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An outbreak of tetanus was observed in lambs from a flock of 600 ewes in the Sabzevar district of the Khorasan province in Iran during the winter of 1996. Over a 35-day period, 19 lambs showed signs of tetanus and 18 of them died. The flock (without lambs) grazed on a poor pasture and was supplemented with alfalfa at night. In this flock ear tagging of lambs was performed seven to eight days after birth. All the lambs were vaccinated against enterotoxaemia at 12 to 20 days of age. Tetanus in these lambs was observed at 15 to 38 days of age.

The clinical signs exhibited initially were mild stiffness, especially in the forelimbs, and asymmetrical positioning of the ears. These symptoms progressed to severe stiffness and rigidity of the neck and limbs. Other symptoms included tremor, hypersensitivity to touching, lockjaw, dilation of nostrils, and recumbency with extension of the limbs in the terminal stages. All affected lambs, with the exception of one, died within five to 14 days of the appearance of the clinical signs.

Haematological parameters (tested in three lambs) and serum calcium and magnesium levels (tested in two lambs) were within normal ranges. Cerebrospinal fluid was taken from the cisterna magna in three lambs and examined for the presence of aerobic and anaerobic bacteria; the results of the cultures were negative. Seven lambs were submitted for postmortem examination. There were no wounds of any significance on the animals other than at the ear tagging sites. Large size, locally made, plastic tags had inadvertently been applied too close to the base of the ears and the point of insertion of all of the tags was covered by a blood clot. Samples from these sites taken from five affected lambs were cultured anaerobically and a pure growth of *C tetani* was observed in two. Histopathological examination of the brain tissues did not reveal any lesions. Following diagnosis of the disease, the ear tagging was stopped in the newborn lambs and no new cases of tetanus were seen in the flock during 1996.

The diagnosis in this outbreak was made on clinical grounds and laboratory findings. There was evidence that the lesions at the ear tag insertion sites were the source of the tetanus toxins as *C tetani* was isolated from two of five samples which were taken from these sites. The time of ear tagging and onset of symptoms was consistent with the incubation period of tetanus (Radostits and others 1994) and, additionally, no other visible wounds were found on the affected lambs.

Vaccination has also been reported as a means of entrance of *C tetani* spores and the development of tetanus in sheep (Char and others 1993, Radostits and others 1994). Although this route of infection can not be ruled out in these cases, the development of a blood clot at the insertion sites may have established anaerobic conditions suitable for the multiplication of *C tetani*. Recent reports have described problems with the use of ear tags in cattle (Wardrope 1995, Johnston and Edwards 1996) and sheep (Hosie 1995). In addition, in Iran, there is no programme for the vaccination of ewes against *C tetani*, nor was there any history in the affected flock of vaccination of pregnant ewes against clostridial diseases.

Tags used for marking farm animals should be inserted approximately one-third of the length of the ear from the base, generally on the upper edge of the ears with the number on top (Banerjee 1991). Furthermore, ear tags should be lightweight, bright in colour and easily inserted into the ear by a competent operator using an applicator which does not spread disease.

References

BANERJEE, G. C. (1991) Textbook of Animal Husbandry, 7th edn, New Delhi, Oxford and IBH Publishing. p 598

CHAR, N. L., RAJESWARL, K. R. & REDDY, B. V. R. (1993) *Indian Veterinary Journal* **70**, 297

HERD, R. P. & RICHES, W. R. (1964) *Australian Veterinary Journal* **40**, 356

HOSIE, B. (1995) *Veterinary Record* **137**, 571

JOHNSTON, A. M. & EDWARDS, D. S. (1996) *Veterinary Record* **138**, 612

RADOSTITS, O. M., BLOOD, D. C. & GAY C. C. (1994) Veterinary Medicine, 8th edn. London, Baillière Tindall. p 677

RAMSAY, W. R. (1973) *Australian Veterinary Journal* **49**, 188

TIMONEY, J. F., GILLESPIE, J. H., SCOTT, F. W. & BARLOGH, J. E. (1988) Hagan and Bruner's Microbiology and Infectious Diseases of Domestic Animals. 8th edn. London, Comstock Publishing. p 215

WARDROPE, D. D. (1995) *Veterinary Record* **137**, 675

Clinically silent rabies infection in (zoo) bats

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RABIES virus belongs to the lyssavirus subgroup of the Rhabdoviridae family (Table 1). Of the four sero- and six genotypes in this subgroup, genotypes 5 and 6 relate to the European bat population and are further subdivided (Bourhy and others 1993, Amengual and others 1997).

Fatal rabies in humans and animals is most commonly caused by geno- and serotype 1. These cases often result from contact with rabid terrestrial animals such as dogs, skunks, raccoons and foxes. In Latin America and the USA, fatal rabies in man may also be related to contact with bats, which are infected by different variants of the rabies virus belonging to genotype 1, represented in the local terrestrial animals (Krebs and others 1996). African bats are, however, also infected with specific geno- and serotypes (Lagos and Duvenhage), of which only the Duvenhage strain has been isolated from fatal rabies in man.

In 1985/86 Denmark and other European countries recorded a large number of infections in free-living bats (*Eptesicus serotinus*) with a Duvenhage-like European bat lyssavirus (EBL-1a) which was considered a possible danger to man (Grauballe and others 1987, Muller 1987, Schneider and others 1987). In Denmark these infections were confirmed in 19 per cent of cases sent for diagnosis. Following a 10-year period with only a low incidence (4·7 per cent), Denmark again saw an increase in cases in the summer of 1997 (26 per cent). Furthermore, England (Whitby and others 1996) and Australia (Frazer and others 1996), previously free from rabies, also experienced sporadic rabies infections in their bat populations.

In the beginning of July 1997, a colony of 42 flying foxes (*Rousettus aegyptiacus*) was imported from a Dutch zoo to one in Denmark. Nine days after their arrival two bats died from rabies, verified by a standard immunological staining of brain smears. A virus cell culture harvested from one of these was subsequently classified as an EBL-1a subgenotype at the Central Veterinary Laboratory, UK, with a 99·8 per cent nucleotide sequence homology with a reference strain (Amengual and others 1997).

Since the colony had been established in an artificial cave which was open to visitors, all the bats were euthanased and the premises disinfected. Of the 40 apparently healthy bats in the colony a further three showed diagnostic evidence of localised rabies virus infection on brain smears, giving an overall infection rate of 11·9 per cent. One of these three animals subsequently proved culture positive for rabies virus.

Ten days after the export of the bats to Denmark, an investigation by the ID-DLO laboratory of the 254 remaining healthy bats in the original Dutch colony showed that 13 per cent elicited weak rabies-positive staining on brain smears in a few localised areas by standard immunofluorescence testing (Bourhy and Sureau 1990). Suspensions of of brain material from 22 of these bats suspected of having rabies were injected intracerebrally into 140 newborn (OF1 strain) mice (six to seven mice per brain suspension) and resulted in the death of 30 animals, of which only one

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TABLE 1: Subgroups of lyssavirus

Virus	Serotype	Genotype	Geographical origin	Original host	Secondary host
Rabies virus	1	1	Worldwide, except eg: Scandinavia, Iceland, UK, Ireland, Australia, New Zealand	Dog, cat, fox, skunk, raccoons, mongoose, bat (Americas)	Mammals, man
Lagos bat virus	2	2	Nigeria and other African countries	Bat (frugivorous)	Cat, dog
Mokola virus	3	3	Nigeria and other African countries	Shrews, rodents	Cat, dog, man
Duvenhage bat virus	4	4	South Africa, Zimbabwe	Bat (insectivorous)	Man (South Africa 1971)
European bat lyssa-virus 1a (EBL-1a)	?	5	Denmark, Germany, Netherlands, Poland, Russia	Bat (insectivorous)	Man (Russia 1985)
European bat lyssa-virus 1b (EBL-1b)	?	5	Netherlands, France, Spain	Bat (insectivorous)	
European bat lyssa-virus 2a (EBL-2a)	?	6	Netherlands, UK	Bat (insectivorous)	
European bat lyssa-virus 2b (EBL-2b)	?	6	Finland, Switzerland	Bat (insectivorous)	Man (Finland 1986)
Australian bat lyssa-virus	?	?	Australia	Bat (frugivorous and insectivorous)	Man (Australia 1997)

showed a weak positive rabies staining reaction. This was an atypical behaviour compared with a reference EBL-1a strain from rabies postive *E serotinus* bats which was injected into control mice, all of which died between day 6 and 25 showing a strong and widely disseminated rabies specific fluorescence staining on brain smears. Negative controls, injected with brain material from non-rabies suspected bats, were killed one to two months later and showed no rabies specific immunofluorescence staining reaction. However, the rabies diagnosis was finally confirmed by nucleotide sequence analysis at the Institut Pasteur though in only two of nine bats suspected of having rabies. The strain was again classified as an EBL-1a subgenotype. Consequently the remaining animals in the Dutch colony were also destroyed.

A similar clinical rabies outbreak has previously been experienced in a Danish closed laboratory bat colony (*Eptesicus fuscus*), newly imported from the USA, in which 50 per cent of the animals died, starting three to four weeks after transfer (Stougaard 1994).

The finding of an EBL-1a subgenotype in the Dutch flying fox colony indicates a northern European source of the original infection (Amengual and others 1997) which was probably introduced into the colony at a point in its history. The colony concerned had been established in 1991 following the importation of animals from 12 different zoos; it had remained a closed colony since its establishment. It is possible, however, that the increased number of cases of rabies seen in free-living bats in Denmark during 1997 may have increased the likelihood of infection passing from free-living to captive bats. The present findings support the hypothesis that persistent, subclinical rabies infections are more common in bats than previously thought and that such infection may lead to clinical outbreaks of rabies following the imposition of an additional stress factor such as transport, the habitation of new dwellings or high environmental temperatures.

The possible transmission of rabies virus from bats to human beings is a matter of continued concern. The new genotype classification system of rabies viruses has demonstrated that, in addition to the African Mokola, Duvenhage, and the new demonstrated Australian (not yet genotyped) rabies related virus strains, the EBL-1a and EBL-2b strains, which are widespread in western Europe, have been isolated from a few fatal cases in man (Table 1). It is therefore suggested that the opening of any private or public bat aviary to visitors be reconsidered, unless the actual bat colony can be confirmed as rabies-free.

Bat species are protected by law. However, individual free-living or captured bats showing abnormal behaviour should, if possible, be collected with due concern for personal safety and forwarded to a diagnostic laboratory in order to exclude or subtype any rabies virus. Though the EBL types have never been demonstrated in terrestrial animals (Bourhy and others 1992, Whitby and others 1996), any injury in humans or domesticated animals caused by a bat suspected of being infected with rabies must immediately be followed by rabies postexposure prophylaxis, despite the weaker efficacy of the conventional rabies vaccine of genotype 1 against infections caused by EBL (Lafon and others 1988, Perrin and others 1991).

References

AMENGUAL B., WHITBY, J. E., KING, A., SERRA COBO, J. & BOURHY, H. (1997) *Journal of General Virology* **78,** 2319

BOURHY, H., KISSI, B., LAFON, M., SACRAMENTO, D. & TORDO, N. (1992) *Journal of Clinical Microbiology* **30,** 2419

BOURHY, H., KISSI, B. & TORDO, N. (1993) *Virology* **194,** 70

BOURHY, H. & SUREAU, P. (1990) Methodes de Laboratoire pour la Diagnostic de la Rage. Paris, Institute Pasteur

FRAZER, G. C., HOOPER, P. T., LUNT, R. A., GOULD, A. R., GLEESON, L. J., HYAT, A. D., RUSSEL, G. M. & KATTENBELT, J. A. (1996) *Emerging Infectious Diseases* **4,** 327

GRAUBALLE, P. C., BAAGOE, H. J., FEKADU, M. & ZOFFMAN, N. (1987) *Lancet* **1,** 379

KREBS, J. W., STRINE, T. W., SMITH, J. S., NOAH, D. L., RUPPRECHT, C. E. & CHILD, J. E. (1996) *Journal of the American Veterinary Medical Association* **209,** 2031

LAFON, L., BOURHY, H. & SUREAU, P. (1988) *Vaccine* **6,** 362

MULLER, W. W. (1987) *Rabies Bulletin Europe* **4,** 17

PERRIN, P., JOFFRET, M.-L., JANETTI, C., BOURHY, H., GONTIER, C., FRITZELL, C., LECLERC, C. & SUREAU, P. (1991) *Vaccine* **9,** 549

SCHNEIDER, L. G., MUELLER, W. W. & HOHNSBEEN, K. P. (1987) *Rabies Bulletin Europe* **4,** 12

STOUGAARD, E. (1994) *Rabies Bulletin Europe* **3,** 5

WHITBY, J. E., JOHNSTONE, P., PARSSON, G., KING, A. A. & HUTSON, A. M. (1996) *Veterinary Record* **139,** 491

First report of bovine neosporosis in dairy cattle in Costa Rica

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NEOSPOROSIS has recently been recognised as a major cause of abortion in dairy cattle worldwide (Barr and others 1990, Duff and Otter 1994, Boulton and others 1995). The disease is caused by the protozoan parasite, *Neospora caninum*, which is closely related to *Toxoplasma gondii* (Dubey and Lindsay 1993). Consequently, it may have a life cycle similar to *T gondii*, which is transmitted by ingestion of oocysts in the faeces of a definitive host or by oral or nasal exposure to tachyzoites or oral exposure to

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