At the zoo where she died, she had been housed with giraffe *Giraffa camelopardalis* and Arabian gazelle *Gazella gazella* and may also have had contact with roan antelope *Hippotragus equinus* and forest buffalo *Synceros caffer*. However, SE has not been diagnosed in any of the animals with which she had been in contact at either of these premises. Her dam is still alive at the time of writing but her sire was euthanased, for management reasons, prior to her birth. The source of the disease in this case remains unknown at present. Feeds that are likely to have contained RDP prior to the July 1988 ban were used at both the zoos in which she had been kept so it may perhaps be possible that some environmental contamination could have taken place. Alternatively we could speculate that she could have acquired the disease from another animal which did not, or has not yet, shown any signs of the disease, or that this could have been a spontaneous case.

During her illness prior to death, this animal showed no locomotor disturbances and the clinical signs were not strongly suggestive of SE (Table 3).

Cheetah *Acinonyx jubatus*

There have been three cases in cheetah (Tables 1 and 2). The first case was in an adult male, that had been born at a zoological park in England on 16th June 1986 and had been exported, with two litter mates, to the Pearl Coast Zoo, Broome, Western Australia, arriving on May 9th 1989 (Peet and Curran 1992). The animal showed signs of illness for three weeks before its death on 7th January 1992 (Table 3). It was assumed that the

animal had acquired the disease prior to leaving the UK, in which case the incubation period must have been at least 26 months. At the zoo where it was born, the cheetahs were fed on cattle and possibly some horse meat from a knacker yard. Although no heads were included, split spinal columns were fed.

The second case in a cheetah occurred in a male (*Duke*) born on 3rd September 1984 at the same zoo (Tables 1,2 & 3). When 22 months old the animal had been moved, via Whipsnade Wild Animal Park where he spent six weeks (from 2nd July 86 to 17th August 86), to the collection where he died in 1992. From August 1986 to the end of 1991, his diet had been sections of cattle and horse carcases from a knacker yard. These sections did not include heads or backbones. He was also occasionally fed on whole stillborn calves, also from the knacker yard. The heads of these were sometimes partially eaten. Other items included, rabbits, one Soay sheep from the same collection which had been given to him in 1990, and wild pigeons which he occasionally caught. From the beginning of 1992 onwards his diet had been only horsemeat from the knacker yard and chickens (Dr John Lewis, pers comm).

The third case (Tables 1,2 and 3) occurred in a seven year-old female (*Saki*) born at the same zoo as the other two. This animal was hand-reared. She was moved from the zoo where she was born on 18th September 1990, spent five months at another zoo, and was then moved again on 22nd February 1991. She died at this zoo on 4th May 1993, 26 months after her arrival, and after showing progressive neurological signs for about six weeks.

These three cheetah had the same dam (*Dusky*) but different sires. The dam had a total of nine cubs. The two litter mates of the one that died of SE in Australia were euthanased. The three litter mates of *Duke* all went to Whipsnade and two remained there but one was moved on to another zoo on 17th August, 15 months after arriving at Whipsnade. The fate of this animal is not known, but the two animals that remained at Whipsnade have since died. Of *Dusky's* other two cubs, one was sent to Whipsnade on 12th September 1990 and was euthanased on 24th July 1993, and the other was *Saki* that died with SE in May 1993.

It would seem likely that the three animals with SE may all have acquired the disease at the zoo where they were born.

Puma *Felis concolor*

One case of SE has been reported in a puma (Tables 1,2 & 3). The case has been described in detail by Willoughby and others (1992), but we list some features relevant to the epidemiology here. This female remained in the collection at which she was born throughout her life. She was separated from her dam at ten months of age. Both parents died of non-neurological diseases. Her diet consisted of chicken and rabbit carcases and parts of cattle carcases deemed unfit for human consumption. These parts included split spinal columns but not heads and, because she appeared to find offals unpalatable, they were rarely offered to her. She had never been fed on sheep or goat tissues but had once, three months before onset of clinical signs and 4.5 months before her death, been fed on meat from two eland that had been culled in the collection. These eland had been born in 1988 and had been fed on commercially-prepared cattle food.

The source of the infection in this case is unknown but, in view of the circumstances outlined above, the authors considered it was possible that the animal had acquired the disease from BSE-infected cattle tissue

(Willoughby and others, 1992).

Ostrich *Struthio camelus*

Schoon et al (1991) reported three cases of spongiform encephalopathy in ostriches from two zoos in North West Germany. The cases occurred in 1986, 1988 and 1989 and the birds were euthanased after showing protracted central nervous symptoms with ataxia, disturbances of balance and uncoordinated feeding behaviour (Table 3). The authors considered that the appearance and pattern of distribution of the lesions observed by light microscopy were similar to those of transmissible SE's in mammals. However, a conclusive diagnosis could not be made and it was suggested that a toxic or nutritional aetiology could not be discounted.

The diet of these birds had included proprietary poultry feed and meat, some of which was from cattle emergency slaughter cases. Cases with similar clinical signs had been observed at both zoos but had not been examined at post mortem.

DISCUSSION

In this review of SE in captive wild animals we have listed 18 cases in eight species at or from seven institutions in the British Isles and mentioned also three cases reported in ostriches in Germany. Summaries of the dates of birth and death of the cases in the British Isles are given by Figs 1 and 2. The pattern of incidence of these cases raises several questions about the epidemiology of these SEs and there are some challenges in devising appropriate control measures in captive wild animals. Here we first consider the possible relationship of these cases with BSE and the significance of their taxonomic distribution, and conclude with some considerations regarding control of the disease.

Possible relationship to BSE

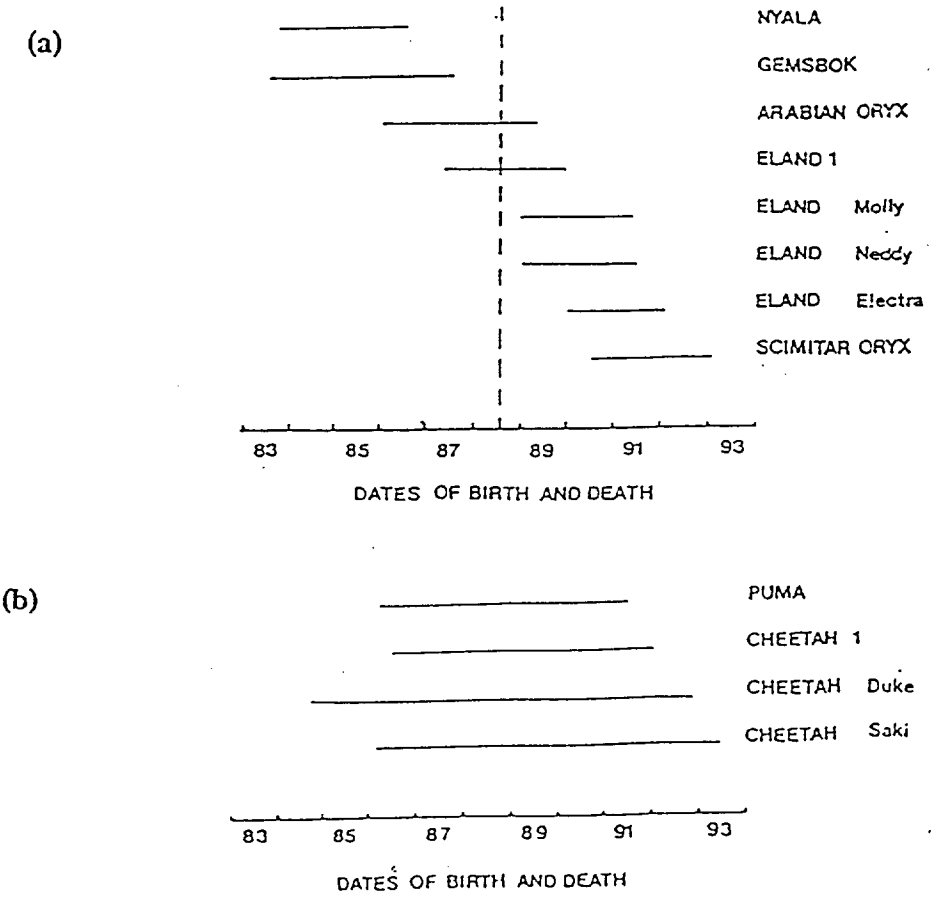
Four pieces of evidence support the hypothesis that these SEs are related to the BSE epidemic. First their temporal and geographic coincidence with the BSE epidemic. Second, the certainty in some cases and likelihood in others that the affected animals or others with which they were in contact had been exposed to feeds containing RDP or, in the case of the carnivores, to sections of cattle carcases that had been deemed unfit for human consumption. Third, the finding reported here on the similarity of the pattern of variation in incubation period in five strains of mice following inoculation of brain homogenates from a kudu and a nyala, to the pattern observed on transmission of BSE. Fourth, there were no significant differences in the character or distribution of histological and ultrastructural lesions between the mice inoculated with the kudu and nyala brain homogenates and those inoculated with homogenates from BSE-affected cows (Jeffrey et al, 1992).

It therefore seems reasonable to consider that at least some of the cases of SE in captive wild animals were caused by the agent of BSE. However, there is no conclusive proof of this and other explanations, although less likely, can be envisaged. One possibility is that SEs may have been present but undetected in some of these species prior to the BSE epidemic. Considering that SEs had been recognised prior to 1980 in six species (man, sheep, goat, mink, mule deer and elk) of the relatively small handful of mammals whose diseases have been studied closely, it would perhaps be surprising if diseases of this type do not naturally occur in some of the other

4050 or so species of mammals. It is almost certainly true that, were it not for increased vigilance for SEs among zoo veterinarians and pathologists, several of the cases reported here would not have been diagnosed. Nevertheless, we think it is unlikely that SEs would have all but completely escaped detection if they had been present in zoo herds in the past. In fact, two were detected prior to the BSE epidemic. An SE, assumed to be scrapie, had been diagnosed in mouflon by 1983 (Wood et al, 1992) and the diagnosis of SE in the nyala predated the detection of BSE in cattle (Jeffrey & Wells, 1988).

Figure 2. **Dates of birth and death of cases of scrapie-like spongiform encephalopathy in zoo animals in the British Isles between 1983 and 1993. (a) Artiodactyla other than Greater kudu and (b) captive wild felids.**

The dotted line in (a) indicates the date of the July 1988 ban on the inclusion of ruminant-derived protein in ruminant feeds.



Patchiness of incidence among taxa

From a taxonomic perspective, the incidence of cases has been strikingly patchy. A wide range of species of wild animals are kept in zoos in the British Isles but cases of SE have only been diagnosed in animals in the orders Artiodactyla and Carnivora. An indication of the number of individuals and range of species of wild Artiodactyla and Carnivora present in collections in the British Isles is provided in Table 5. This presents statistics on populations of all the species, in these orders, kept by member organisations of the Federation of Zoological Gardens of Great Britain and Ireland, in December 1989. At that time there were 2675 individuals of 62 species of artiodactyla and 1227 individuals of 69 species of carnivora in these collections. These numbers underestimate the total numbers of wild animals maintained in captivity in the British Isles since not all those who keep them are members of the Federation of Zoos.

Among the Artiodactyla, cases have occurred only in the family bovidae, and within this, incidence has been limited to certain closely-related species within each of the subfamilies bovinae and hippotraginae (Table 5). Similarly, among the carnivora, all the observed cases have occurred in the felidae and, of these cases, three out of four have occurred in cheetah. Joints of meat from cattle unfit for human consumption have certainly been fed to species of large cats other than cheetah and puma and, almost certainly, have been fed to other large carnivores, notably wolves and bears. If the BSE agent has been the cause of the zoo animal SEs then how do we explain the taxonomic patchiness of the incidence?

Firstly, as we have already pointed out, the animals in which the disease has been diagnosed were among those which either were, or could have been, exposed to proprietary ruminant feeds or to tissues from cattle carcases unfit for human consumption. However, a considerably greater range of species was exposed to these foods than those in which cases have occurred, and detection of the disease in clusters of closely-related species (eg eland, kudu and nyala; puma and cheetah) suggests that genetic factors may also have influenced risk. However, there are other possible explanations. One is that the range of species in which the disease has been detected so far is that in which the incubation period is relatively short, and that the actual range may prove to be wider and to include individuals of other species which are still incubating the disease at present. Alternatively, the clusters could reflect variations in the intensity of exposure. The three cheetah cases could for example all have been subjected, in some way, to particularly intense exposure at the premises at which they were all born.

Thus, knowing nothing about variation in intensity of exposure and being cognisant of the possibility that other cases may emerge, no firm conclusions can yet be drawn regarding possible between-species variation in susceptibility.

It is notable that, although the diets of many species of smaller carnivores and omnivores kept in zoos have included RDP, so far no cases have been detected in any of these animals.

Table 5 **The number of species and of individuals of wild animals in the Orders Carnivora and Artiodactyla held by members of the National Federation of Zoological Gardens of Great Britain and Ireland on 31st December 1989, and the population sizes at that time of the species in which SE's have been diagnosed.** Although populations change from year to year, this gives a rough index of the size of the national populations during the period under discussion.

NB: We are unable to say how closely these numbers represented the population at risk, since we do not know the source or sources of infection, the degree of exposure, or how susceptibility varies between species.

Order	Family/ Subfamily	Species	No. species	N*
Artiodactyla			62	2675
	Bovinae		14	304
		T. oryx		35
		T. angasi		26
		T. strepsiceros		17
	Hippotraginae		10	270
		O. dammah		65
		O. gazella		29
		O. leucoryx		5
Carnivora			69	1227
	Felidae		23	543
		F. concolor		16
		A. jubatus		42

*N is the number of individuals in the National Federation's collections on 31st December 1989.

Sources of infection

Epidemiological studies of BSE provide convincing evidence that cattle acquired the disease through the consumption of proprietary feeds which contained RDP (Wilesmith and others 1988, 1991, 1992a, 1992b). It is thought that, if it occurs at all, transmission is very rare in cattle (Taylor 1992, Wilesmith and others 1992b, 1993), and thus that, following the July 1988 ban on inclusion of RDP in ruminant feeds, the disease will not be maintained in the British cattle population (Taylor, 1991).

Wilesmith and others (1993) reported 449 cases in cattle born after the July 1988 food ban. They attributed the occurrence of these cases to exposure to foodstuffs manufactured before July 1988 which remained in the supply chain or on farms after this date, and considered that exposure from this source would have continued for at least four months after the statutory ban. More recently, it has been reported that there have been 2857 cases in cattle born after July 1988, the majority of which were born within three months of the ban (Matthews cited by Anon 1993). However, as is shown above, it is not easy to explain the cases in the zoo

ungulates born after July 1988 in this way because the diets of some did not include feeds containing RDP and because birth dates were, in some cases, many months after July 1988 and feed turnover rates on the premises were quite high.

It appears that the disease in greater kudu may have a different epidemiology to that in cattle. The pattern of incidence in kudu suggests that transmission occurs between animals through contact, and that the disease could (or has) become endemic in one herd. The epidemiology of SE in kudu appears therefore to be more like scrapie in sheep and chronic wasting disease in deer in both of which transmission occurs by direct or indirect contact (Kimberlin 1990, Williams and Young 1992).

If it were not for the possibility that the eland's paddock may have become contaminated as a result of transport of pieces of knacker cattle through it, the cluster of cases in animals born after July 1988 would be hard to explain other than as a result of transmission between animals as we have postulated occurs in the kudu. The origin of the disease in the scimitar-horned oryx is unknown.

Control of SE in zoo animals

The July 1988 ban on inclusion of RDP in ruminant feeds in (Order 1988), and the September 1990 ban on the feeding to any animal of specified offals (brain, spinal cord, spleen, thymus, tonsils and intestines) from cattle over six months of age (Amendment Order, 1990), were instituted to prevent the further exposure of animals fed on these feeds to the BSE agent. The 1990 order concerns the feeding of offals from domestic bovines but, in view of the cases in zoo ungulates reported here, it would be advisable not to feed animals on the offals of any of the species in which cases have occurred or of those in which cases could occur. In the absence of knowledge of the tissue distribution of infectivity in zoo animal cases it would seem prudent to avoid the use of any zoo animals that could have been exposed to the BSE agent, as food for others.

Most zoo animal herds are small and, in order to maintain the genetic diversity of captive populations, animals have to be moved between zoos for breeding. This involves both local and international movements and these are inevitably associated with some risks of accidental disease transmission. Furthermore there is growing interest in the reintroduction of captive-bred animals to the wild for species conservation programmes and these 8reintroductions provide a route for accidental transmission of infectious diseases into free-living populations (Woodford & Rossiter, 1993).

Assessing and minimising the risks of spread of SE within and between species of captive wild animals presents particular difficulties for several reasons (Cunningham, 1991; Kirkwood & Cunningham, 1992; see also Williams & Young, 1992). First, introduction of the disease may have very serious consequences to the conservation value of a captive population. Secondly, infected animals cannot be identified in life and, third, the incubation period is prolonged and may vary within and between species.

It is likely that the range of species exposed to feeds that may have contained the BSE agent is considerably greater than the range of species in which SE's have so far been detected, and cases may yet emerge in other species. Furthermore, if the kudu SE was caused by the BSE agent then it is possible that BSE-related SE's that may emerge in other species could prove to be transmissible as appears to be the case in the

kudu. There may also be a possibility that individuals of some species could carry and transmit the disease without showing any clinical signs. It would seem prudent therefore to avoid movements of animals that could have been exposed to the BSE agent or their offspring or in-contacts, into populations that have not been exposed, unless the damage that this would cause to the conservation breeding programme outweighs the risk of the introduction of an SE.

Further work

There is a need for continued vigilance for SEs in zoo animals. The detection of the disease on histopathological examination of animals which had shown no-clinical signs or signs not suggestive of an SE indicates that ideally animals which may be suspected of dying of other causes should also be monitored.

To elucidate the routes by which transmission could have occurred in greater kudu, an investigation of the tissue distribution of infectivity has been initiated using tissue samples collected from cases at the Zoological Society of London.

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