

**Infectious diseases of saiga antelopes and
domestic livestock in Kazakhstan**

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Chapter 1

Introduction

This thesis combines an investigation of the ecology of a wild ungulate, the saiga antelope (*Saiga tatarica*, Pallas), with epidemiological work on the diseases that this species shares with domestic livestock. The main focus is on foot-and-mouth disease (FMD) and brucellosis. The area of study was Kazakhstan (located in Central Asia, Figure 1.1), home to the largest population of saiga antelope in the world (Bekenov *et al.*, 1998). Kazakhstan's independence from the Soviet Union in 1991 led to a dramatic economic decline, accompanied by a massive reduction in livestock numbers and a virtual collapse in veterinary services (Goskomstat, 1996; Morin, 1998a). As the rural economy has disintegrated, the saiga has suffered a dramatic increase in poaching (Bekenov *et al.*, 1998). Thus the investigation reported in this thesis includes ecological, epidemiological and socio-economic aspects, all of which were necessary in order to gain a full picture of the dynamics of the infectious diseases of saigas and livestock in Kazakhstan.

The saiga is an interesting species to study because it is one of the few wildlife populations in the world that has been successfully managed for commercial hunting over a period of more than 40 years (Milner-Gulland, 1994a). Its location in Central Asia, an area that was completely closed to foreigners during the Soviet era, means that very little information on the species and its management has been available in western literature.

The diseases that saigas share with domestic livestock have been a particular focus of this study because of the interesting issues related to veterinary care and disease control in the Former Soviet Union (FSU). Soviet Central Asia is only just opening up to the outside world, and the area is in rapid transition. Until very recently, little was known about their methods of animal husbandry, disease control and veterinary measures. The disease status of their animals and the state of veterinary research were not disclosed, either within the country or externally. Even now, a large proportion of the scientific literature is not in the public domain, and thus difficult to obtain. Of the literature that is available, most is in Russian and not accessible to western audiences. Within the western literature, there has been some work carried out on the disease-mediated interactions between livestock and wildlife (Christiansen & Thomsen, 1956; Anderson & Trehwella, 1985; Freeland &

Boulton, 1990), but it is still a relatively under-researched field, and one that is receiving increasing recognition as important both for conservation and for livestock disease control.

Figure 1.1

Map of Asia

Thesis Aims

The study aimed to answer the following questions:

- What are the effects of recent socio-political changes in Kazakhstan on livestock husbandry and veterinary practices, and hence on disease prevalence in Kazakhstan?
- Are there effective disease control measures and monitoring programs operating at present in Kazakhstan?
- Foot-and-mouth disease and brucellosis have been reported in saiga. Are these diseases or other major viral diseases currently circulating in the saiga population?
- What are the key factors that should be included in a model of FMD dynamics in a saiga population?
- Are infectious diseases a cause for concern for the saiga population, or could they be so in the near future?
- How have the socio-political changes in Kazakhstan in the last decade affected the saiga antelope with respect to their relation with humans, and are these changes likely

to have affected the interaction between livestock and saigas, through disease or other factors?

- If Kazakhstan embarks on a program to attempt eradication of brucellosis or FMD from the domestic livestock population, could saiga constitute a threat of failure to this program?

To attempt to answer these questions, baseline data were required. This necessitated collection of a large amount of information in the field:

- The current prevalence of major infectious diseases in the saiga population
- The current saiga population structure and ecology (sex ratio, age structure)
- The current prevalence of foot-and-mouth disease virus (FMDV) infection among domestic ruminants located in or close to saiga range areas and their current level of immunity against FMD, and the important factors that are associated with immunity
- The current prevalence of brucellosis infection among domestic ruminants located in or close to saiga range areas, and the important factors that are associated with infection
- The prevalence of other major infectious diseases among domestic ruminants located in or close to saiga range areas, and the important factors associated with infection
- Changes in disease control policies after independence
- Changes in the veterinary care of livestock after independence
- The reliability of official statistics
- Meteorological data for modelling the spread of FMD

Data were collected from two saiga populations, one of which is subject to heavy human pressure and the other of which is relatively undisturbed. This comparison allowed an assessment of the effects of human disturbance on saiga ecology and the prevalence of infectious diseases. Data were also collected for livestock from villages outside the saiga range, for comparative purposes and in order to assess veterinary procedures in the case of a disease outbreak. These data were then analysed using a range of laboratory methods, statistical and epidemiological models in order to provide answers to the questions listed above. The t-test was used to test the significance between means, and the χ^2 , Mann-Whitney U, Fisher exact and Kruskal-Wallis H tests (Mead & Curnow, 1983; Bland, 1987; Fowler *et al.*, 1998) were used to test the significance between proportions, where $p < 0.05$ was considered significant. Throughout the thesis, $p < 0.05$ is abbreviated as *, $p < 0.01$ as **

and $p < 0.001$ as ***. Where an expected value for χ^2 was less than 5, Fisher exact results were used.

The transliterations of place names have been done using the Russian spelling, rather than the Kazakh spelling. Many place names have 4 or 5 different, but similar spellings. In these cases the most recent Russian version is used throughout the thesis, and no reference is made to other ways of spelling. In cases where the name of a place has been changed the old name is in brackets after the new name. Appendix 1 contains the transliteration method.

Chapter 2

The Saiga Antelope (*Saiga tatarica*)

2.1 Introduction

The saiga antelope (*Saiga tatarica*, Pallas) is a nomadic herding species, found in the semi-arid deserts of Central Asia: in Kazakhstan, Russia and Mongolia. It is about the size of a domestic goat; its most striking feature is the protuberant nose (Figure 2.1). The saiga is listed in appendix 2 of CITES (Convention for International Trade in Endangered Species), and is classified as 'vulnerable' by the IUCN (World Conservation Union).

The saiga belongs to the order Artiodactyla and the family Bovidae (Nowak, 1991). There are two sub-species: *Saiga tatarica tatarica* in Russia and Kazakhstan, and *Saiga tatarica mongolica* in Mongolia. *Saiga tatarica tatarica* are found in four distinct populations, three of these are in Kazakhstan, the fourth is in Kalmykia, Russia. Currently the populations in Kazakhstan make up more than 80% of total saiga numbers (Bekenov *et al.*, 1998). This study concentrates on the populations in Kazakhstan, in particular the Betpak-dala and Ustiurt populations.

There are few sources of data on saiga biology, and values for biological parameters tend to be stated in the literature without supporting data or confidence limits. Most of the published work draws heavily on the findings of Bannikov *et al.* (1961) and thus applies to the Kalmykian population rather than the Kazakh. Some aspects of the biology of the Kazakh population (e.g. longevity, fecundity, herd structure) are different from the

Kalmykian population (Fadeev & Sludskii, 1982). The discussion below is based on the review of saiga ecology by Bekenov *et al.* (1998) unless otherwise indicated.

2.2 Saiga range area, past and present

Saigas inhabit the steppe, semi-desert and desert. They require an even terrain with watering places and pasture with low-growing vegetation. In the 17th century saigas were found from the Polish border to mid-Mongolia (Milner-Gulland, 1991). There was heavy hunting in the 18th and 19th centuries, but saigas were still numerous and wide spread in and around Kazakhstan in the first half of the 19th century (Figure 2.2). In the second half of the 19th century widespread hunting caused the numbers of saiga to fall everywhere, and its range area consequently decreased in size (Sludskii, 1955). This continued into the early 20th century, driving the species to the verge of extinction.

Fig_2_1

Fig 2.1

Fig 2.2

A hunting ban was introduced in 1919 (Zhemchuzhnikov, 1926), but it was the rural depopulation caused by enforced collectivisation of the peasants in the years after 1929 (Asanov & Alimaev, 1990) that allowed saiga numbers to recover (Bannikov *et al.*, 1961; Zhirnov, 1982; Milner-Gulland, 1991). In the late 1930s they appeared again in areas they had previously inhabited (Sludskii, 1955). It is clear that saigas are vulnerable to overhunting; however they have the ability to rapidly recover their numbers. The increase in population size and geographical range carried on into the 1960s, when saigas appeared for the first time on *sovkhozes* (state farms). There was some worry among Soviet scientists that transmission of diseases could occur between saigas and livestock as they were sharing pastures and water points (Kindyakov, 1967; Starchikov, 1971).

Between 1960 and 1990 the geographical range area of the saiga remained essentially the same, although suitable habitat within this area gradually decreased due to human settlements, land cultivation etc., and gaps appeared between the main range areas of Betpak-dala, Ustiurt and Ural (Figure 2.3). A survey of 14,000 marked saiga calves between 1986 and 1993 showed that these three separate populations did not mix during this period (Grachev & Bekenov, 1993).

The overall area of the saiga's summer range in Kazakhstan is 300,000 – 350,000 km² (almost ?of the size of Spain); that of the winter range is 49,000 – 105,000 km². In central

parts of the summer ranges population density is usually 5 – 20 animals per km², but may sometimes reach up to 50 – 80 animals per km². The density in winter is much greater and may reach up to 140 per km². This may be a cause of increased contact, and possibly competition, with domestic livestock during the winter months. The Betpak-dala winter range is the largest of the three populations, covering 39,000 – 78,000 km² (usually about 60,000 km²). It has, in the past, been home to the largest saiga population; population density in winter averages around 45 saigas per km² (Fadeev & Sludskii, 1982). In comparison, the winter range of the Ustiurt population covers only 2,800 – 12,300 km², and population density is 11.6 – 19.5 saigas per km² (Fadeev & Sludskii, 1982).

2.3 Population size

Aerial and ground counts of saigas were made for the first time in the period 1950-54; these estimated the total population in Kazakhstan at 900,000 (Sludskii, 1955). Annual aerial counts of saiga populations began in 1954, when licensed hunting of the saiga for commercial purposes was introduced. Figure 2.4 illustrates the changes in estimated saiga numbers in Kazakhstan between 1961 and 1994 (Table A2.1, Appendix 2).

Fig_2_2

Figure 2.3

Fig 2.4

The highest numbers of saigas recorded in Kazakhstan were observed during the periods 1958 – 1960 and 1971 – 1974 (Fadeev & Sludskii, 1982; Bekenov *et al.*, 1998). During the interval between these peaks, population numbers decreased as a result of bad winters, summer droughts and intensive harvesting. The most drastic reduction took place in 1975 when 500,000 saiga were killed during the hunting season (Fadeev & Sludskii, 1982).

The estimates of saiga population sizes from 1954 to the present day were calculated by scientists at the Institute of Zoology (part of the Kazakhstan Academy of Sciences) in Almaty, and were based on aerial surveys performed in April every year. Unfortunately, the estimates are given without supporting data or confidence limits, and the methods used for the aerial surveys are not described in the literature; it is thus difficult to assess their accuracy. According to Iu.A. Grachev (senior scientist at the Institute for Zoology and Animal Genetics, Almaty), the surveys were performed in the same manner every year. A transect method was used, with photography of herds to aid counting. It thus seems likely that they are reasonably precise, although there is no way of assessing their inaccuracy.

According to Caughley *et al.* (1976), the most potent source of bias in any form of census by aerial survey is counting bias. They found that aircraft speed, height above the ground and strip width were the most significant factors of counting bias. Unfortunately, none of this information is available for the saiga censuses. There are several other sources of error in estimation of population size; one of the main errors is missing individual animals, often caused by observer error (Caughley & Sinclair, 1994). The geography of the semi-desert is such that an individual saiga is unlikely to be hidden by vegetation, but saiga colouring could make them difficult to spot at certain times of year. Censuses are timed to coincide with the periods when the saiga's coat contrasts with the vegetation and when they are aggregated into herds, thus minimising the chances of underestimation. A further cause of sampling error is related to animals not being evenly distributed over an area, however this is less of a problem with the transect method than with the quadrat method of survey (Norton-Griffiths, 1978). The enormous size of the range area and the uneven density of saigas mean that it would be easy to miss large herds, and thus underestimate the population size, if the entire area was not covered by the survey. On the other hand, if by chance the area surveyed was where saiga density was at its highest, an overestimation of population size could occur.

The individual saiga populations have been under different pressures, and are, for clarity, depicted in separate graphs (Figure 2.5a – c). Weather conditions often vary greatly in the different ranges, e.g. in Betpak-dala a severe winter caused a large-scale die-off 1971 – 1972, whereas the same year in Ustiurt and Ural higher than normal mortality was not experienced. In the Betpak-dala range area there are more human inhabitants than in the Ustiurt range area, thus poaching is likely to be greater there. In Betpak-dala the soil is richer and the number of livestock is far greater, thus saiga in Betpak-dala are more likely both to contact domestic livestock and to experience disturbances from humans. This may have an indirect effect on the saiga through competition for forage and transmission of endoparasites and infectious diseases from domestic livestock sharing the same pastures.

2.4 Seasonal changes in herd size

The saiga has a discrete yearly cycle, with rutting occurring in December and the vast majority of females giving birth within the space of 10 days in May (Fadeev & Sludskii, 1982). During the rut, saigas are mainly organised into small harem herds of 50 or fewer animals. After the rut harem herds join together, forming larger groups of 51 – 500 animals

(Fadeev & Sludskii, 1982). Herds are usually mixed, although there may be some smaller groups of only males, often those weakened by the rut.

Towards spring, saigas congregate in the calving areas, forming groups of tens of thousands of animals which disperse over the entire calving area without forming distinct herds. Up to 70% of the population may congregate in one area. The main calving areas in Betpak-dala cover an area 400 x 700 km, in Ustiurt 300 x 300 km and in Ural 200 x 300 km. The number of animals gathered to calve varies from 10,000 to 200,000 (usually 50 – 100,000), the population density at this time of year ranges from 5 – 600 animals per km², (usually 15 – 400). There are very few smaller groups outside of these large congregations.

After calving, saigas continue migrating towards their summer range. Some years the entire group moves at once, other years they move in stages as the calves grow old enough to follow their mothers (at roughly 10 days of age). In summer, the saigas gradually disperse, forming smaller herds. In June 30% of herds consist of less than 10 individuals, and only 2.9% have more than 500 individuals (Figure 2.6). Towards autumn, saigas gather in larger herds in preparation for their mass migration south. In August 18.9% of herds contain more than 500 animals. By October, 28% of herds consist of more than 500 saigas, and it is common to find groups of several thousand animals. By November, migration is mostly over and small harem herds begin to form (Fadeev & Sludskii, 1982).

Figures 2.5 a - b

Fig_2_3

Fig_2_4

Figure 2.5 c

Figure 2.6

Saiga herd structures are extremely fluid; groups often disperse and new ones assemble, although family groups (females with calves) are more stable. The fluidity of saiga herds increases the risk that infectious diseases are passed on to large numbers of individuals. Although the density of saigas in their summer range is characteristically much less than in their winter range, circulation of FMDV in the saiga population throughout the summer has occurred (Kindyakov *et al.*, 1973).

2.5 Migrations

Being migratory animals, saigas characteristically change their habitat according to the season. They migrate between their summer and winter ranges prompted by their need for new pastures or the presence of deep snow. The periods, routes, distance and speed of migration vary from year to year and in different areas, depending on climatic conditions, the number of watering places, the degree of disturbance experienced by the animals, various artificial obstacles on migration routes etc.

The saiga winter ranges are in the deserts south of latitude 48° (Figure 2.3). The chief factor restricting winter distribution is the depth and density of snow cover. Seasonal migration occurs in spring and autumn. Saigas migrate north to north-west in early spring, briefly halting their migration to gather in enormous herds for calving in early May. The position of spring calving varies from year to year because it is related to the distance travelled by saigas moving north from their winter range. This distance varies depending on the temperature, rainfall, condition of pastures and the accessibility of watering places. The most important factor is the distance away from human inhabitation. Females continue migrating towards the summer range when the calves grow big enough to leave. Some males stay with the females, others form separate herds migrating ahead. In general they have reached the summer ranges by June.

The summer ranges are located in semi-desert regions, at a latitude of 48-49°. The general direction of the autumn migration is south to south-east. In autumn, saigas gather in large herds in preparation for migration, which occurs with intervals of hundreds of kilometres separating groups of migrating animals. There are several different migration routes used. The spring migration routes are roughly similar to the autumn routes, but surveys using marked saiga calves have shown that some animals use other routes to return to their winter range (Grachev & Bekenov, 1993). The evidence suggests that animals form new groups in their winter and summer ranges, and that their migration routes do not remain constant from year to year. This regrouping is the result of various local migrations within the winter and summer ranges. These irregular movements are not confined to fixed times or directions, and in general involve only part of the population, sometimes occurring as a response to drought or deep snow, other times due to human disturbances.

Saigas generally migrate at a rate of 5 – 20 km per day, although this may increase to 40 – 45 km in poor weather conditions or if water is scarce. Females with young calves migrate

at a rate of 10 – 15 km per day. The Betpak-dala population covers 600 – 1200 km each way (as the crow flies) during migration, the Ustiurt population covers 300 – 600 km and the Ural population 200 – 300 km (Fadeev & Sludskii, 1982).

2.6 Reproduction

One of the ecological peculiarities of the saiga is its high rate of reproduction and recruitment. Females are sexually mature at 8 months of age and up to 95% produce young in their first year. Females can breed until death at around 10 years of age, often producing twins annually (Bannikov *et al.*, 1961). Triplets also occur, although this is not common. A mean of 0.92 (range 0.54 - 1.52, SD 0.239) embryos per juvenile (<1 year old) female and 1.74 (range 1.47 – 2.0, SD 0.138) embryos per adult female were recorded in saigas culled and necropsied between 1986 and 1996. Thus in years with a favourable climate the population size can increase rapidly; a population increase of up to 60% in a single year has been recorded. The high recruitment rate is also encouraged by the high percentage of breeding females in the population (Fadeev & Sludskii, 1982). Males become sexually mature at about 2 years of age, but they suffer a much higher mortality rate than the females, due to fighting during the rut and selective poaching. It is rare to find a male over 5 years of age.

2.7 Factors limiting population growth

2.7.1 Mortality caused by abiotic factors

Episodes of very high mortality are occasionally observed due to climatic conditions (Zhirnov, 1982). Saigas may become severely malnourished or starve to death when the snow cover is deep (30cm or more) or there is a layer of ice over the snow, usually in combination with low temperatures and strong winds. This set of climatic conditions is known as a *dzhut*. In the winter of 1971 – 1972 it was estimated that 400,000 saigas died in the southern part of Betpak-dala (Fadeev & Sludskii, 1982). In the winter of 1975 – 1976 at least 100,000 saigas died over an area of 15,000 – 20,000 km². Adult males weakened by the rut suffer especially high mortality in bad winters, when they may comprise 70 – 80% of the fatalities (Bannikov *et al.*, 1961).

Bad weather during the calving period may increase neonatal mortality, with calves dying from hypothermia or malnutrition because they are too weak to suckle. Summer droughts may also cause high mortality, mainly affecting females and calves. The effects are usually indirect, increasing mortality from predators, disease, lack of food etc. Mortality rates vary

greatly from year to year and between different populations; for example, in 1993 a mortality rate of 29.2% was estimated among saiga calves from birth to autumn in Betpak-dala, whereas in Ustiurt calf mortality was 82.9%.

Dzhuts occur approximately once every 10 – 12 years (Bannikov *et al.*, 1961), and summer droughts about 3 years in 10 (Milner-Gulland, 1994b). Climate induced mortality is limited somewhat by migration, and local migrations have been known to occur in direct response to bad weather conditions (Fadeev & Sludskii, 1982; Zhirnov, 1982).

2.7.2 Predation

Another factor that influences mortality is predation, mainly by wolves, and possibly by feral dogs or dogs owned by shepherds. Some authors suggest that wolves kill up to 25% of the saiga population (Rakov, 1956), but it is impossible to assess the impact of wolves without knowing the importance of weak or injured saigas and other species of prey and carrion to the wolf's diet (Filimonov & Laptev, 1975).

2.7.3 Diseases

Large-scale outbreaks of FMD were recorded in saigas between 1955 and 1974. Brucellosis has been recorded in the saiga, but the effects on the population are not known. Both FMD and brucellosis are discussed in greater detail in chapter 3. Necrobacillosis, caused by *Bacterioides necrophorum* (formerly *Fusobacterium necrophorum*), has been observed once in the saiga; it was thought to be the cause of death of about 5,000 – 6,000 saigas in Betpak-dala in the summer of 1978, when saigas had been sharing pasture with sheep (Petrov *et al.*, 1979). Strains of the plague and toxoplasmosis have been isolated from saiga remains (Peisakhis *et al.*, 1979; Galuzo *et al.*, 1963), but it is unclear whether saigas actually were carriers of these diseases.

When large numbers of saigas died in 1981, 1984 and 1988, pasteurellosis was thought to be the cause after *Pasteurella haemolytica* was isolated from many of the corpses (Fadeev & Sludskii, 1982; Aikimbaev *et al.*, 1985; Fadeev, 1986; Khakin & Sedov, 1992). The estimated number of deaths from pasteurellosis in the May 1988 outbreak was 270,000; most of these were calves and females in the birth area in Betpak-dala. *Pasteurella haemolytica* is a bacterium that occurs as a commensal organism in the upper respiratory and digestive tracts of a number of animal species (Carter & Chengappa, 1991). It is

commonly found in the nasopharynx of healthy cattle, sheep, goats and bighorn sheep (*Ovis canadensis*) (Aitken & Al-Sultan, 1985; Dunbar *et al.*, 1990; Wray & Thompson, 1973; Queen *et al.*, 1994; Carter, 1991; Foreyt & Lagerquist, 1994). In bighorn sheep it appears to act as an opportunistic pathogen (Queen *et al.*, 1994), e.g. when an animal has lowered immunity due to some form of stress or disease, *P. haemolytica* may invade, often causing pneumonia which kills the animal.

P. haemolytica has been isolated from nasal and tonsillar swabs in healthy saigas that on post-mortem examination had clinically normal lungs (Nametov & Lundervold, unpublished data). The mass mortality of saigas in 1988 that was attributed to pasteurellosis, was thought by some senior scientists at the Kazakh Veterinary Science Research Institute (*KazNIVI*) to have been caused by something the saigas encountered when grazing close to a military base. The whole area was closed off after the saigas died, and senior officials from Moscow were flown in to take part in the investigation. Scientists at *KazNIVI* felt the incidence was ‘hushed up’, and too quickly attributed to pasteurellosis.

Identification of the prevalence of *P. haemolytica* carriers is unlikely to be of any great significance with regard to preventing outbreaks of pasteurellosis. A more important factor would be to identify the primary causes of stress or disease that allowed invasion of *P. haemolytica*. This is difficult unless a disease outbreak is ongoing, and was therefore not attempted. Thus, although pasteurellosis is thought to be a significant cause of mass mortality in saigas, the study did not focus on this disease.

2.7.4 Density dependent and –independent factors

Natural populations of animals remain relatively stable, albeit fluctuating within limits, even though they have an intrinsic ability for rapid population growth (Sæther, 1997). Density dependence restricts population growth when competition for scarce resources lead to increases in the mortality or decreases in the fecundity of individuals (Caughley & Sinclair, 1994). Measuring density dependence is a difficult task in the absence of reliable information on the carrying capacity, which can be hard to estimate from field data. The relationship between the growth rate and the population density can be very difficult to quantify, making density dependence difficult to detect, even when it exists (Sæther, 1997).

Density dependent factors, although difficult to prove, have been recorded in numerous mammals, such as bighorn sheep, kudu (*Tragelaphus strepsiceros*), red deer (*Cervus elaphus*), reindeer (*Rangifer tarandus*), roe deer (*Capreolus capreolus*), Soay sheep (*Ovis aries*), white-tailed deer (*Odocoileus virginianus*) and feral donkey (*Equus asinus*) (Skogland, 1985; Owen-Smith, 1990; Clutton-Brock *et al.*, 1991; Gaillard *et al.*, 1998). Density dependence is accepted to occur in the saiga, although the evidence for it is inconclusive (Milner-Gulland, 1991). Lyudvigovich (1974) demonstrated a linear relationship between rainfall in the summer before conception and the average number of calves per female saiga on Barsa-Kel'mes island. This may be an effect of forage limitation caused by density dependence, as the animals could not leave the island. Data from the Kalmykian saiga population indicate that the number of calves per female only decreased at high population densities, as might be expected in a herding mammal, however, there were few data points. Additionally, other influences, such as climate, were not controlled for (Milner-Gulland, 1991). Coulson *et al.* (2000) recently demonstrated density dependent reduction in fecundity of the saiga through an increase in the number of singletons and a decrease in the proportion of twins born. This may be related to the females' poor body condition at conception, caused by forage limitation in the autumn. The fact that female fecundity is strongly affected by body condition before mating has been documented for other species (Owen-Smith, 1990). In domestic ewes, it has been reported that improving nutrition can increase ovulation rate (Downing *et al.*, 1999; Perez *et al.*, 1997). However, the effects of poor body condition may be indirect, e.g. through decreasing fertility and increasing resorption of embryos.

Juvenile survival varies considerably from year to year and across different populations and species (Gaillard *et al.*, 1998), and thus seems highly sensitive to limiting factors, regardless of whether these are stochastic or related to changes in population density. Adult survival across different populations and different species generally varies little from year to year, and thus seems buffered against most limiting factors. Gaillard *et al.* (1998) reported density dependent adult survival in Soay sheep. The Soay sheep and the saiga antelope are similar in that both populations show persistent instabilities with up to 60% of the population dying in some years. Density dependent adult survival has not been demonstrated in the saiga, however, the data are insufficient for analysis, so it cannot be assumed not to exist.

Fowler (1981) presented evidence suggesting that the population growth rate of ungulates was most affected by the number of animals present when the population size reached a value of about 0.6 of the carrying capacity, i.e. density dependence was not linearly related to population size. In Soay sheep and African buffalo (*Syncerus caffer*), the major effects of population density occur in a non-linear way at high population densities (Sinclair, 1977; Clutton-Brock *et al.*, 1991; Grenfell *et al.*, 1992). Similarly, in wild reindeer (*Rangifer tarandus*), adult mortality increases only when the winter food resources are severely depleted due to overcrowding (Skogland, 1985). If density dependence in the saiga is linear then some effects may be seen even at low population size, in which case available data can be analysed for density dependent effects. However, if density dependence in the saiga is not linear, like many other ungulate species, and if the saiga population has not reached 0.6 of carrying capacity in the last 50 years, then density dependence may not be detectable, given the poor data available. The maximum population size in Betpak-dala that was estimated by aerial counts was 995,000 in 1974 (Fadeev & Sludskii, 1982), however this is unlikely to be anywhere near the carrying capacity, considering that between 12% and 39% of the population have been culled (officially) every year since 1954. Indeed the saiga population may not have reached carrying capacity during the last 300 years, considering that they were hunted in large numbers as early as in 1690 (Vernad, 1939).

Saiga population size varies greatly during the year due to high mortality rates and a seasonal birth period of very short duration. As migratory animals with large areas in which to roam, saigas may not experience the same density dependent limitations that ungulate populations experience when in a confined area, such as a nature reserve or an island. It is, therefore, interesting that the saigas on Barsa-Kel'mes Island (a nature reserve in the Aral Sea) showed a clear relationship between rainfall and fecundity. Saigas can generally reduce the impact of climatic changes by moving to a different area. Of course they do not have unlimited space in which to roam. In early times they may have been limited solely by their habitat requirements, nowadays human competition is an essential factor. It is possible that there could be some density dependent effect in the winter range, where saiga density is higher, and competition with domestic livestock is greater than in the summer range. This density dependent effect could be related to the availability of winter forage, which could be exacerbated by a severe winter.

During the saiga calving season up to 200,000 animals congregate together for about 10 days, a behaviour which has probably evolved to minimise the risk of predation to each calf during this vulnerable time. Even at this high concentration, density dependent neonatal mortality is unlikely to outweigh the risks of predation (Milner-Gulland *et al.*, in review). Calves can follow their mothers by 10 days of age, and saigas will then move on if there is insufficient forage. However, if the dam is in poor body condition due to forage limitation in winter caused by density dependence, the calf may be born weak, and additionally the dam may produce very little milk. In this way density dependent mortality of neonatal saigas could occur.

Another potential cost of aggregation in the birth areas is that the possibility for transmission of disease and parasites is much greater. An increase in the number of nematode eggs in the faeces around parturition (peri-parturient rise) is common in many host species, in particular ewes, sows and goats (Urquhart *et al.*, 1987), and is likely also to occur in saigas. In sheep this seems to result from a temporary relaxation in immunity around parturition (Urquhart *et al.*, 1987). The importance of the peri-parturient rise in livestock is that it may cause loss of production in lactating animals, and clinical disease in susceptible young stock. In the calving areas of the saiga, the conditions are generally ideal for parasite transmission; high density of hosts and warm wet weather. High parasite burdens could theoretically cause a decrease in milk production of the dam. Following a severe winter, adults may be in poorer than average body condition at the time of calving, and parasite loads that under normal circumstances are tolerated may cause clinical disease. Lowered immunity is common in nutritionally stressed animals, making them more susceptible to disease (Mims *et al.*, 1998). A disease such as FMD will spread extremely rapidly in a densely populated area, and may cause huge losses even in a healthy population.

The population dynamics of ungulates are characterised by a strong influence of density independent factors (Sæther, 1997), in particular climatic conditions. Density independent factors are well documented in the saiga. Climate is the main limiting factor, affecting both mortality and fecundity. In Ustiurt, both the number of breeding females and the average number of embryos per female were noticeably lower after a severe winter in 1990/91. In 1992, when conditions were favourable, the fertility rate in the Ustiurt population was unusually high. Lower than average fertility was also seen in the Betpak-dala population

following a severe winter in 1993/94. Summer droughts may also affect fecundity - infertility is more common in drought years, especially amongst juvenile females.

One of the problems with the study of density dependence is that the effects of density independent factors may be influenced by population density. For example in Canada, female calf survival in red deer (*Cervus elaphus canadensis*), is influenced by both climate and population density (Sauer and Boyce, 1983). Wild reindeer herds show a relationship between calf survival rate and population density, which in turn is related to the amount of available forage during late winter (Skogland, 1985). In white-tailed deer, red deer and mountain goats (*Oreamnos americanus*), the recruitment rate (number of calves per female) is influenced by a combination of population density and winter severity (Sæther, 1997). Coulson *et al.* (2000) showed that twinning rate of saiga antelopes in Betpak-dala significantly decreased with increasing population numbers and with decreasing winter temperature. These examples show how density independent factors may limit population size, alone or in combination with population density, in particular by effects on juvenile mortality and fecundity.

2.8 Interactions with humans and livestock

2.8.1 Interactions with domestic livestock pre-independence (1960 – 1991)

In most regions in Kazakhstan, saigas have shared pasture and watering places with cattle, sheep and horses for centuries. Shepherds move with their livestock several times a year; in summer they live in *yurtas* (round felt tents) in areas with access to water, and in winter they live in *zimovkas* (small houses with barns, built on pastures situated away from the main farm). Saigas may approach watering points used by the shepherds at night or early in the morning when the human inhabitants are asleep, however, they rarely have close contact with livestock. Under some circumstances, such as severe droughts or *dzhuts*, saigas have approached human settlements, even towns, in search of food.

According to Fadeev & Sludskii (1982) there is no real competition for forage between saigas and domestic livestock, as saigas eat vegetation which domestic livestock do not eat, and there is enough food for both domestic and wild animals. Certainly in the summer ranges this is true. In the saiga winter range, which is smaller than the summer range, there has historically been a higher concentration of both saigas and domestic livestock, in particular in the area of Betpak-dala between the Chu river and the Chu-Iliskiy hills, and in

the Moinkum sands (Figure 2.7). If the total number of animals (livestock and saigas) in the saiga winter range increases such that they reach or exceed the sustainable level of removal of vegetation there will be competition for food. In the Ustiurt range areas there have historically been fewer livestock (Kunaev, 1985), therefore possibly also less competition for forage.

Fig 2.7 (file fig2_5)

Soviet scientists have reported degradation of vegetation in some areas of Betpak-dala, especially around the river Chu (Babaev & Kharin, 1991; Zhambarkin, 1995). Zhambarkin (1995) suggested that there was a stocking rate of about 1.5 hectare per sheep in Moinkum in the 1980s, and that at this stocking rate the pasture would have provided the sheep with 45 – 55% of their needs, the rest being provided from grain feed. At these stocking rates there would probably have been competition for food between grazing animals (S. Robinson, pers. comm.), however it is difficult to say anything for saigas as they are so mobile; they have in some years moved far south, where there is no evidence for degradation.

Several scientists believed there was significant competition for forage between domestic livestock and saigas in the 1980s, especially in the winter range of the saiga where livestock remained on the pasture for long periods of the year (Babaev & Kharin, 1991; Zhambarkin, 1995). Livestock damaged the pasture, causing a loss of palatable plant species. However, recent analysis of satellite images of Betpak-dala taken between 1982 and 1994 shows that this may not have been the case (Sarah Robinson, pers. comm.).

2.8.2 Interactions with domestic livestock post-independence (1991 - 1999)

Since the independence of Kazakhstan in 1991, there has been a massive decline in livestock numbers (Kulekeev, 1998). The difficult economic conditions have led to rural-urban migration, further reducing human density in rural areas. Official statistics on human population change suggest that between a quarter and a fifth of inhabitants have left their villages since 1992 (Goskomstat, 1997). The dramatic changes in agriculture during recent years (discussed in chapter 4) may have a large impact on the saiga. There is greater concentration of livestock around the villages, rather than on the remote pastures, which may substantially reduce saiga contact and forage competition with livestock.

2.8.3 Management of saiga populations before 1988

Hunting saigas for commercial purposes in Kazakhstan has been licensed since 1954. In the 1960s it was carried out by specialised state-run hunting organisations, *promkhoz*s, which in 1973 came under central administration, 'Kazglavokhot'. This formed part of the hunting union 'Okhotzoprom', which in 1989 received exclusive rights over the hunting and commercial exploitation of the saiga. The *promkhoz*s were involved in protecting saigas; hunters received premiums for culling wolves in winter, and in spring they went to the birth areas to ensure that the saigas remained undisturbed. Although not officially involved in anti-poaching measures, hunters would apprehend any poacher they encountered and hand him to the relevant authorities (V.V Ukrainskii, pers. comm.).

The Institute of Zoology made yearly recommendations with regard to how many animals should be culled, and from which age and sex class. This was based on the spring census, estimates of fertility, mortality and population growth, analyses of sex and age structure, and evaluation of natural limiting factors.

2.8.4 Hunting methods

There are two main methods used for commercial hunting – hunting at night with powerful lights and hunting during the day using portable net traps. An experienced team of four or five hunters can harvest 100 – 150 animals during a five or six hour hunt. Net traps have the advantage that entire herds can be driven into them with the aid of motorcycles, allowing hunters to select the animals according to gender and age. This method is commonly used in the Ural and Ustiurt populations, but not in Betpak-dala, as the terrain is too flat for this method to be efficient (V.V Ukrainskii, pers. comm.). Each *promkhoz* has its own storage and preparation points, where the carcasses are taken to be dressed and prepared for sale.

2.8.5 Management of saiga populations after 1988

Before 1988, all legally killed saigas were sold to the state. In 1988 border controls with China were lifted, allowing the legal export of saiga horns to their principal market for the first time since 1921 (Milner-Gulland, 1991). Simultaneously, private co-operatives were allowed to set up trade. However, the hunting pressure on the saiga was so intense, and the prices so high, that for the 1990 season controls were reintroduced such that in Kazakhstan only the state registered co-operatives were allowed to hunt.

‘Okhotzoprom’ currently has exclusive rights over the hunting and commercial exploitation of the saiga. The Ministry of Ecology and Natural Resources grants the hunting permits, and decides when the hunting season opens (usually 1st October to 30th November). The Institute of Zoology is still responsible for making recommendations about the numbers culled, however, a lack of government funding in recent years has led to insufficient organised data collection, such that scientists at the Institute have little scientific evidence upon which to base their recommendations (A.B. Bekenov, pers. comm.). Even so, they recommended a ban on hunting in 1999, based on low population estimates acquired from incomplete aerial and ground surveys of the three populations. The ban was implemented by the Ministry of Ecology and Natural Resources.

2.8.6 Poaching before 1988

The saiga antelope has never received adequate protection from hunters due to the logistical problems associated with monitoring animals that migrate through areas covering 350,000 – 400,000 km² (nearly twice the size of UK). However, during the Soviet era there was at least a functional hunting inspection organisation, controlled by the Department of Hunting and Fishery. During this time, most people living in villages in the saiga range area had jobs, and only a small proportion of villagers might occasionally poach saigas for meat. The poachers left the horns outdoors to rot as there was no market for saiga horn because the border to China was closed (A. Grachev, pers. comm.).

2.8.7 Poaching after 1988

Poaching of saigas increased greatly after access to the Chinese market was gained in 1988. The rapid economic decline in Kazakhstan during the early 1990s, accompanied by a substantial increase in unemployment, encouraged young men to earn easy money by hunting saigas. Middlemen drove around most of the villages in the saiga range area, buying horns to sell onto the gangs who smuggled the horns across the border to China.

Whereas previously saigas were shot mainly in autumn and winter for meat, they were now shot all year round for their horns. Organised gangs of poachers used motorcycle teams to increase hunting efficiency. These poachers would often take only the horns, leaving the meat to rot. The reduced funding to the hunting inspection bodies meant they were unable to control the situation. Between 1989 and 1994 there was a 40% reduction in funding for hunting inspection (Iu.A. Grachev, pers. comm.). A major concern about selective poaching for horns is that the proportion of adult males may become very small, which can affect the population dynamics of the species (Milner-Gulland *et al.*, 1995).

By 1993 – 1994, there seemed to be a slight reduction in poaching, possibly because saigas became scarcer or because the price of horns fell. However, the continuing economic decline in Kazakhstan into the late 1990s has led to many ordinary villagers trying to supplement their income by hunting saigas. Meat is taken home to eat or sold to local women who take it to nearby towns to sell on the black market. Very few villagers receive any cash income, and many families have already eaten or bartered away most of their livestock, hence some families depend on saiga for their livelihood. As the economic situation continues to deteriorate, the situation may worsen and hunting may become more indiscriminate; there are already reports of calves and pregnant females being shot for meat (personal observation; W. Fitz, pers. comm.; anonymous poachers, pers. comm.).

In 1998 control of the hunting inspectorate was transferred to ‘Okhotzooptom’. Senior Figures in the former Department of Hunting and Fishery are privately concerned that this could lead to a conflict of interest, especially now that all hunting has been banned (S.M. Akhmetov, pers. comm.). Funding for hunting inspection is continuing to decline; by 1999 it had decreased by 80% with respect to the 1989 figures (Iu.A. Grachev, pers. comm.). The only way to scientifically assess the rate of poaching and its impact on the saiga would be to monitor saiga population size by performing complete, annual (ideally biannual) aerial surveys, to investigate the consumption of saiga meat by the human population and to have anti-poaching teams that keep accurate records of encounters with poachers.

Chapter 3

Diseases under study

3.1 Introduction

The diseases under study have been selected because of their known or potential importance to the saiga population, or because of the role saigas in Kazakhstan may play in the epidemiology of the diseases. Foot-and-mouth disease, brucellosis, bluetongue, epizootic haemorrhagic disease, rinderpest and peste-des-petites-ruminants are studied.

3.2 Foot-and-mouth disease

Pigs have been excluded from this review, as pigs were not sampled in the study. Extremely few pigs are kept in Betpak-dala, the main study area, and although pigs play a significant part in the epidemiology of foot-and-mouth disease in some countries (Report, 1990; OIE, 1999), they are unlikely to be of importance in Kazakhstan, a Muslim country. Unless otherwise stated, the information in sections 3.2.1 to 3.2.6 has been taken from a review of foot-and-mouth disease by Donaldson (1993).

Foot-and-mouth disease (FMD) was first described in Italy in 1546 by Hieronymus Frascatorius (R.P. Kitching, pers. comm.) It is perhaps the most contagious disease known to human or veterinary medicine, and for those countries free of the disease it is that most feared by the farming community and the veterinary service (Donaldson, 1987). FMD is an acute, febrile condition of cloven-hoofed animals and is characterised by the formation of vesicles in and around the mouth, on the feet and on the teats and mammary gland. Muscular lesions, especially in the myocardium, are common in young animals and may lead to death (Mohiyuddeen, 1958). FMD in adult animals does not usually result in a mortality rate of above 5% except in rare circumstances. However, in young stock, especially under conditions of dense stocking, mortality rates of up to 90% may result. In general, FMD is not thought to affect humans, however in 1966 one person in Kazakhstan was diagnosed with FMD type A, and another with type O (Kindyakov *et al.*, 1972).

3.2.1 The virus

The causal agent of foot-and-mouth disease is a virus of the Picornaviridae family. There are seven distinct serotypes of the virus (A, C, O, Asia 1, SAT 1, SAT 2, SAT 3) which do not cross protect against each other (Donaldson, 1987). The survival of airborne virus is mainly influenced by relative humidity (RH): above 60% RH virus survives well, below 55% it will be quickly inactivated (Donaldson & Ferris, 1975). Wind-borne spread can occur over considerable distances if climatic factors are favourable (most importantly RH

higher than 55 – 60%) (Gloster *et al.*, 1981; Donaldson *et al.*, 1982). At temperatures below freezing point the virus is stable almost indefinitely (Donaldson, 1987).

3.2.2 Diagnostic Techniques - Antibody assays

Antibody assays are used to determine evidence of past infection or vaccination. The classical procedure of virus neutralisation (VN, also termed serum neutralisation) has been applied for many years (Anderson, 1978). In recent years, enzyme-linked immunosorbent assay (ELISA) procedures have been applied to FMD serology. The liquid-phase blocking sandwich ELISA by Hamblin (1986) was designed to mimic the traditional VN test, and appears to do so. The ELISA is sensitive, more specific to serotypes, and results are more reproducible than those obtained by VN (Hamblin *et al.*, 1986). This is the test which is most widely accepted and best validated, and is used at the World Reference Centre for Foot and Mouth Disease at Pirbright Laboratories, UK. The interpretation of individual positive results needs to be made cautiously, as spurious results may occur, however, there is no indication that false negatives occur (Forman, 1993).

Current serological tests for FMD detect antibody to the structural, capsid proteins of the virus. Antibodies to the capsid proteins are induced by both vaccination and infection. A profiling ELISA has recently been developed to differentiate between vaccinated and infected animals. The test detects antibody to the non-structural (NS) proteins Lb, 2C, 3A, 3D and the polyprotein 3ABC, of FMD virus. Mackay *et al.* (1998) showed that sera from naive cattle, experimentally infected cattle and vaccinated cattle could be differentiated on the basis of the presence or absence of antibody to the structural and/or NS proteins. The assay was validated with field sera, which demonstrated that antibodies to 3ABC and usually one or more of the other non-structural proteins are detected only in animals reported to have shown clinical signs of FMD (Mackay *et al.*, 1998).

3.2.3 World wide distribution

The World Organisation for Animal Health (OIE, formerly Office International des Epizooties) publishes a list of notifiable diseases, grouped by classification. List A are transmissible diseases which have the potential for very serious and very rapid spread irrespective of international borders. List B are transmissible diseases considered of public health importance within countries, and which are significant in the international trade of animals and animal products (OIE, 1999a). FMD is classified as list A, and according to

the OIE, FMD remains a threat to all continents. In general, FMD type C is found in South America, central Africa and occasionally in the Middle East. Types A and O are mainly found in South America, northern Africa, the Middle East, Europe and Asia (including the central Asian republics, Figure 3.1). Asia 1 is found in the Middle East and south-east Asia, the SAT types are found in central and southern Africa (B. Newman, pers. comm.).

In 1998 and 1999, Oceania, North and Central America, as well as western Europe were FMD free (Figure 3.2). Eastern Europe, Asia, Africa and South America all suffered episodes of foot and mouth disease (OIE, 1998a); . FMD continued to be enzootic throughout most of the Middle East in 1998, and was present in the countries of south and south-east Asia (OIE, 1999b).

The FMD situation in the Commonwealth of Independent States (CIS), in particular the Transcaucasian area, has been of great concern to the OIE since the identification in Armenia of a new strain of FMD type A very close to a variant from Iran for which, at the time of writing, there is no vaccine available (FAO, 1998b). Outbreaks of FMD in Central Asia are discussed in chapter 4.

3.2.4 Pathogenesis

FMDV most commonly infects via the respiratory route, especially in ruminant species, where very small doses can initiate infection (Sellers, 1971). Apart from the respiratory route, infection can also occur by ingestion, or when there is a break in an animal's integument, i.e. its skin or mucosa. Following replication in the mucosae and associated lymphoid tissues of the upper respiratory tract, virus enters the blood stream in which it may circulate for three to five days. A secondary phase of replication is initiated by blood-borne virus in organs such as lymph nodes, adrenals, kidneys, mammary gland and the epithelial tissues in and around the mouth and feet where vesicles are produced.

3.2.5 Transmission and Immunity

An important feature of FMD is that virus excretion occurs before infected animals manifest clinical signs, thus movement of incubating animals is a hazard. The length of the incubation period is variable, and depends mainly on the virus strain, dose of exposure and the route of entry. With natural routes and high exposure doses the period can be as short as 2 to 3 days, or up to 10-14 days if the exposure doses are low (Donaldson, 1987).

During the acute phase of disease, which generally lasts 3-4 days, all excretions, secretions and tissues contain virus. The duration of protective immunity (to homologous challenge) in unvaccinated cattle and sheep is effectively lifelong (R.P. Kitching, pers. comm.).

Map 3.1

Fig 3.1

Fig 3.2

Animal movement is by far the most important mechanism of transmission of FMD, followed by movement of contaminated animal products such as milk, meat, offal, untreated hides and skins. Next are people who have been in contact with incubating or diseased animals, and vehicles that have been used to transport them. Finally airborne spread and spread by carriers, but these are less frequent and require a particular sequence of events. In Kazakhstan, outbreaks of FMD have often coincided with the annual migrations to and from summer pastures, when livestock are moved along migration corridors situated adjacent to the territories of other farms (Omirezhanov *et al.*, 1976). Similarly, dairy farms near migration routes in Saudi Arabia frequently experience outbreaks of FMD soon after Bedouin tribes have moved by with their animals.

Cattle are more readily infected by airborne virus than other species, possibly due to their higher tidal volume (air-intake per breath). They usually exhibit very clear symptoms of FMD, unlike sheep and goats which often have mild or unapparent signs, easily missed. Sheep transported from market to market have caused outbreaks in cattle that had been through the same markets e.g. in Morocco in 1983. In Central Asia, subclinically infected sheep bought at a market in Uzbekistan were identified as the source of the outbreak of FMD in cattle in Kazakhstan in May 1998 (Zh.O. Omirezhanov, 1998, pers. comm.).

Within herds, transmission is most likely by close contact between animals. High stocking densities facilitate spread of FMD virus, because a high level of challenge is maintained both from infectious animals and the environment. This was thought to have been the cause of the rapid spread of FMD on a state farm in southern Kazakhstan in November 1971, after an outbreak occurred when infected cattle were bought from a farm in a neighbouring administrative district (*raion*) (Kindyakov *et al.*, 1972).

One feature during outbreaks in cattle has been a high attack rate amongst in-calf heifers and first-lactation cows, in spite of multiple vaccination. The underlying causes have not been resolved, but heat stress, underlying infection, hypersensitivity and reduced immunity through pregnancy may all have been involved. Higher incidence in pregnant animals has also been observed among saigas in Kalmykia (Russian Federation), where an epidemic of FMD in 1957 killed many females in the late stages of pregnancy, but only one infected male was found (Smirnov, 1960).

The occurrence of FMD at a critical period of a husbandry cycle may also have serious consequences. For example, in the 1989 – 90 epidemic in Tunisia, 50,000 lambs died when FMD struck shortly after the beginning of the lambing season. This does not only occur in farmed livestock, it may occur in any situation where animals are at high density during parturition. For example, in the spring of 1967 an estimated 50,000 saiga calves died following an outbreak of FMD during the period of mass calving in Betpak-dala (Fadeev & Sludskii, 1982).

3.2.6 Seasonality

The seasonal pattern of outbreaks in parts of Africa has been explained by Rweyemamu (1970) and Dawe (1978) as a consequence of climate and increased animal contact. During the dry season animals congregate at a few watering places and there is an increased opportunity for dissemination of virus. Rweyemamu (1970) observed that the first foci of an epidemic in Tanzania began in an area where there was opportunity for contact at watering points between livestock and wildlife towards the end of the dry season. Dawe (1978) suggested similar factors might explain outbreaks in Malawi. In Kazakhstan, saigas are known to share watering points with domestic livestock, and it was believed that the outbreak of FMD among saigas in Betpak-dala in 1969 was probably from water (Starchikov, 1971).

In Assam, India, Chakrabarty *et al.* (1979) found that outbreaks of FMD were more common in winter and the monsoon. He attributed the winter epidemics to climatic conditions favouring virus survival, and the monsoon epidemics to the fact that floods forced movement of livestock to the highlands where large congregations were herded in close contact with wild herbivores. In Pakistan, Haq (1951) related the occurrence of FMD in the spring months to the increase in animal movement at this time of year. Several authors have noted increased FMD outbreaks in relation to the mass movement and congregation of animals during specific times of year (Ramaraq, 1988; Sharma *et al.*, 1991; Voh *et al.*, 1993; Woloszyn, 1952). Plotnikov (1972) analysed seasonal incidence data from Kazakhstan, Uzbekistan and western Siberia and found that there were two major peaks of disease incidence, one in early summer and one in late autumn. These periods coincided with the annual migrations to and from summer pastures. Kindyakov (1972) and Omirzhanov (1976) also observed an increase in FMD outbreaks in Kazakhstan during these periods.

Dawe (1978) suggested that rough, abrasive grazing ground may have an influence on the incidence of FMD by causing injuries to mucosal and epithelial surfaces, producing additional entry points for virus. This could be a contributing factor to some of seasonal variation in disease outbreaks. There is a possibility this could occur in the autumn months in Kazakhstan, as the vegetation on the steppe is more abrasive at this time of year (personal observation).

3.2.7 Vaccination and control

In South America farmers protect their livestock herds from infection with FMDV by erecting double fences (Donaldsen, 1993) which prevent direct contact between their livestock and animals in transit. In South Africa, game fences are used to protect livestock from infection by contact with wildlife (personal observation).

The aim of vaccination is to protect animals against the production losses that FMD may cause. Vaccination will not necessarily prevent immunised animals that are exposed to infection from replicating and excreting virus (C. Hamblin, pers. comm.). There have been reports of disease outbreaks in vaccinated livestock (Woolhouse *et al.*, 1996), most recently in south-eastern parts of Kazakhstan (A. Namet, pers. comm.). This may be due to the gradual antigenic transition of virus strains away from parent strains which seems to occur in the field situation, rendering vaccines less effective (Kitching *et al.*, 1989). Outbreaks in vaccinated young stock may be related to interference by maternally-derived antibodies. Susceptibility to infection precedes the ability to respond to vaccination in the presence of maternally-derived antibodies (Kitching & Salt, 1995).

3.2.8 FMD carriers

Animals in which FMDV persists for more than 4 weeks are commonly referred to as carriers (Salt, 1993). However, not all persistently infected animals are potentially able to transmit the disease (Thomson, 1996). After recovery from FMD, up to 80% of cattle and 15-20% of sheep may become persistently infected (Sutmoller & Gaggero, 1965; Burrows, 1968; Sharma, 1978; Salt, 1993; Thomson, 1996). The carrier-state can also become established in immune animals. There is a possibility that these carriers can initiate fresh outbreaks when brought into contact with fully susceptible animals, but transmission from livestock carriers to susceptible livestock has not been reproduced under experimental

conditions (Thomson, 1996), even when the carriers were stressed by road transport (Sellers, 1971). There has been laboratory confirmation from field samples that cattle and African buffalo carrying FMD may have initiated new outbreaks of disease (Dawe *et al.*, 1994; Hargreaves, 1994), however there are no unequivocal reports of transmission of FMDV from persistently infected domestic livestock to susceptible animals. However, sub-clinically infected, vaccinated cattle can transmit FMDV to susceptible animals for up to 7 days post-infection (Donaldson & Kitching, 1989). Field evidence has shown that routine vaccination of cattle reduces the establishment of carriers in endemic areas (Anderson *et al.*, 1974), probably indirectly by reducing clinical cases and thus challenge doses of FMDV (Salt, 1993).

The duration of the carrier-state varies with the host species, the strain of virus and possibly other factors. The maximum recorded periods for different livestock species are over 3 years in cattle, 12 months in sheep and 4 months in goats (McVicar & Suttmoller, 1972; McVicar & Suttmoller, 1968; Sharma, 1978; Singh, 1979; Thomson, 1996). Pigs do not become carriers (Salt, 1993).

3.2.9 FMD in wildlife

In the early 1900s in southern Africa it was thought that FMD was predominantly a disease of cattle, which spilt over into the surrounding wild ungulate populations (Donaldson, 1993). However, as more focal outbreaks occurred in the absence of cattle movement and in the presence of wild ungulates, this theory was dispelled (Bengis *et al.*, 1987). Large-scale serological surveys conducted in southern Africa in the late 1950s and early 1960s showed that nearly all the buffalo (*Syncerus caffer*) populations had high antibody titres to one or more of the SAT type FMD viruses, whereas only a few individuals of other species showed antibody titres; typically of a much lower order (Bengis *et al.*, 1987; Condy *et al.*, 1969). Hedger (1976) reported virus isolation from 50 – 65% of wild African buffalo sampled. More recent studies have also found high antibody titres in buffalo (Falconer & Child, 1975; OIE, 1998d; Paling *et al.*, 1988).

Extensive research work in southern and eastern Africa in 1960s and 1970s, showed that only buffalo remain long term reservoirs of virus after FMD infection, although clinical disease in buffalo is uncommon (Hedger, 1976; Anderson *et al.*, 1979). The African buffalo is now known to act as a significant reservoir of FMDV (serotypes SAT 1, SAT 2

and SAT 3), which may infect domestic livestock (Bengis *et al.*, 1987; Dawe *et al.*, 1994). It is generally accepted on circumstantial grounds that African buffaloes are the major source of FMDV infection for domestic livestock in southern Africa (Thompson *et al.* 1992; Condy *et al.* 1985; Vosloo *et al.* 1995; R.P. Kitching, pers. comm.). In a survey of wildlife in Tanzania, the presence of antibodies to FMDV serotypes A and O were found in 90 – 100% of buffalo sampled in a Katavi National Park, an area almost devoid of cattle (Hamblin *et al.*, 1990). It has not been established how these buffalo may have become infected with types A and O.

The African buffalo has been shown to carry virus for up to 5 years (Hedger *et al.*, 1972; Condy *et al.*, 1985), but its ability to transmit virus to other buffalo or cattle is limited (Condy & Hedger, 1974; Hedger, 1976; Anderson *et al.*, 1979; Anderson, 1986; Bengis *et al.*, 1987; Bengis *et al.*, 1986; Vosloo *et al.*, 1995). Transmission from carrier buffalo to cattle has been demonstrated experimentally (Hedger & Condy, 1985; Dawe *et al.*, 1994), but in the experiment by Hedger & Condy, buffalo and cattle had been kept in close contact for two years, an extremely unnatural situation in the wild. Only one outbreak of FMD in cattle in Africa has been reported as initiated by buffalo; based on laboratory findings it was concluded that buffalo were the source of an outbreak in Zimbabwe where they had been sharing a watering hole with cattle (Dawe *et al.*, 1994).

FMD has been reported in water buffalo (*Bubalus bubalis*, *Bubalus arnee*) (Natarajan *et al.*, 1993; Akhtar & Haq, 1993; Cleland *et al.*, 1995), which have been shown to carry FMDV for up to two months (Moussa *et al.*, 1979; Samara & Pinto, 1983), furthermore there is indirect evidence for the virus persisting for up to two years (Thomson, 1996). Camels (*Camelus dromedarius*) are susceptible to FMDV, but infection remains unapparent (Fassi-Fehri, 1987; Higgins, 1986; OIE, 1987). Camelids in South America, such as llama (*Lama glama*), alpaca (*Lama pacos*) and vicuna (*Vicugna vicugna*), are also susceptible to FMD (Espinoza & Ameghino, 1993; Fassi-Fehri, 1987), but their role in its epidemiology is unclear. In the West Bank (Palestinian Authority) in 1994, there was an outbreak of FMD in a holding of 36 gazelles (species not mentioned) and ibexes, of which 20 died (OIE, 1995). There was no evidence that the gazelles in the West Bank acted as reservoir of disease, however other wildlife species such as wild boars may act as disseminators of FMDV (OIE, 1995). Outbreaks of FMD in saiga antelopes were reported in the USSR during the 1950s and 1960s, these are discussed in section 3.2.10.

A survey of a mixed farm in Kenya in 1988 showed antibodies to FMDV in cattle, sheep, goats and eland (*Taurotragus oryx*), but not in oryx (*Oryx beisa*) or in camels (Paling *et al.*, 1988). Antibodies to FMDV have also been demonstrated in bushbuck (*Tragelaphus scriptus*), hartebeest (*Alcelaphus buselaphus*), impala (*Damaliscus korrigum*), sable (*Hippotragus niger*), springbok (*Antidorcas marsupialistopi*), tsessebe (*Damaliscus lunatus*), waterbuck (*Kobus defassa*, *Kobus ellipsiprymnus*) and wildebeest (*Connachaetes taurinus*) (Condy *et al.*, 1969; Falconer & Child, 1975; Paling, *et al.*, 1979; Paling *et al.*, 1988; Hamblin *et al.*, 1990); Anderson *et al.*, 1993).

Experimental infection with FMDV causes clinical disease and measurable antibody response in impala, eland, greater kudu, sable, warthog (*Phacochoerus aethiopicus*) and bush pig (*Potamochoerus porcus*) (Hedger *et al.*, 1972; Ferris *et al.*, 1989). Mild clinical disease has been observed in wildebeest (Anderson *et al.*, 1975), but in an experiment by Hedger *et al.* (1972) none of the four wildebeest succumbed to artificial or natural infection, nor were antibody responses recorded.

Cyclical epidemics in impala are believed to arise from contact with buffalo (Bengis *et al.*, 1987; Dawe *et al.*, 1994). In November 1995, FMDV type SAT 2 was reported in impala in the Kruger National Park (Republic of South Africa). It spread through contiguous impala herds along two main water courses. Infection rates of 30 – 59% were recorded in different herds. At the end of the epizootic, a total area of 3200 km² had been affected. The outbreak lasted from September 1995 to July 1996, but FMD lesions were not seen in wildlife species other than impala (OIE, 1997a).

Experimental infection of British deer took place in the 1970s, and it was demonstrated that sika (*Cervus nippon*), muntjac (*Muntiacus reevesi*), and roe deer exhibited generalised symptoms of FMD, whereas fallow deer (*Dama dama*), showed very mild or no clinical disease and red deer remained symptomless (Forman *et al.*, 1974; Gibbs *et al.*, 1975). In Kazakhstan, susceptibility of maral deer (a sub species of the red deer), roe deer, saiga antelope and wild ass to FMDV infection has been demonstrated experimentally (Bukhtiyarov, 1967b; Bukhtiyarov *et al.*, 1967).

Wild ruminants other than African buffalo are thought not be carriers of FMD virus, with the exception of the kudu which may persist in the carrier state for up to 140 days (Hedger

et al., 1972; Pinto & Hedger, 1978). Persistence of virus was found in sable antelope up to 56 days (Ferris *et al.*, 1989). There has been one report of wildebeest carrying virus for up to 45 days, (Hedger *et al.*, 1972), but other authors have found no viral excretion after 7 days (Anderson *et al.*, 1975; Falconer & Child, 1975; Pinto & Hedger, 1978). Bushbuck, eland, hartebeest, impala, springbok, tsessebe, wart hog and bush pig have been reported not to carry virus for more than 7 days (Anderson *et al.*, 1975; Falconer & Child, 1975; Hedger *et al.*, 1972; Paling *et al.*, 1988; Pinto & Hedger, 1978), although there is one report of eland carrying FMDV for up to 32 days (Thomson, 1996).

Studies on British deer have shown that muntjac and roe deer may carry FMDV for 10 – 14 days (Forman *et al.*, 1974; Gibbs *et al.*, 1975), sika and red deer may carry virus up to 28 days (Gibbs *et al.*, 1975), whereas fallow deer may carry up to 63 days (Forman *et al.*, 1974). White-tailed deer in the USA have in one study been shown to carry FMDV for up to 11 weeks (McVicar *et al.*, 1974), but in another they were reported not to carry FMD (Copland *et al.*, 1993). Kazakh scientists have showed that saiga antelope may carry FMDV for up to 10 days (Nagumanov, 1972).

3.2.10 FMD in saiga

Outbreaks of foot-and-mouth disease were recorded among saigas in Kazakhstan in 1955, 1956, 1957, 1958, 1967, 1969, 1971, 1972 and 1974 (Kindyakov *et al.*, 1959; Fadeev & Sludskii, 1982; Kindyakov *et al.*, 1972; Starchikov, 1971; Bekenov *et al.*, 1998). Epidemics of FMD among saigas have resulted in the death of 3 – 10% of the population (Sokolov & Zhirnov, 1998). In Kalmykia in the spring of 1957, 40,000 saigas (9 – 10% of the population) died during one epidemic. Mortality was high amongst calves (66.7% in newborn, 78.8% in calves aged up to one month), but also amongst females in the late stages of pregnancy (Bannikov *et al.*, 1961; Sokolov & Zhirnov, 1998). An estimated 50,000 calves died in the spring of 1967, when the FMD outbreak in central Kazakhstan spread over an area of over 100,000 km² (Fadeev & Sludskii, 1982). Mortality of adult saigas was observed, but to a much lesser degree. Adults were visibly weakened, and in general remained in one area instead of migrating. The horns of males infected with FMDV become stunted; this is recognisable for the rest of their lives. In 1969 the epidemic arrived later in the summer, and the mortality rate, especially amongst calves, was much lower than two years previously (Fadeev & Sludskii, 1982). This may have been because the calves were older and therefore suffered less severe forms of the disease. In cattle it is

well known that FMDV affects the cardiac muscle of newborn animals causing death (Mohiyuddeen, 1958). This is less common in older calves (K. O'Brien, pers. comm.).

FMDV transmission between wild buffalo and cattle sharing watering holes has been reported in Zimbabwe (Dawe *et al.*, 1994), thus the brief contact that saigas have with domestic livestock may be sufficient to allow FMDV transmission. There is much anecdotal evidence citing saigas as vectors of FMDV in Kazakhstan. FMD has been reported on farms following migration of saigas nearby, and when saigas had apparently shared pasture and watering places with domestic livestock (Starchikov, 1971). In June 1967, FMD type A-22 ASSSR 550/65 was isolated from saigas and also from clinically diseased domestic livestock in 4 *oblasts* (administrative regions), all in saiga range areas (Kindyakov *et al.*, 1973). Again in October 1969, identical FMDV was isolated from affected saigas and from outbreaks in domestic livestock in saiga ranges (Nagumanov *et al.*, 1975). In April 1967, an outbreak of FMD in livestock on a farm was traced to an infected saiga carcass brought onto the premises by hunters (Nikitenko, 1968). Many farms were affected in this outbreak, and at the time of the outbreaks mass migration of sick saigas was reported in the same areas (Kindyakov *et al.*, 1970). In 1968, saigas in Kzyl-Orda *oblast* were moved from one *raion* to another; some of these were infected with FMDV and caused outbreaks of disease in Kzyl-Orda (Starchikov, 1971). These data indicate that FMDV has in the past circulated between saigas and domestic livestock.

From the late 1950s to the early 1980s, Soviet scientists believed that saigas not only acquired infection from domestic livestock, but were carriers of FMDV themselves (Kindyakov, 1967; Nagumanov *et al.*, 1975; Starchikov, 1971; Kruglikov *et al.*, 1985). After the outbreak of FMD among saigas in South Kazakhstan (formerly Chimkent) *oblast* in 1970, the Veterinary Committee wanted to prevent domestic livestock grazing near saigas, however this was not implemented (Starchikov, 1971). Vaccination of domestic livestock in the saiga range area was considered the most effective way of controlling the disease (Kindyakov *et al.*, 1972). For example; in 1970 mass vaccination of livestock against FMDV type O-194 was implemented in South Kazakhstan *oblast* to prevent infection of saigas. The same year, livestock in West Kazakhstan (formerly Ural) and Atyrau *oblasts* were vaccinated against type A-22 to prevent infection of saigas (Kindyakov *et al.*, 1972). Following the introduction in 1974 of an improved vaccine used in the routine vaccination of domestic livestock (discussed in section 4.5.1) there have

been no further outbreaks of FMD in saigas. However, scientists fear that the recent collapse of the livestock vaccination program (discussed in chapter 4) may allow FMD once again to spread to the saiga population.

Several Soviet scientists suggested that saigas are highly susceptible to FMDV infection (type A and O) under both experimental and natural conditions (Bukhtiyarov, 1967a; Bukhtiyarov *et al.*, 1967; Kindyakov, 1967; Nagumanov *et al.*, 1975; Starchikov, 1971). FMDV has been shown to increase in virulence when passaged through saigas (Kindyakov, 1967; Bukhtiyarov *et al.*, 1967).

Nagumanov (1972) showed that after intra-lingual inoculation of FMDV saigas experienced a latent period of 6 hours before viral excretion occurred for an average of 83 hours (range 42 – 234), as shown in Table A2.2 (Appendix 2). Following experimental infection of saigas by contact with infectious material, the mean time at which FMDV was first detected in saigas was 36 hours (range 24 – 48, SD 12.83), in domestic livestock it was 45.2 hours (range 6 – 96, SD 32.46, Table A2.3, Appendix 2). The mean duration of viral excretion amongst saigas was 183 hours (range 144 – 240, SD 28.5), amongst domestic livestock it was 180 hours (range 248 – 312, SD 28.5) (Nagumanov, 1972). Saigas become infected with FMDV faster than domestic livestock, although the difference is not statistically significant. Starchikov (1971) also reported that saigas become infected with FMDV faster than domestic livestock. Saigas experienced clinical symptoms for longer (13 – 17 days), and of a much more severe nature than the domestic livestock, which showed clinical symptoms for only 6-9 days (Bukhtiyarov, 1967b). 75% of saigas (N=8) died after they were infected with FMDV, whereas during the same experiment there was zero mortality amongst the 20 domestic livestock infected. The ages of the experimental animals were not recorded. However, too much confidence cannot be placed upon the mortality rates observed in this experiment, as saigas are notoriously difficult to keep in captivity, and this may have biased the results. In a recent experiment (Ivanov *et al.*, 1999) over 70% of the captured saigas (N=27) died before the experiment actually started.

Saigas develop antibodies to FMDV earlier, and at a higher titre than domestic livestock, and the response lasts longer (Bukhtiyarov, 1967a). Following experimental infection, the duration of antibody response of saigas was >200 days (the duration of the experiment), in

domestic livestock the mean duration was 109 days (range 60 – 200, SD 58.33, Table A2.4, Appendix 2) (Bukhtiyarov, 1967b). However, as in domestic livestock, immunity to one serotype (e.g. type A) did not confer immunity to a different serotype (e.g. type O). From observations in the field during the epidemic of 1969, it seems that the saiga can retain immunity for at least two years. Clinical disease was mainly seen in animals born in 1968/69, leading to the conclusion that older animals were protected by immunity acquired in the 1967 outbreak (Fadeev & Sludskii, 1982). Although saigas were probably originally infected from domestic livestock, the saiga population appeared to be endemically infected during the period 1955 – 1974.

3.3 Brucellosis

Brucellosis is one of the most important zoonoses (diseases transmitted from animals to humans) that affect human welfare and livestock health (Nicoletti, 1990). It is considered by the World Health Organisation (WHO), the Food and Agriculture Organisation of the United Nations (FAO) and the OIE as the most widespread zoonosis in the world (WHO, 1997), and is classified as a list B disease by the OIE. Despite advances made in diagnosis and therapy, the prevalence of brucellosis in many developing countries is increasing (WHO, 1997; Abuharfeil & Abo-Shehada, 1998). As an indication of the importance of this disease, plans to eliminate ovine, caprine and bovine brucellosis from the European Union were expected to receive over half of the total European Commission funding for animal disease control measures in 1997 (WHO, 1997).

3.3.1 The bacteria

Brucellosis is caused by a bacterium, *Brucella*, which was first identified by Bruce in 1887 (Corbel, 1997). *Brucella* organisms are intracellular pathogens, and can survive within phagocytes of the host (Nicoletti, 1990). This provides protection from host defence mechanisms; an important feature in incubation period and latency. *Brucella* organisms are fairly resistant to environmental conditions, and can survive pickling, smoking, chilling and freezing. *B. abortus* may persist in an aborted foetus for 75 days during cool weather (Carter & Chengappa, 1991), and in liquid manure for up to 8 months (Abdou, 1986). *B. melitensis* may survive in soft cheeses for at least 100 days (Abdou, 1986), however it is easily destroyed by heat treatment such as pasteurisation (Carter & Chengappa, 1991).

Six species of *Brucella* are presently known, of which *Brucella melitensis* (main hosts are sheep and goats), *Brucella suis*, *Brucella abortus* (main host is cattle), and *Brucella canis* have public health implications. When transmitted to humans, these bacteria may cause human brucellosis. *Brucella ovis* (found in sheep and goats) and *Brucella neotomae* (found in desert wood rats) have not been reported to cause disease in humans. Camel, porcine and canine brucellosis, caused by *Brucella abortus*, *Brucella suis* and *Brucella canis* respectively, are not discussed in this chapter as they are not thought to be important in the epidemiology of brucellosis in saigas in Kazakhstan.

3.3.2 Diagnostic Techniques - Antibody assays

The Rose Bengal Plate Test (RBPT) is recommended by the FAO for screening samples for the determination of herd and flock prevalence (FAO, 1993). It is easily performed in the field, as very little laboratory equipment is necessary. The ELISA (Greiser-Wilke *et al.*, 1991) is the test currently used as a screening test for brucellosis at the Veterinary Laboratory Agency (VLA), Surrey, UK. The complement fixation test (CFT) (Corbel & Macmillan, 1996) is used for confirmation of positives.

Compared to the RBPT and CFT, the ELISA has been demonstrated as the most sensitive and most specific test in several studies of sheep and cattle (Saravi *et al.*, 1995; Abuharfeil & Abo-Shehada, 1998). The RBPT has 97 – 99% specificity and 98% sensitivity (A.A. Macmillan, pers. comm.), whereas the specificity of the ELISA is 99.7% and the sensitivity 99% (Saravi *et al.*, 1995). Both the ELISA and RBPT rarely give false negatives, however occasional false positives may occur (Nicoletti, 1990; Abuharfeil & Abo-Shehada, 1998), therefore one single test is not sufficient to confirm brucellosis; a combination of two tests should be performed (Mahajan & Kulshreshtha, 1991), preferably RBPT and ELISA (Abuharfeil & Abo-Shehada, 1998).

Comparisons of various serological tests on sera of red deer have yielded inconsistent results. However, Thorne *et al.* (1978) found that by using a combination of two serological tests; RBPT and CFT, they obtained 100% positive agreement on sera from wild North American red deer found to be *Brucella* culture positive at necropsy. They concluded that serologic test procedures to detect brucellosis in red deer are the same as for cattle. Morton & Thorne (1981) investigated the frequency of agreement between pairs of serological tests and found 87% correlation between the RBPT and the CFT. They also

found that 98% of CFT and 80% of RBPT correctly identified animals that were known to carry brucellosis (they were culture positive at necropsy). The ELISA test currently used at the VLA has been tested in a variety of wildlife species with good results (A.A. Macmillan, pers. comm.). It has not been used in saigas, however, there is no reason to believe that it should be any less sensitive or specific in this species.

3.3.3 World wide distribution

Bovine brucellosis, caused mainly by *B. abortus*, is the most widespread form of brucellosis (Figure 3.3). In 1998, cases of bovine brucellosis were widespread in South and Central America, Africa, the Middle East, central and southern Europe, Asia and parts of the USA and the Commonwealth of Independent States (CIS) (discussed in more detail in chapter 4) (T. Chillaud, pers. comm.). Ovine and caprine brucellosis have a more limited geographic distribution, but remain a major problem in the Mediterranean region, the Middle East, western Asia, parts of Africa, Latin America and the CIS (Corbel, 1997, Figure 3.4). The distribution of human brucellosis generally follows that of livestock brucellosis. Figure 3.5 shows the distribution of cases of human brucellosis reported to the WHO in 1998.

In Mediterranean and Middle Eastern countries the annual incidence of human brucellosis varies from less than 1 up to 78 cases per 100,000 population; however, over 550 cases per 100,000 have been reported from confined endemic areas in regions where no animal control measures are applied (WHO, 1997). Up to 77 cases per 100,000 people have been reported from certain communities in Italy and Spain, countries in which animal control measures are mandatory (WHO, 1997).

Map 3_2

Figure 3.3 bovine bru

Fig 3.4

Figure
brucellosis in 1998. Source: WHO

3.5 World-wide distribution of human

3.3.4 Pathogenesis

Brucella organisms are highly infectious and usually gain entrance to the body as a result of ingestion of food or water contaminated by the foetus, placenta or vaginal discharges of an infected animal (Carter & Chengappa, 1991). They may also enter through broken skin or through the genital tract during mating (cattle and pigs only). Sexually immature and non-pregnant females are quite resistant to infection (A.A.P. Macmillan, pers. comm.). The organism passes from the point of entry via the lymphatics to the regional lymph nodes, then into the bloodstream. The predilection sites in ungulates are the endometrium, foetal placenta and mammary gland (Nicoletti, 1990).

3.3.5 Bovine brucellosis

In cattle infection is almost always caused by *B. abortus* (Carter & Chengappa, 1991). The characteristic sign of brucellosis in cows is abortion after the fifth month of gestation, in bulls the main clinical symptom is orchitis (Nicoletti, 1990). The incubation period is variable, ranging from 3 weeks to 8 months (Morgan, 1996). About one third of infected cows abort (Carter & Chengappa, 1991), however less than 20% abort more than once (Nicoletti, 1990). The most important factor contributing to the length of the incubation period seems to be the stage of gestation; the incubation period is inversely related to it (Morgan, 1996).

At the time of abortion or parturition, approximately 15% of infected cattle may be seronegative (Nicoletti, 1990). Calves that acquire infection *in utero* or when sexually immature may remain infected for life, but they are usually seronegative until they abort or have an infected calving (Nicoletti, 1990). It can therefore be difficult to identify carrier animals. Nearly all infected cows develop permanent udder localisation with shedding of bacteria in the milk, and cows may remain infected for several years (Carter & Chengappa, 1991). There is no correlation between levels of antibodies and acquired immunity, and the immunity acquired from natural infection is not always sufficient to prevent reactivation of infection or re-infection (Carter & Chengappa, 1991).

Bovine brucellosis caused by *B. melitensis* has recently become an important problem in Israel, Kuwait, Saudi Arabia, Spain and Italy (Corbel, 1997), especially in intensive dairy farms (WHO, 1997). In the 1980s, it was found that only 0.2% of cattle in Kazakhstan were likely to be infected with *B. melitensis* (Amiraeiev *et al.*, 1986). However, during the last five years bovine brucellosis caused by *B. melitensis* has become a serious problem (M.M. Rementsova, pers. comm.). *B. abortus* vaccines commonly used in cattle do not effectively protect against *B. melitensis* infection, while the use of the *B. melitensis* Rev-1 vaccine has not been fully evaluated in cattle (Corbel, 1997). Thus bovine *B. melitensis* infection is emerging as an increasingly serious public health problem.

3.3.6 Ovine and caprine brucellosis

B. melitensis is the main cause of brucellosis in both sheep and goats. The disease occurs largely in sexually mature females, and the main sign is abortion in the latter part of pregnancy. Males may develop orchitis. The syndrome resembles that in cattle, but excretion of *B. melitensis* from the vagina is more prolonged than in cows infected with *B. abortus*, and may last up to three months. Infection of the udder is common, but shedding of the bacteria in milk may be intermittent. Interference with fertility appears to be minimal, and infected births may occur without abortion.

According to specialists on brucellosis in Kazakhstan, sheep and goats do not become infected with *B. abortus* (N.P. Ivanov, pers. comm.). Nevertheless, in Europe and North America there is evidence that *B. abortus* may transmit to sheep and goats, (Carter & Chengappa, 1991). However, it rarely infects sheep and goats, and even when it does, abortion is not a feature and the disease is not contagious (A.A.P. Macmillan, pers. comm.). Ovine brucellosis caused by *B. ovis* is primarily a genital disease of rams; clinical disease is uncommon in ewes (Corbel, 1990).

3.3.7 Human brucellosis

Transmission of brucellosis to humans is through consumption of contaminated or untreated meat and dairy products and by direct contact with infected animals, animal carcasses and abortion materials. Occasionally infection may occur by inhalation of infected dried materials (Abdou, 1986). The occurrence of the disease in humans is thus dependent on the animal reservoir, and can always be traced to an animal source (Madkour & Gargani, 1990). In humans the illness may be mild and self-limiting or severe and recurrent (WHO, 1997). *B. melitensis* is the most frequent cause of human brucellosis, and

the disease is usually severe (Corbel, 1997; Madkour & Gargani, 1990; Nicoletti, 1992). The symptoms of brucellosis include undulating fevers, rheumatic and neuralgic pains (Carter & Chengappa, 1991). The epidemiological significance of *B. abortus* for humans is quite different from that of *B. melitensis* (Abdou, 1986). In foci of bovine brucellosis the background infection rate in the human population is high, but the great majority of human infections are subclinical (Abdou, 1986). The highest prevalence in man has been reported in countries with high rates of ovine and caprine brucellosis, such as the Middle East and Mediterranean countries (Abdou, 1986).

Reported incidence in endemic disease areas varies from <0.01 to >200 per 100,000 population per annum (Corbel, 1997). Although human brucellosis is a notifiable disease in many countries, official figures do not fully reflect the number of people infected each year, and the true incidence has been estimated to be between 10 and 25 times higher than the reported figures indicate (WHO, 1997). In the USA it was estimated that the actual incidence was 26 times higher than reported (Wise, 1980). In a survey in a randomly selected human population in Saudi Arabia, seroprevalence to *Brucella* was found to be close to 20%, with more than 2% of these having active disease (Alballa, 1995). Similar figures may be expected from most countries in which the disease is endemic in the animal population. Brucellosis can be prevented in humans by controlling, or better, eliminating the disease in the animal population and avoiding consumption of raw meat, milk and milk products (WHO, 1997).

3.3.8 Vaccination and control

Vaccination of humans has not played a major role in control of brucellosis, due to the lack of development of an effective vaccine without serious side-effects (Nicoletti, 1992). Control of human brucellosis is therefore achieved through its control in livestock. There are three strategies of control measures recommended by the FAO/OIE/WHO joint committee (Abdou, 1986):

1. *Test and slaughter*. This is the most successful method, and was used successfully in Great Britain and Sweden.
2. *Vaccination program combined with test and slaughter*. This is used in countries where brucellosis is endemic and large-scale animal husbandry is common (e.g. Kazakhstan in the 1980s).

3. *Test and vaccination.* This is used in countries where slaughter is difficult due to financial implications (currently the situation in Kazakhstan).

It is commonly accepted that eradication should only be attempted where the individual animal prevalence is 2% or less, otherwise vaccination is recommended (Abdou, 1986). As the eradication procedure proceeds, vaccination needs to be stopped when the prevalence drops to 0.2% or less (Abdou, 1986). In Sweden an eradication program was thus achieved in 14 years (1944 – 1957) (Madkour & Gargani, 1990).

The FAO and OIE recommend the living vaccine strains *B. abortus* strain 19 for use in cattle and *B. melitensis* strain Rev-1 for use in sheep and goats (FAO, 1993). Most vaccinated female calves lose their serum antibody titres within 16 to 18 months, but their relative immunity to infection with virulent organisms is considered life-long (Abdou, 1986; A. Macmillan, pers. comm.). Ewe lambs inoculated with Rev-1 are protected for 2 – 3 years, whereas female goats are protected for at least 4 – 5 years (Abdou, 1986).

Programs for the control of bovine brucellosis have had a marked effect on the incidence of infection in man. This has been demonstrated in many countries where successful control and eradication programs have been in force, such as the USA, Cyprus, Denmark, Japan, Great Britain (Abdou, 1986). The effectiveness of widespread vaccination of sheep and goats in reducing human brucellosis has been shown in the Mongolian People's Republic, where, following vaccination of 33 million sheep, the incidence rate of human brucellosis declined from 4.8 per 100,000 to 0.23 per 100,000 in 7 years (Kolar, 1987).

3.3.9 *Wildlife reservoirs*

Brucellosis is maintained in wildlife populations throughout the world. In North America and Europe, *B. abortus* has been isolated from American and European bison (*Bison bison*, *Bison bonasus*), big horn sheep, chamois (*Rupicapra rupicapra*), moose (*Alces alces*), fallow deer, mule deer (*Odocoileus hemionus*), roe deer, white tailed deer, sika deer, reindeer (*R. t. groenlandicus*, *R. t. arcticus*), pronghorn (*Antilocapra americana*), red deer (*C. e. canadensis*, *C. e. nelsoni*), fox (*Chifris argentina*) and red fox (*Vulpes vulpes*). *B. suis* has been isolated from arctic fox (*Alopex lagopus*), grizzly bear (*Ursus arctos*), hare

(*Lepus europaeus*, *Lepus timidus*) and grey wolf (*Canis lupus*) (Moore & Schnurrenberger, 1981; Rausch, 1972; Thorne *et al.*, 1978; Aguirre & Starkey, 1994; Fowler, 1985; Kita & Anusz, 1991; McCorquodale & DiGiacomo, 1985; OIE, 1996a).

Serological surveys of African wildlife have demonstrated antibodies to *Brucella* spp. in African buffalo, bushbuck, eland, impala, Grant's gazelle (*Gazella granti*), oryx, Thomson's gazelle (*Gazella thomsoni*), topi, waterbuck, wildebeest, giraffe (*Giraffe camelopardalis*), hippopotamus (*Hippopotamus amphibius*), zebra (*Equus burchelli*), Jackal (*Canis mesomelas*), spotted hyena (*Crocuta crocuta*) and wild hunting dog (*Lycan pictus*) (Hamblin *et al.*, 1990; McCorquodale & DiGiacomo, 1985; Moore & Schnurrenberger, 1981; OIE, 1997a; Paling *et al.*, 1988; Sachs *et al.*, 1968; Thimm & Wundt, 1990; Waghela & Karstad, 1986).

In Russia, brucellosis has been confirmed in maral deer (a subspecies of the red deer) and in wild and domesticated reindeer (McDiarmid, 1975; OIE, 1997a; Tretyak, 1973). 40% of reindeer in Siberia are positive to brucellosis; 2% have clinical signs related to *B. suis* infection, while *B. abortus* is more common in forested areas where reindeer have contact with cattle (OIE, 1996a). In Kazakhstan, brucellosis is found in wildlife inhabiting all geographical zones; prevalence is not dependent on the landscape type or altitude (Postricheva, 1966). Serological surveys of maral deer, mountain sheep (*Ovis ammon*), mountain goats (*Capra sibirica*), roe deer and saiga antelopes (discussed in more detail in section 3.3.10) have demonstrated antibodies to brucellosis (Postricheva, 1966; Rementsova, 1987). *B. suis* has been recovered from maral deer, and *B. melitensis* from saiga antelope (Postricheva, 1966; Rementsova *et al.*, 1969).

The epidemiological role of wild animals in brucellosis has remained unexplained to date, except for the specifically established role of hares in the epidemiology of brucellosis among swine in Denmark (Christiansen & Thomsen, 1956). In North America, *B. abortus* is endemic in many populations of free-living bison and red deer, whereas reindeer and semi-domesticated reindeer are known reservoirs of *B. suis* (Fowler, 1985). According to Fowler (1985) it is more likely that wildlife have contracted *B. suis* infection from domestic reindeer than the reverse. However, there are no confirmed reports that wild ungulates are a source of infection for domestic animals under natural conditions, and

there have been no published reports of wild ungulates hindering eradication of bovine brucellosis (Hudson *et al.*, 1980; Fowler, 1985; McCorquodale & DiGiacomo, 1985).

It has been confirmed experimentally that American bison can transmit *B. abortus* to cattle (Davis *et al.*, 1990), however the risk of disease transmission from wild bison to cattle is poorly understood. No cases have been documented (Meyer & Meagher, 1995) although four outbreaks of brucellosis in Wyoming have been attributed to red deer and/or bison, because a cattle source could not be identified (US Department of Agriculture, 1997). Wild animals could be involved in mechanical transmission by scattering of contaminated material on farms, pasture areas or near water points (Moore & Schnurrenberger, 1981).

3.3.10 Brucellosis in the saiga

Serological studies of saigas in Kazakhstan have demonstrated antibodies to brucellosis (Postricheva, 1966; Rementsova *et al.*, 1969; Rementsova, 1987). Postricheva (1966) and Rementsova (1987) reported 1.7% and 1.3% seropositivity in saigas respectively. Rementsova *et al.* (1969) tested serum from 575 saigas culled in three separate locations in Betpak-dala, and found 2.9% were seropositive. Out of 118 samples inoculated into guinea pigs, 3 yielded cultures of *B. melitensis*. These had all been taken from female saigas.

Rementsova *et al.* (1969) demonstrated that infection of saigas with brucellosis can be produced through relatively small infective doses of *B. melitensis* in comparison to the doses necessary to infect sheep. Antibody production was recorded two weeks after infection, and titres were still measurable after 120 days (the duration of observation). On autopsy, *B. melitensis* was isolated from cultures taken from lymph nodes, however these cultures grew poorly in comparison with those isolated from sheep infected with identical doses of *B. melitensis* in a parallel study. Saigas retained the causative agent for a longer period than sheep, but it did not become more virulent on passaging through the saigas. Thus the results of the study revealed that the saiga is more sensitive to infection with *B. melitensis* than sheep, but non-pregnant saigas excrete the agent in insignificant numbers in the environment. This was demonstrated by the fact that when infected saigas were housed with sheep, contact infection did not occur in the latter, nor did a healthy saiga housed with the two infected saigas contract infection (Rementsova *et al.*, 1969). Non-pregnant females are quite resistant to infection and it is possible this was the reason the healthy saiga and sheep did not acquire infection through contact. In a parallel study

saigas did become infected through contact with sheep, but the intensity of infection was higher in this instance as one of the sheep had aborted (Rementsova *et al.*, 1969).

The saiga is susceptible to brucellosis, however, it is unknown what the effects of brucellosis are on the saiga population itself. Saigas may become infected in pastures used by farm animals and consequently retain and carry the infection for long distances (Rementsova, 1987). If they are carriers of brucellosis they may play a role in disseminating the bacteria during their migrations, however it is clear that under natural conditions with comparatively low concentrations of saigas in their range, the wider dissemination of infection in the environment is unlikely. However, the saiga birth areas may serve as potential reservoirs of infection because of the high concentration of birthing females (Lundervold *et al.*, 1997; Rementsova *et al.*, 1969). The experimentally observed sensitivity of the saiga to brucellosis explains why retention of *Brucellae* is seen among saigas under natural conditions (Rementsova, 1987).

3.4 Other diseases

3.4.1 Rinderpest and Peste-des-Petites-Ruminants

Rinderpest and peste-des-petites-ruminants (PPR) are both viral diseases that cause high mortality in cattle and sheep / goats respectively, and are on list A of the OIE. Neither of these diseases are thought to be present in Kazakhstan or the other central Asian Republics (Kyrgyzstan, Uzbekistan, Turkmenistan, Tajikistan), however they have in the last decade been reported in neighbouring countries Russia and China, and in Mongolia, only 50 km away from the border to Kazakhstan (OIE, 1999e; OIE, 1987; Rweyemamu, 1996). Russia reported rinderpest as recently as in 1998 (OIE, 1998e). Livestock in the eastern and south-eastern parts of Kazakhstan are unlikely to have much contact with livestock across the border as there are large mountain ranges hindering travel. Several countries bordering the southern part of Central Asia have also experienced outbreaks of rinderpest during the last decade, e.g. Pakistan, which experienced rinderpest in 1994 and Afghanistan, which saw an outbreak in 1995 (Rweyemamu, 1996). Turkey, a country heavily involved in trade with Kazakhstan, reported rinderpest in 1991, 1994 and 1996 and PPR as recently as in 1999 (OIE, 1999e). It is possible that either rinderpest or PPR could be present in remote areas of Kazakhstan.

Rinderpest has been diagnosed in many game species but predominantly in buffalo, kudu, and eland (J. Anderson, pers. comm.). It caused massive mortality in wild ungulate species in the 19th Century, when it was introduced by cattle into the Cape region, and spread right through south and east Africa (Plowright, 1982). PPR has been diagnosed on serological evidence in camels, Arabian oryx and several Middle Eastern antelope species (J. Anderson, pers. comm.). Considering the susceptibility of African wildlife to rinderpest, it is likely that the saiga may also be susceptible to rinderpest and / or PPR.

3.4.2 Bluetongue

Bluetongue is an arthropod-borne viral disease of domestic and wild ruminants, particularly sheep. It has been described as this century's most economically devastating affliction of sheep (Alexander *et al.*, 1994) and is classified as a list A disease by the OIE. Bluetongue virus is the type-species of the genus *Orbivirus* in the family of Reoviridae. Twenty-four serotypes exist (Verwoerd & Erasmus, 1994). It is transmitted by midges of the *Culicoides* spp. Evidence of infection by orbiviruses transmitted by *Culicoides* midges has been found world-wide north and south of the equator, from 50°N to 30°S in America and 40°N to 35°S in Europe, the Middle East, Africa, Asia and Australia (Sellers, 1991). In Africa and the Middle East *Culicoides imicola* is the main vector of bluetongue, and *C. obsoletus* and *C. pulicaris* are suspected vectors (Bouayoune *et al.*, 1998; Papadopoulos, 1991). The distribution of bluetongue closely follows the spatial and temporal distribution of its *Culicoides* vectors (Figure 3.6) Gawkes, 1995).

Another disease transmitted by *Culicoides* spp. is African Horse Sickness (AHS). Until the late 1980s, AHS virus seemed unable to survive beyond its traditional endemic zones in sub-Saharan Africa for more than 2 years, however the outbreaks of AHS in Spain, Portugal and Morocco which persisted for 5 years (1987 – 1991) seemed to establish a new pattern in virus survival in an epidemic zone (Mellor & Boorman, 1995). It is thought to be due to the all-year-round persistence in the area of the adult *C. imicola*, the major vector. Its continuous presence in parts of Iberia may be due to some recent moderation in the climate, and further northerly extensions in the range of *C. imicola* in response to 'climatic moderation' may occur, substantially increasing the area of Europe 'at risk' to all diseases transmitted by this vector (bluetongue, AHS and EHD). In 1999 bluetongue was reported in Greece (OIE, 1999f), Turkey (OIE, 1999g), and Bulgaria (OIE, 1999h). The OIE has

little information on bluetongue prevalence from the FSU and it is very difficult to obtain any information.

Bluetongue has not been officially reported in Kazakhstan, which lies at a latitude of about 54°N to 42°N, however there are several *Culicoides* spp. present in Kazakhstan, e.g. *Culicoides dewalphi*, *Culicoides montanus*, *Culicoides obsoletus*, *Culicoides scotius*, *Culicoides ukumensis*, which could potentially act as vectors for Orbiviruses (G. Auezova, pers. comm.). Bluetongue has been reported in Russia (Sergeev *et al.*, 1980; Vishnyakov *et al.*, 1994) and in the Xinjiang and Inner Mongolia provinces of China, which border Kazakhstan (Hawkes, 1995; Regen *et al.*, 1995).

Figure 3.6 World-wide distribution of bluetongue (Hawkes, 1995)

In most animals, infection with bluetongue virus only rarely results in disease (Daniels *et al.*, 1995). The vast majority of cattle exhibit no signs of disease, however viraemia is typically prolonged, thus cattle act as important reservoir hosts (MacLachan *et al.*, 1991). In sheep, many infections are unapparent; in particular, local breeds seem more resistant to disease than the imported European breeds (Daniels *et al.*, 1995). Clinical signs are extremely variable (Verwoerd & Erasmus, 1994); they include hyperaemia of nasal and buccal mucosae, which in serious cases may progress to oedema of the tongue with necrotising mouth lesions, accompanied by rumen stasis and secondary pneumonia (Verwoerd & Erasmus, 1994). If clinical signs are mild it is easy to assume the animal is suffering from pneumonia. Mortality rates between 2% and 45.6% have been reported

(Parsonson, 1991). A good review of the pathogenesis, clinical signs and pathology of bluetongue is given by Verwoerd & Erasmus (1994).

The susceptibility of wild ruminants to bluetongue virus has been established by experimental infection of blesbok (*Damaliscus albifrons*) (Nietz, 1933), and evidence of unapparent infections has subsequently been found in many other species (Verwoerd & Erasmus, 1994). Serological studies in North America have revealed antibodies to bluetongue virus in mule deer, red deer, pronghorn antelope and bison (Vestweber *et al.*, 1991; Chomel *et al.*, 1994; Aguirre *et al.*, 1995; Dunbar *et al.*, 1999). In Mexico antibodies to bluetongue virus have been reported in mouflon (*Ovis musimon*) (Aguirre *et al.*, 1992).

In studies of wild African game, antibody to bluetongue virus has been reported as widespread in species such as eland, nyala (*Tragelaphus angasi*), bushbuck, waterbuck, roan antelope (*Hippotragus equinus*), hartebeest, kob (*Kobus kob*), duiker spp. (*Cephalophus* spp.), buffalo, elephant (*Loxodonta africana*), black rhino (*Diceros bicornis*) and white rhino (*Ceratotherium simum*) (Formenty *et al.*, 1994; Karesh *et al.*, 1995; Anderson & Rowe, 1998). African antelopes do not seem to develop clinical disease, but other wild ruminants may develop severe disease, e.g. pronghorn, desert bighorn and white-tailed deer (Verwoerd & Erasmus, 1994). Bluetongue has not been reported in the saiga, and it is not known if saigas are susceptible to infection.

Scientists at the Institute for Animal Health (IAH) use a blocking ELISA with monoclonal antibody to detect group specific antibodies to bluetongue virus. This method was developed by Anderson (1984), and has been validated in both domestic livestock and wild ruminants. More recently, Afshar *et al.* (1995) demonstrated that a competitive ELISA using a group-specific monoclonal antibody could detect bluetongue virus-antibodies in llamas and wild ruminants. The authors proposed that the C-ELISA be used as a rapid and specific test for serodiagnosis of bluetongue virus infection in llamas and other wild ruminants.

3.4.3 Epizootic haemorrhagic disease

Epizootic haemorrhagic disease (EHD) of deer is caused by a virus of the genus *Orbivirus*, which is thus related to the bluetongue virus. It is also transmitted by the *Culicoides* spp. of insect vectors. EHD virus commonly causes anorexia and lameness, but virulent serotypes

may cause fatal infections. Epidemics of EHD in North America have resulted in high mortality in white-tailed deer and pronghorn antelope (Thevasagayam *et al.*, 1996a; Brodie *et al.*, 1998). The disease has also been reported in black-tailed deer (*Odocoileus hemionus*), mule deer, red deer, pronghorn antelope and mouflon (Shope *et al.*, 1960; Aguirre *et al.*, 1995; Aguirre *et al.*, 1992; Chomel *et al.*, 1994; Dunbar *et al.*, 1999). There have been epidemics of EHD in cattle in Africa (Thevasagayam, *et al.* 1996). EHD has not been studied as intensely as bluetongue, and it is not known if it occurs in the saiga antelope. It was therefore decided to screen the samples for antibodies to EHD virus.

3.5 Management of infectious disease in wildlife

Infectious disease may pose a serious threat to wildlife populations. There are a variety of options available for disease management in wildlife species, such as treatment, quarantine, vaccination or culling of infected individuals or populations, however, few attempts at disease control have demonstrated clear benefit (Woodroffe, 1999). The method chosen depends on the disease, the host species and the management policy.

3.5.1 Treatment

Wild animals kept in zoological parks may be treated for infectious diseases using the same methods as in domestic animals (Flach *et al.*, 1998; Samour *et al.*, 1995). Wildlife on game ranches may be treated if the manager deems it worthwhile, e.g. in Namibia cheetah cubs reared in captivity are treated with antibiotics if they show signs of bacterial infection (personal observation). In many National Parks, e.g. Kruger in South Africa, it is uncommon to treat wildlife unless the species are very rare (D. Forbes, pers. comm.).

3.5.2. Vaccination

Vaccination is used in free-living species, most notably in foxes in Europe, where oral vaccination against rabies is practised (Muller, 1996). Inactivated rabies vaccines have also been used in African wild dogs (*Lycaon pictus*) (Gascoyne *et al.*, 1993), however the results of vaccination have been variable. Much effort has been put into research, modelling different control methods for rabies in wildlife (Frerichs, 1975; Anderson *et al.*, 1981; David & Andral, 1982; Ball, 1985; Voigt *et al.*, 1985; Coyne *et al.*, 1989; Smith & Harrison, 1991; Murray, 1993). Lack of good data on rabid foxes means it is difficult to use the models for accurate predictions. The effect of vaccination of free-living bison against brucellosis has also been modelled. Peterson *et al.* (1991) evaluated annual vaccination of female bison calves. Simulations suggested that after 20 years the

proportion of the herd infected with brucellosis might be reduced from 0.69 to between 0.5 and 0.2, depending on the vaccine efficacy. If eradication of the disease is the goal, this does not seem like a worthwhile option if the vaccine has low efficacy.

3.5.3 Quarantine / movement restriction

Botswana, Namibia and South Africa all have an FMD free zone, recognised by the OIE, where vaccination is not practised (OIE, 1999i). Any animal coming from outside this zone must be quarantined and certified FMD free before it can enter. This applies also to wildlife species that are being moved, for example to a new game reserve.

3.5.4 Culling

Culling of infected animals is a very common method for dealing with disease outbreaks in wildlife. In the Yellowstone National Park bison are known to carry brucellosis, they can therefore be shot if they leave the park (Berger & Caine, 1999). In the UK, badgers (*Meles meles*) are thought to act as a wildlife reservoir for tuberculosis (caused by *Mycobacterium bovis*). Control measures against bovine tuberculosis (TB) have for years been based on culling of infected social groups of badgers (Anderson & Trehwella, 1985). In Australia, the national Brucellosis and Tuberculosis Eradication Campaign (BTEC) requires that TB be eliminated from all feral and domestic bovids. In order to achieve this, water buffaloes (feral and domestic) have been subjected to a massive capture, testing and control campaign. All reactors are culled (Freeland & Boulton, 1990). In South Africa the TB testing program requires that all African buffalo that test positive to TB are culled (Discovery, 1999).

Several governments have made provisional arrangements for which control measures to use in the face of an outbreak of exotic disease. The Ministry of Agriculture, Fisheries and Food (MAFF) used a model by Smith & Harrison (1991) to evaluate the effects of different control regimes on the potential transmission of rabies in foxes in Britain. Based on this model, MAFF decided on a policy of culling all foxes within a 19 km radius of the rabies outbreak, using poison baits. In Australia, much research has been carried out into the potential spread of FMD if an outbreak was to occur, and the role wildlife may play (Pech & Hone, 1988; Hone & Pech, 1990; Egglestone & Korn, 1993; Garner, 1993; Garner & Lack, 1995). Hone & Pech (1990) estimated that with the current opportunistic surveillance scheme in Australia, it would take 23 to 358 days to detect an outbreak of

FMD among feral pigs. Pech and Hone (1988) found that unfeasibly high culling rates of feral pigs would be required for rapid disease eradication in the event of an outbreak of FMD.

3.5.5 Management of wildlife diseases in the Soviet Union

Diseases of wildlife were researched in the Soviet Union, however there seems to have been no real attempt at management of wildlife diseases, except for vaccination of livestock, which was seen as a way of indirectly controlling transmission of diseases between wildlife and domestic populations. During some of the major outbreaks of FMD in saigas, farmers would try to keep saigas away from their livestock (Starchikov, 1971). However, this was for the protection of the livestock, not the benefit of the saiga. Experiments were conducted on saigas to identify their susceptibility to particular infections (Nagumanov *et al.*, 1975), however scientists seem not to have attempted to identify how transmission occurred in the field. There was no attempt to identify whether, for example, FMDV could circulate continuously within the saiga population, or whether infection from an outside source, such as from domestic livestock, was necessary. It is possible that control measures for diseases in wildlife populations were not considered because the basic epidemiology of the infections in the field situation had not been researched.

Chapter 4

Livestock Production and Animal Health in Kazakhstan

4.1 Introduction

This chapter is meant to provide a background to the livestock production systems in Kazakhstan. It is essential to have an insight into the workings of the agricultural industry

during the time of the Soviet Union to understand what is happening in Kazakhstan today. Very little information specific to Kazakhstan is available in the English language. Most of the information in this chapter was collected in Kazakhstan by the author, much of it during the expeditions undertaken as part of the study (see Chapter 5, in particular section 5.10), and it had to be translated by the author from Russian to English.

4.2 Kazakhstan

4.2.1 Geography

Kazakhstan is located in Central Asia (Figure 4.1), it became an independent nation-state for the first time on 16th December 1991. There were 19 *oblasts* (administrative regions) until 1997 (see Figure 4.2), when several were amalgamated. Currently there are 14 *oblasts*, which are all divided into smaller *raions* (districts).

4.2.2 Economy before the Gorbachev era (pre 1987)

Until the 19th century Kazakhstan's economy was based on livestock rearing and minor trade (Matley, 1989). In the 1920s the Russians began a process of industrialisation in the north of the country (de Broek & Kostial, 1998), and in the post-war era, Kazakhstan's economy became integrated with the Soviet military industrial complex with the establishment of further heavy industries. These industries supplied uranium, grain and metals to factories in Russia (CIA World Factbook, 1998). The 'Virgin Lands Campaign' (discussed in section 4.3.1) gave a second boost to the country with the transformation of vast arid lands into agricultural areas and turning the region into a major grain producer. Kazakhstan was the largest grain exporter to other parts of the Soviet Union, and also exported about 300,000 tons of meat every year (de Broek & Kostial, 1998).

4.2.3 The transition from centralised planning to a market economy (1987 to 1997)

According to Soviet sources, industrial output in Kazakhstan grew by 17% between 1985 and 1989, and agricultural output was growing at an average of 15.4% a year between

Figure 4.1 and 4.2

Fig chap4_1

1986 and 1989 (Anonymous, 1990). Overall output then decreased by 40% in the period 1991 – 1995 (Goskomstat, 1996). Broek and Kostial (1998) suggest that the output decline can be explained as a correction to the inefficiently high production levels at the onset of the transition to a market economy. The first reforms towards a market system began with Gorbachev's reforms in the areas of production and trade between 1987 and 1989. The

dissolution of the USSR and the collapse of demand for Kazakhstan's traditional heavy industry products resulted in a sharp contraction of the economy (Gaynor, 1996).

4.2.4 The present situation (1998 to 1999)

Currently, Kazakhstan's economy is based on its mineral wealth, fossil fuels and on agriculture, which remains one of the key activities in the Kazakh economy, providing close to half its economic output (Factbook, 1999), it was, however, the sector most affected by the transition process. 18% of the labour force work in agriculture and forestry, 24% in industry (Kulekeev, 1998). The economic situation for the majority of the population in Kazakhstan seems to have consistently worsened since independence.

4.3 Historical Changes in Livestock Production

4.3.1 Collectivisation and the Virgin Lands Campaign (1929-1960)

Traditionally, Kazakhs have been nomadic livestock producers of horses and sheep. Prior to Russian settlement and colonisation in the 19th century, Kazakhs moved between ecological zones to seasonally available grazing areas (Matley, 1989). Kazakh nomads of the steppes and deserts travelled north in the summer and south in the winter, over distances up to 1,200 km (van Lewen *et al.*, 1994). Forced collectivisation of the nomads began in the years after 1929; during the following years, livestock were destroyed or died from starvation so that there were few livestock left in the rangelands (Asanov & Alimaev, 1990). By the 1940s, official recognition that sedentarisation of the nomads was a disastrous failure led to a repopulation of the arid ranges (Kerven *et al.*, 1996). During the Virgin Lands Campaign (1954-1960) 17 million hectares of land in northern Kazakhstan were ploughed up to plant wheat (Foster, 1997). Large state farms (*sovkhos*) were constructed, and existing collective farms (*kolkhoz*) were expanded and many converted to state farms (Rementsova, 1987). Water supply systems and basic infrastructures, as well as social amenities, were constructed in the rangelands during the 1960s (Foster, 1997).

Thus each farm came to resemble a village, each with its own administration. Although collectivisation initially put an end to nomadism, a system of maximising pasture use did evolve, and each farm was given areas of summer and winter pastures with recommendations for ideal stock numbers based on estimates for carrying capacity. A partially-sedentarised state-organised form of nomadic animal husbandry thus developed in the desert (Asanov & Alimaev, 1990). The main difference between the collectivised

grazing system of the latter Soviet era and the tribal grazing pattern that prevailed until the late 19th century was in the shorter distance the livestock migrated seasonally and the higher degree of settlement during the Soviet era. During the late 1950s and 1960s these farms were successful, being supplied with all their inputs from the state. Livestock numbers increased (Figures 4.3 – 4.4) and new breeds were introduced to improve production (Foster, 1997). There was a small number of privately owned livestock on all farms – the state allowed individuals to keep some livestock for personal use. These were not kept with the collective herds unless the owner was a shepherd.

Dependence on cultivated fodder increased progressively from the mid-1960s as reliance on natural pastures declined (Gilmanov, 1995). According to Coulter (1996) the rapid expansion of livestock production in the 1960s resulted in a number of problems in the early 1980s, including overgrazing of pastures, poor disease control and poor nutrition due to the lack of protein supplement and forage. Rementsova (1987) observed that when state farms were formed by amalgamation of collective farms, veterinary prophylactic measures were often overlooked. Squires (1996) states that agriculture has had a significant impact on the environment, resulting in serious problems such as soil erosion, salinisation and pollution of waterways, with 26% of the whole country eroded (700,000 km²). Gioldyieva & Viesiliova (1992) believe 60% of the area of Kazakhstan is affected by land degradation in some way, and that animal production is the chief cause of this.

4.3.2 Farming pre-independence (1980-1991)

Collective farms were owned by their members, but had to deliver an assigned amount of produce to the state every year. State farms were state enterprises in which each worker was paid a wage by the state, with bonuses if the state quota was exceeded. At their peak in the 1980s, state farms contained an average of 540 workers and technicians, vets, accountants etc. with their families, and each had a school, a hospital, social facilities etc. (Coulter, 1996). Collective farms and state farms were similar with regard to organisation, management and marketing arrangements, however, collective farms tended to be much more diversified and of smaller hectareage than the state farms (Kerven *et al.*, 1996). In 1991 there were 430 collective farms in Kazakhstan, averaging around 38,000 hectares of which on average 9,800 were cultivated. There were 2120 state farms, averaging 80,000 hectares each, of which only on average 14,000 hectares were cultivated. However, farm

size varied from 5,000 to 300,000 hectares, depending on the type of land used (Coulter, 1996). The majority of these farms had some sort of specialisation, e.g. grain production, sheep rearing. All farms were split into smaller units that were run separately, each with their own production targets to reach.

Figures 4.3 – 4.4

Fig chap4_2

In Betpak-dala (a desert area in central Kazakhstan where one of the populations of saiga antelope are found), most farms were state farms. Farms in the south of Betpak-dala sent their shepherds 400 km north during the summer so the livestock could make use of the better pastures found there (A. Namet, pers.comm.). In the winter they grazed on pastures nearer the state farm, supplemented by feed grown on the farm or provided by the state. Migration was primarily conducted on horseback, along special migration corridors that were separated from the land belonging to farms passed on the way. In some places the separation was only 50 metres (Kindyakov *et al.*, 1972) thus migration may have allowed some contact between migrating animals and livestock on the farms bordering the migration corridor. This must have increased the risk of infectious diseases spreading through the country, even though contact was minimised. Migration benefited from considerable technical support; tractors were used to help transport the shepherds' families, *yurts* (large tents), food and petrol for the water pumps. The shepherds were sent supplies of food and petrol once a month and a veterinary surgeon visited to check the livestock. By way of this semi-nomadic system, farms managed to sustain high numbers of livestock. However, they did rely on winter-feed supplementation and technical support for the summer migrations.

Livestock on farms in the north of Betpak-dala did not migrate, but spent the summer months dispersed throughout the territory of the farm. Shepherds moved 2-3 times every season, and there was some annual rotation to prevent over grazing of pastures (Kerven *et al.*, 1996). In winter some livestock were based at the farm, but most were found at *zimovkas* (houses built by the state farm away from the main farm, where shepherds spent the winter). Sheep and cows were kept in outhouses at the *zimovka*, and relied entirely on supplemented feed all winter. Horses grazed permanently outdoors.

4.3.3 Post-independence (1991 onwards)

The transition from a command economy to a market economy began soon after independence, as described in section 4.2.3. The transfer of ownership of state and collective farms to private entities began in earnest in 1993, though some degree of private animal ownership had been permitted throughout the Soviet period (van Leeuwen *et al.*, 1994). Figures 4.5 a–c show the change in types of agricultural organisations from 1990 to 1995. The basic mode of transfer involved establishment of ownership agreements, making former workers and managers into stockholders who assumed both assets and liabilities of

these “firms”. In the initial phase, the large state-owned agricultural enterprises disposed of their yields through a state procurement system. These state-run

factories (e.g. abattoirs with processing plants) also underwent privatisation, and many were bankrupted such that they were unable to pay for the produce when it was delivered. Even when they did pay, the price was often fixed by a compulsory contract at a level way below realistic market prices (Foster, 1997).

After the state-run factories were privatised, sales of yields were opened to private buyers. Later, the sources of agricultural inputs (fuel, spare parts, fertiliser, veterinary medicines and vaccines) were also privatised (Pope, 1997). Most of these inputs were in the past supplied by Russia or other regions of the Former Soviet Union (FSU). When Kazakhstan departed from the FSU it had no infrastructure to start its own production, and input prices, now fully covered by privatised farms, began to rise (Foster, 1997). The devaluation of the *tenge* in 1994 resulted in serious consequences for the large agricultural enterprises that were now paying for their own inputs. Being cash poor, due to a variety of reforms and the

Fig chap4_3

Figures 4.5 a–c

see Figure 4.6

devaluation, most had to buy inputs based on credit from the state-owned suppliers or barter away their livestock in exchange for inputs (Pope, 1997).

According to World Bank estimates in 1993, the prices of inputs used in agriculture increased by 18.8 times while output prices increased by only 7.8 times. This was not reversed in subsequent years and was the key factor in the sector's growing financial losses. There were 37% loss-making farms in 1992, whereas 80% of farms reported losses in 1995 (de Broek & Kostial, 1998).

Collapse of markets for many livestock products contributed to a massive reduction in production (Figure 4.6), with stockpiles of unsold wool and hides being formed (Easterley & Fischer, 1996). During privatisation, livestock numbers in Kazakhstan decreased dramatically; for example the number of small ruminants (sheep and goats) dropped by over 60% in just 3 years, from 34,2 million in 1993 to 13,7 million in 1996 (Goskomstat, 1997). The number of cattle dropped 54% in 4 years, and the number of horses dropped 39% in the same period (Figures 4.7 – 4.9).

There are four main types of private farming entities; partnerships, joint stock companies, producers' co-operatives and peasant farms (Kulekeev, 1998). All these received their land from the government free of charge, and there is no land tax for them to pay if they are carrying out agricultural production within the limits of established quotas (Article 8,2 of Republic of Kazakhstan civil code 1995, Anonymous, 1995). In general, the farms that were privatised as enterprises retained most of the features of the old state farms. The peasant farms were the only private farms not classified as legal entities, and are thus not liable for the registration, taxation and minimum charter capital applicable to other types of legal entities (Gaynor, 1996). Whichever farm structure was chosen, all individuals had the legal right to redeem their land share certificates for a share of farm assets (such as livestock or access to machinery) and demarcated land plots (Robinson & Milner-Gulland, 1998). If the farm had land used for summer grazing away from its main territory (e.g. farms in southern Betpak-dala), individuals would be given two land plots, one in each area. However, it has been noted that those individuals who left the farm first received the worst land (Gaynor, 1996). The small scale of operation of peasant farmers, coupled with the lack of knowledge about the market economy, has led to difficulties for many of them

(Squires, 1996). However, it seems clear that individual peasant farmers and family plots are playing an increasing role in livestock production (Figures 4.10a – b).

Fig chap4_4

(Figures 4.7 – 4.8).

Figure 4.9

4.3.4 Privatisation

To individuals on farms, the privatisation process meant every worker was entitled to his or her own land share which could be used independently, or be given up to a privatised collective. Prior to privatisation, the state committee for privatisation assessed the value of the farm property, taking into account debt, inflation and depreciation, and established a standard property share for each member of the farm (Gaynor, 1996). Coefficients were used to calculate the size of the property and land shares per household. The size of coefficients could be decided using one of several different methods; e.g. in accordance with the role the person had on the farm (e.g. a farm worker would receive a higher coefficient than a housewife), or based on the length of time a person had worked on the farm. The *raion* authorities decided which method was to be used, then the farm management was supposed to conduct an informal consultative process to try and establish a consensus on the structure of the new type of farming entity (Robinson & Milner-Gulland, 1998).

With regard to migration, article 80 of the civil code stipulates that each *oblast* will grant land to be permanently set aside as migration corridors for livestock. Users of these tracks will be obliged to observe ecological requirements and to only drive animals during time periods agreed with the veterinary regulatory bodies. Article 50 stipulates that *oblast* and *raion* administrations can create temporary stock driving tracks for seasonal use, but these must be in agreement with the private owners on whose land the track is to lie. In practice, however, migration of livestock has almost stopped because it is uneconomical (personal observation).

4.3.5 The decline in livestock numbers post-independence

As soon as privatisation of farms began, the government dramatically cut down financial and technical support to the farms then later withdrew it altogether (Gaynor, 1996, personal observation). In many cases this happened before the workers became aware of their rights to land shares and property shares, or before they had a chance to claim them.

As a result many farms functioned as if they were still state farms, however, they had no state input and with the collapse of the livestock markets they were forced to barter their livestock in return for coal, diesel, sugar etc. Many coal companies that could not afford to pay their workers bartered coal for livestock, and the workers were given meat in lieu of wages (personal observation). Barter sales accounted for a third of agricultural sales in 1995 (de Broek & Kostial, 1998). In general, workers on the farms did not receive enough sheep to keep a sustainable flock (Robnson & Milner-Gulland, 1998), and numbers

gradually diminished as sheep were slaughtered to feed their owners' families (note that in soviet times goats were only kept by private individuals). Thus the overall number of livestock in Kazakhstan rapidly diminished, and the ratio of collective to private livestock changed dramatically (Figures 4.10a – b).

Fig chap4_5

Fig 4.10a-b

4.3.6 *Misreporting*

During the time of the Soviet Union there was a ceiling placed on the number of animals an individual could own (Gaynor, 1996). This encouraged under-reporting of the number of privately owned animals on farms. At the same time, the administration of the farms were under great pressure to perform well, and would therefore over-report the numbers of collective livestock on the farm. Over-reporting of production was common all over the Soviet Union, and has been verified by numerous sources (Easterley & Fischer, 1996). It is important to note the Soviet mentality of bribing authorities, a way of life that seems to have become ingrained in the population.

Although the state no longer dictates the number of livestock an individual can keep, it does levy a tax per head of livestock, thus people may still lie about the number of stock they own. Officially, livestock must be registered at the *Selsoviet* (village council) to have a veterinary certificate issued. The animal is given a brief examination by a veterinary surgeon, and the owner is issued with identity documents. If it is from another area, it may be vaccinated according to local conditions. The entire procedure costs about 250 *tenge* for cattle (currently about USD 1.8, or equivalent to 10 loaves of bread from the market). Without this certificate, the animal cannot (in theory) be sold. Officials in the Almaty Veterinary Committee believe that nearly all animals are registered by their owners spontaneously because of the fear of diseases. They give as proof that in rural areas everyone knows each other and it would be difficult for a farmer to lie about the number of animals he keeps. *Raion* statistical offices are responsible for conducting random unannounced checks on private stock numbers (staff at *Goskomstat*, pers. comm. 1997), but whether these actually occur is another matter.

Gross underreporting of private stock has been observed in Kyrgyzstan (ULG Consultants Ltd, 1994), and according to Kerven *et al.* (1996), the state can no longer afford to pay the salaries of those who used to reside on farms and regularly obtain data on production.

However, our study found that most of the farms that have been privatised as large enterprises retained many of the features of the old state farms, including the system of recording livestock statistics on herd size, births, deaths etc. (personal observation). Pope (1997) noted that coverage of statistics on former state farms and co-operatives by *Goskomstat* (Agency of the Republic of Kazakhstan on Statistics) remains good, and may contain some degree of accuracy, but those from the private sector are almost certainly unreliable. However, even information acquired from *Goskomstat* often contains columns of figures that do not add up to the total stated, and other figures seem to “change” on a frequent basis. As an example the Statistical Bulletin of 1998 states that there were 18,507,000 sheep and goats in 1995 (Kulekeev, 1998), whereas the Statistical Bulletin of 1997 states that there were 19,853,900 sheep and goats in 1995 (Goskomstat, 1997); a difference of nearly 1.35 million. During the last few years the state sector and large enterprises accounted for only 30% of total production, with the smaller peasant farms, household plots and *dachas* (small plots of land for growing vegetables, given to all families under the Soviet system) accounting for 70% (Pope, 1997). Hence the majority of livestock production may not be covered by official statistics at all, and the official statistics quoted in this thesis (e.g. Figures 4.3 – 4.9 and 4.12 – 4.16) may be dubious.

4.4 Animal Health in Kazakhstan

4.4.1 The Veterinary Services pre-independence

During the Soviet period the veterinary structure was centralised and under the control of the Ministry of Agriculture in Moscow. All veterinary surgeons were employed by the state. The central government in Moscow delegated responsibility for less important animal diseases to local institutes in the republics, whereas policies for combating the more important diseases were decided centrally. In Kazakhstan, prevention policies for foot-and-mouth disease were co-ordinated from Moscow, whereas the Veterinary Committee in Almaty (the capital of the republic) decided brucellosis policy. There were veterinary committees in every *raion*; these were responsible for enforcing disease prevention policies and routine disease surveillance programs, such as vaccination and serological testing of livestock. All farms employed veterinary surgeons, there was at least one on each separate unit of the farm. In addition animal technicians were employed as assistants to the veterinary surgeons. Antibiotics were widely used to treat conditions such as respiratory infections and mastitis. Specific information about animal diseases at the time of the Soviet Union is difficult to come by, because it was classified as top secret in

case of biological warfare. Scientists and officials are still unwilling to officially release information on diseases.

4.4.2 The Veterinary Services post-independence

At the time of writing the competent authority is the National Veterinary Committee (NVC), which was created in 1995. Although the NVC is within the Ministry of Agriculture, it is self-controlled. The state veterinary services on the regional and local level are composed of regional and municipal veterinary committees and veterinary stations, whose staff receive salaries from the government. There are about 7,000 veterinary surgeons employed in the state veterinary services, in addition there are others employed by farms or working privately (Morin, 1998a).

A private veterinary service working for the state and farming enterprises has gradually developed since independence, as the veterinary organisation has had to adapt itself to the new needs of private farmers. The authorities aim to reach a situation in which 80% of animals are treated by the private service within a few years (Morin, 1998a). There are also plans for privatisation of veterinary stations. Privatisation of veterinary services has developed mainly in urban areas, in contrast to rural areas where most livestock owners cannot afford to pay for this service (personal observation). In general, all large enterprises (not peasant farms) have a veterinary surgeon or animal technician on the premises, but this is far fewer than during the Soviet period. *Raion* veterinary stations have received some funding to employ veterinary surgeons on large co-operative farms, but this varies from *raion* to *raion*. Many vets do not receive income from the state, and provide their services in exchange for food or other items from the farm (personal observation).

The government in the newly independent Republic of Kazakhstan kept disease prevention policies similar to those used during the Soviet period, even retaining the old Soviet veterinary legislation. This legislation is very limited in comparison with that enforced in the European Union, and has not been updated sufficiently to cope with modern day trade in animal products. Movements of animals and animal products require certification from the veterinary services, but there seem to be no rules concerning imports from specific countries. This may be because of the new international borders that have been formed since the collapse of the USSR. Morin (1998a) reported that epidemiology and disease surveillance are in need of considerable improvement. There is no list of notifiable

diseases, only lists of infectious diseases for which control and eradication is carried out by state veterinary services, and a list of infectious diseases authorising isolation of animals and confiscation of products. If a cow aborts, the owner is required by law to notify a state veterinary surgeon, however, there seems to be no requirement to notify major diseases unless the animal is in transit. Inspectors involved in the 1998 European Commission's Audit of the Veterinary Services of Kazakhstan recommended that Kazakhstan should not be placed on the list of countries authorised to export meat and fishery products to the EU (Morin, 1998a). It is interesting to note that neighbouring country Kyrgyzstan was audited the same year, and the report recommended that Kyrgyzstan be approved for export to the EU, conditional on several improvements. Export would be restricted by allowing only certain categories of horses from specific regions. The improvements were, in particular, the completion of the list of notifiable diseases and including *Equidae* other than horses in the monitoring programmes (Morin, 1998b). One reason for the difference in the auditors' perception of animal health in Kazakhstan and Kyrgyzstan may be the fact that the Ministry of Agriculture and Water Resources in Kyrgyzstan provided the auditors with information regarding equine diseases. In general, they seemed better organised and more able to control their borders. The Kazakh authorities did not organise for the auditors to visit The Ministry of Agriculture, and were unable to provide basic information regarding important equine diseases whilst the auditors were in Almaty. Furthermore, the auditors found evidence of export licenses issued for caviar shipped to the EU (contrary to EU law).

Vaccination programs and routine disease surveillance rapidly disintegrated after Kazakhstan's independence due to lack of funding (N.P. Ivanov, pers. comm.). Since 1992 vaccination of livestock has at best been sporadic. The government has had to cut down dramatically on spending, and the agricultural sector, including the veterinary services, has had to take its share of the burden. Plans for vaccination programs have consistently been made without regard to the availability of funding (see section 4.5.2). The yearly plans for obligatory vaccinations and prophylactic measures made by the NVC are based on statistics received from the *oblast* centres. Every three months, each *raion* provides the *oblast* veterinary committee with its statistics on animal diseases and the number of registered animals in the *raion* (Vidon, 1999). Vaccines are supposed to be distributed by the NVC to the *oblast* centres according to the plan, and from there to the *raions* according to the number of livestock found there. However, many veterinary surgeons (both state and

private) report that the quantity of vaccines delivered from the state is often insufficient. Vaccinations are performed either by private or state veterinary surgeons.

Animal health is not a priority in Kazakhstan, a fact demonstrated by a senior veterinary official who, in 1997, declared on national television that trying to eradicate FMD was not worthwhile. The government does not provide sufficient funds for veterinary salaries, vaccines, drugs etc. The bankruptcy in 1997 of the main producer of vaccines (Biokombinat, in Almaty) had a further unfortunate effect on vaccination programs because vaccines have since had to be bought from abroad at great expense. Some veterinary institutes and research centres have started small-scale production of vaccines, e.g. against brucellosis (personal observation), and these may become more common in the future. However, at the moment, the laboratories lack both the capital and the materials with which to produce large quantities of vaccines.

State veterinary laboratories are financed by the government budget. A central laboratory is located in the capital (Astana), and regional laboratories are found in each *oblast*. A new financing system was introduced in 1999, which included a plan of “anti-epizootic measures”. For the year 1999, the government allocated 105 million *tenge* (about 1,312,000 USD) for the entire republic (Morin, 1998a). A proportion of the money is set aside for laboratory diagnosis of diseases. There are 75 diseases listed by the government, of which every regional laboratory chooses 25 – 30 diseases (dependent on the local situation) that they will carry out tests for free of charge. Tests for other diseases on this list are charged on a commercial basis. In general, the veterinary laboratories in Kazakhstan have poor facilities and limited modern equipment, but they do keep extensive records, and appear to be functioning satisfactorily for the testing they are doing (Morin, 1998a).

Another recent change in legislation has been to do with state procurement of vaccinations and medicines. The government has decided that only certain types of drugs and vaccines are to be provided free of charge, these are all on an official list. Any company wishing to sell drugs to a regional veterinary committee must go through a tender overseen by the Central Veterinary Committee, which forces the regional committee to buy from the supplier with the lowest prices. This system was set up in an attempt to combat corruption. In the past, state veterinary officials commonly accepted bribes from drug companies to

buy drugs for use on state farms. These were either highly overpriced, or not at all useful (A.N. Namet, pers. comm.).

The veterinary services have had to face many problems following the agricultural reforms. The increasing number of small private farmers is making disease control on a national basis more difficult. The closure of factories producing veterinary drugs and vaccines means that the availability of vaccines is not always assured, and most drugs are very expensive. Very little information is available on how outbreaks of infectious disease are managed, but it seems to be below EU standards, for example quarantine has not been imposed on farms suffering from sheep pox, a list A disease, and there is no slaughter policy in cases of outbreaks of FMD.

It is difficult to obtain reliable information on the disease status of livestock in Kazakhstan; hence the current animal health situation with regard to major diseases is not completely clear.

4.5 Foot-and-mouth disease in Kazakhstan

4.5.1 Outbreaks of foot-and-mouth disease

Before independence

In the 1950s each republic in the USSR decided its own policy for foot-and-mouth disease (FMD) control, a widespread disease at the time. Around 1960 a central FMD institute for the whole of the USSR opened near Moscow (Research Institute for Animal Health, Vladimir), and from here came regulations on how to prevent and treat outbreaks of FMD. The institute produced monovalent vaccines, which were used all over the USSR (Zh.O. Omirzhanov, pers.comm). In the 1950s and early 1960s FMD type O-1 was dominant in Kazakhstan, especially in the northern and central *oblasts* (Figure 4.11). During the Virgin Lands Campaign there was an increase in FMD outbreaks (Figure 4.12), possibly associated with the amalgamation of livestock herds from different collective farms.

In 1966 FMD type A-22 was identified for the first time in Kazakhstan. It originated from Kyrgyzstan and quickly spread to South Kazakhstan, Dzhambul and Almaty *oblasts* (Rakhmanov *et al.*, 1991). In 1967 a mass vaccination campaign was set up against FMD type A-22 in all the areas affected, followed up by systematic vaccination of livestock to create a buffer zone in Kazakhstan. In 1972 FMD type O-194 was introduced from

Uzbekistan (where it had occurred the previous year) into South Kazakhstan *oblast*, just across the border. There was alarm over the outbreaks in Kzyl-Kum *raion* (located in the saiga range area, near Betpak-dala) due to the possibility of transmission to saiga antelopes in their winter range (Kindyakov *et al.*, 1972).

The incidence of FMD fell rapidly after mass vaccination was started in 1967, but it was not until the introduction of a much-improved vaccine in 1974 that the FMD situation stabilised (Figures 4.13 and 4.14). Following the introduction of a very good polyvalent vaccine in 1980, FMD was finally eradicated in 1985 (Ivanov *et al.*, 1995). However, FMD reappeared in 1988, again in South Kazakhstan *oblast*, with an outbreak of FMD type A-22 which spread to 5 farms. In the period 1988 to 1991 there was at least one outbreak of FMD every year, but no large-scale epidemic (Figure 4.15).

This sudden re-appearance of FMD was seen by scientists as a result of less stringent border controls and more intensive links between farms (Ivanov *et al.*, 1995). The fact that all outbreaks during this period occurred in border areas (Figure 4.16) indicates that infection may have spread across the border from neighbouring countries. For example, in 1989 there was one outbreak of FMD type O in East Kazakhstan *oblast* near the Chinese border, and another two in Kzyl-Orda and South Kazakhstan *oblasts* near the border to the Uzbek republic (Ivanov *et al.*, 1995).

Figure 4.11

Figure 4.12

Fig chap4_6

Figure 4.13

Figure 4.14

Fig chap4_7

Figure 4.15 (graph)

Figure 4.16 (map 1988 - 1991)

Fig chap4_8

Current situation

Kazakhstan was free from FMD in the period 1991 – 1995, and then in 1996 there was an outbreak of type O on two farms in South Kazakhstan *oblast* (OIE, 1998c). This was thought to have arrived from Tajikistan or Iran, via Uzbekistan (A. Baizhanov, pers. comm.). May 1998 saw outbreaks of FMD type O which spread to four *oblasts*; Almaty, Dzhambul, South Kazakhstan and Kzyl-Orda. In June 1999 there were four outbreaks, also type O, on farms in Kzyl-Orda *oblast*. The outbreaks after 1995 are likely to have been caused by the deterioration in vaccine coverage in recent years, combined with a lack of stringent border controls, in particular the borders with Kyrgyzstan and Uzbekistan.

With respect to the regional situation, FMD was reported in 8 countries of the CIS between 1996-1999; Kazakhstan, Kyrgyzstan, Uzbekistan, Turkmenistan, Tajikistan, Georgia, Azerbaijan and Armenia (Sultanov & Kutumbetov, 1998). Russia has not reported FMD since an outbreak of type O in 1995, however, it has not yet been put on the OIE list of FMD-free countries because it practises vaccination. The People's Republic of China

reported several outbreaks of FMD type O between 1997 and 1999; these were not close to the border with Kazakhstan (OIE, 1999b; OIE, 1999c). Kyrgyzstan reported FMD type O to the OIE in April 1997 and October 1998. Uzbekistan has not reported FMD to the OIE, but FMD type O has been circulating there in recent years (Zh.O. Omirzhanov, pers.comm). The FAO believes the FMD situation in CIS countries is very serious, and may be a threat to many other countries, including Europe (FAO, 1999).

According to Dr Avilov (Chief of the Main Veterinary Department, Moscow) between 1986 and 1991 there was an active buffer zone in the USSR, which is now disappearing (FAO, 1998c). His opinion is that the deterioration has been caused by indefinite borders, absence of border controls, local conflicts and the generally poor economic situation. Outbreaks of FMD in CIS countries in recent years has been due to both types A and O. Type Asia-1 is also suspected in the republics of Central Asia, since it was isolated in Iran in September 1999 (OIE, 1999j). The presence of FMD in countries bordering Kazakhstan, combined with the lack of stringent border controls and the breakdown of vaccination programs means that Kazakhstan has a high risk of continually re-importing the disease, even if it manages to eradicate it within the country.

4.5.2 FMD Control

Measures pre-independence

The mass vaccination campaign set up in 1967 against FMD type A-22 involved vaccination of all cattle, sheep and goats in the areas affected, using a monovalent vaccine produced at Vladimir (Kindyakov *et al.*, 1972). The success of this campaign encouraged systematic vaccination of ruminants in the areas shown in Figure 4.17. As an example, cattle in South Kazakhstan *oblast* were vaccinated at least 5 times a year every year during the period 1967-1970. However, it was not until the introduction of the much-improved

monovalent vaccine in 1974 that the FMD situation stabilised (Rakhmanov *et al.*, 1991). In the early 1980s trivalent vaccines (against FMD types A, O and C) were used, then in the late 1980s trivalent vaccines against type A, O, and Asia-1 were used in the same areas. The first vaccination course gave immunity lasting 7 months in cattle, with 70% effectivity against type A and 100% effectivity against type O (Ivanov *et al.*, 1990).

In the case of a suspected outbreak of FMD, the official policy in the 1980s was for the head veterinary surgeon at the farm to inform the regional veterinary officer, who would take samples of blood and vesicular epithelium from suspected cases and send these to *KazNIVI* for analysis. All suspected cases were immediately isolated, and the farm was quarantined (Rakhmanov *et al.*, 1991). If FMD was diagnosed by the FMD laboratory at *KazNIVI*, a committee from *KazNIVI* was sent to the farm to ensure isolation of infected animals, disinfection of the premises and vaccination of all in-contact animals according to the regulations set up by the Soviet government. In cases where only a small number of animals (15-20) were affected, these were slaughtered and the carcasses incinerated, otherwise infected animals were isolated until recovery. Ring vaccination of up to 50km was used, and the farm was quarantined. All animals were re-vaccinated every 6 months for 2 years after the outbreak (Zh.O. Omirzhanov, pers. comm.).

Figure 4.17

Fig chap4_9

FMD Control Measures post-independence

The Veterinary Committee continued the vaccination scheme set up by the Soviet authorities, however, they disregarded the financial aspect of the scheme. The official plan for 1994 was to vaccinate 880,000 cattle and 290,000 sheep in Kazakhstan against FMD in the areas detailed in Table 4.1 (Ivanov *et al.*, 1995). However, due to lack of funding only 60,000 doses of bivalent cultured FMD vaccine against types A and O were acquired (from Vladimir, Russian Federation). These were used to vaccinate about 12% of the cattle in South Kazakhstan *oblast*. Due to lack of vaccines these animals were not re-vaccinated.

Table 4.1 The official plan for vaccination of ruminant livestock against FMD in 1994 in Kazakhstan. Source: Ivanov *et al.* (1995)

<i>Oblast</i>	<i>Number of cattle</i>	<i>Number of small ruminants</i>
Almaty	70,000	0
Dzhambul	50,000	0
East Kazakhstan	40,000	50,000
Kzyl – Orda	50,000	0
Semipalatinsk	20,000	40,000
South Kazakhstan	400,000	100,000
Taldykurgan	250,000	100,000
Total	880,000	290,000

Following the outbreak of FMD in South Kazakhstan *oblast* in 1996, the official plan for 1997 was to vaccinate one million cattle in the same areas as in 1994 (roughly half the cattle population). Only about 200 000 animals were actually vaccinated, all in Dzhambul and South Kazakhstan (roughly 39% of the cattle population in these *oblasts*) (Zh. O. Omirzhanov, pers. comm.).

In 1998 there was preventative vaccination of 362,569 cattle, 23,481 small ruminants and 300 camels in South Kazakhstan *oblast*, and in Dzhambul *oblast* 139,125 cattle were given the initial course of vaccines with a further 118,860 re-vaccinated (Zh. O. Omirzhanov, unpublished data). According to official livestock numbers, this meant roughly 95% of cattle in these two *oblasts* were vaccinated that year. However, according to officials at the Almaty veterinary committee, the vaccines were ineffective, and did not prevent the outbreak of FMD in May 1998. Emergency supplies of FMD vaccines were bought from Vladimir in order for ring vaccination to be employed in all affected areas, and the outbreak was stopped after having spread to 4 *oblasts*.

Officially, the current plan is still to vaccinate cattle in all the areas mentioned in Table 4.1. In the autumn of 1999, the government bought 1.7 million doses of bivalent FMD vaccines (against type A and O), to vaccinate all livestock in South Kazakhstan *oblast* (A. Namet, pers. comm.). Other *oblasts* at risk, e.g. Dzhambul *oblast*, were not included in this vaccination program, and unless funding is increased dramatically, the trend of the last 5 years is likely to be followed, with only a proportion of livestock in Dzhambul and South Kazakhstan *oblasts* vaccinated on a regular basis.

The current policy of handling a suspected outbreak of FMD is based on the guidelines issued at the time of the Soviet Union. If a veterinary surgeon suspects FMD, he takes

samples of blood and vesicular epithelium from the animals affected and sends them to the *oblast* central laboratory for analysis by histopathology (electron microscopy), virus isolation (by cell culture and cytopathology) and serology. Once FMD is confirmed, generally within 6 – 7 days, the entire farm is quarantined. Sick animals are treated, and the premises are (theoretically) disinfected. Movement restriction and ring vaccination are applied as control measures (chapter 8, section 8.4.7 details a case report).

The government can, and does, compulsory vaccinate against FMDV, and meets both the costs of the vaccine and the administration by a state veterinary surgeon, at no cost to the farmer. However, the government does not have powers to compel slaughter of infected animals (i.e. there is no ‘stamping out’ policy). There is no obligatory notification of suspected outbreaks of FMD, although farmers are supposed to contact the company they sell their livestock to (if they are contracted), and the company director generally calls a veterinary surgeon to investigate. The company compensates the farmer 80 – 90 % of the value of any stock lost; this is included in all standard contracts. There is no government compensation, but most companies are covered by insurance policies, and thus recoup their losses. Their policy is similar to that applied in developing countries trying to control FMD, such as Zimbabwe, and only slightly better than that applied in countries where FMD is endemic, e.g. India and Kenya. In countries free of FMD (e.g. USA and most EU countries), and other countries where eradication programs are enforced (e.g. Colombia, Russia, South Africa), there is a strict policy of slaughtering all animals on the premises where there is a confirmed case of FMD, coupled with indefinite quarantine and movement control policies. Ring vaccination may be used in widespread outbreaks, more commonly in countries not free of FMDV.

4.5.3 Epidemiological research by scientists in Kazakhstan

Epidemiological research was limited in Kazakhstan prior to 1991, however, attempts were made to trace the sources of FMD outbreaks in order to prevent further epidemics. Omirzhanov *et al.* (1976) stated that results from their epidemiological studies indicated that infection was coming from Russia and the Altai corner (eastern Kazakhstan, bordering Russia and China, and only 50 km from the Mongolian border) and especially from neighbouring Kyrgyzstan and Uzbekistan. They concluded that most FMD outbreaks occurred in spring or autumn when susceptible animals were concentrated in large

numbers as they migrated to the summer pastures and back. It was very difficult to implement anti-FMD measures on the summer pastures, because these were usually remote with no facilities for effectively isolating infected animals or herds. In general veterinary surgeons only visited once a month. Once an outbreak occurred, FMDV could rapidly spread to all susceptible herds on the pasture.

The study by Omirzhanov *et al.* (1976) concentrated mainly on the areas of agricultural-economic cooperation between Kazakhstan, Kyrgyzstan and Uzbekistan. At the time, the main road in the south from Almaty to Dzhambul ran via Bishkek (the capital of Kyrgyzstan, formerly Frunze), then on through Chimkent to Tashkent (the capital of Uzbekistan). The researchers were especially concerned about the movement and transport of livestock between Kyrgyzstan and Kazakhstan. For example, livestock from Kyrgyzstan were slaughtered at abattoirs in Kazakhstan, and there was close contact between the human populations of Dzhambul *oblast* and its neighbouring *oblast* in Kyrgyzstan with respect to sale of livestock and livestock products at markets. Analysis of data from outbreaks in Dzhambul *oblast* showed that most infected livestock were from *raions* bordering Kyrgyzstan; 59% of the *oblast* foci of FMD infection were found in these areas. This was thought to be related to the movement through these areas of livestock from Kyrgyzstan migrating to and from their summer pastures (Omirzhanov *et al.*, 1976).

4.6 Brucellosis

4.6.1 Historical Background

Brucellosis was rife all over the Soviet Union prior to 1952, when a mass vaccination campaign of livestock was launched. By 1959 the average overall prevalence of ovine brucellosis in the Soviet Union had been reduced by 50% (Rementsova, 1987) and the incidence (number of new cases) per annum had decreased by an average of 20-25%. In Kazakhstan brucellosis has been endemic for over 60 years (Rementsova *et al.*, 1969). In 1954, for example, 95% of cattle farms and 100% of sheep farms in Almaty *oblast* harboured brucellosis infection (Rementsova, 1987). In the 1950s and 1960s the proportion of abortions in livestock attributed to *Brucella* infection fluctuated between 47% and 63% in cows and between 60% and 80% in ewes (Rementsova *et al.*, 1969).

During the Virgin Lands Campaign (1954 – 1960), veterinary prophylactic measures were often overlooked when livestock from different collective farms were combined into large

herds on new state farms without prior veterinary examination or vaccination. As a result there was an increase in disease spread as infected and susceptible herds were mixed. On one state farm there was an acute epidemic of ovine brucellosis just before lambing, which affected up to 70% of animals in individual flocks. Within a few months, 40% of the farm workers detailed to care for the sheep during the lambing period were afflicted with brucellosis (Rementsova, 1987).

The prevalence of brucellosis in livestock began to decrease after introduction of the Soviet vaccination campaign in the 1950s. The percent of aborted sheep placentae positive to brucellosis dramatically decreased between 1954 and 1964, from 60 - 80% in all *oblasts* in 1954, to an average of 3.2% in 1964 (Rementsova *et al.*, 1969). According to official statistics, the prevalence of brucellosis continued to fall until 1989 in cattle and until 1995 in small ruminants. This is extremely surprising, considering that the incidence of human brucellosis increased by over 200% between 1966 and 1986 (see section below), and humans are invariably infected by animals or animal products. Scientists in Kazakhstan have attempted to point out this fact now that they can speak more freely than during Soviet times (T. Grushina, pers. comm.). The official prevalence in cattle rose gradually from 1.38% in 1989 to 1.91% in 1994, then decreased again, according to official statistics, whereas the prevalence in small ruminants rose only briefly in 1996 and 1997 (to 0.96% in 1997), then decreased to 0.47% in 1998 (Figure 4.18). Several scientists believe the decrease is an artefact caused by poor serological surveillance in recent years (see section 4.6.4). The true situation in livestock is very unclear.

4.6.2 Human brucellosis

Human brucellosis is one of the main disease problems in Kazakhstan, after tuberculosis (T. Grushina, pers.comm.). The incidence of human brucellosis per 100,000 per annum in Kazakhstan was gradually falling between 1956 and 1966 (Figure 4.19), probably related to the ongoing livestock vaccination campaign. It was not possible to obtain any data for the years between 1967 and 1986, but during this period the incidence per 100,000 per annum increased over threefold, from 4.7 in 1967 to 15.3 in 1986. For the next three years human incidence gradually fell, until it was 10.8 in 1989 (Figure 4.19).

Fig 4.18

Fig 4.19a

Fig chap 4_10

Fig 4.19b

Fig chap4_11

According to official statistics, reported brucellosis cases in humans more than doubled from 1989 to 1992, and by 1992 had reached 19.8 per 100,000 per annum, nearly twice as high as in 1956, and more than four times as high as in 1965. This may have been related to the poor economic situation in the Soviet Union during *perestroika* and *glasnost*, with lack of funding for veterinary programs. After 1993 the reported incidence again dropped, decreasing by 37% to 13.2 in 1998. It is uncertain whether the fall after 1992 is real, or an artefact caused by poor disease diagnosis and reporting, now that the medical services have deteriorated dramatically and free health care is (in practice) no longer available.

According to Dr N.P. Ivanov (Brucellosis Laboratory, *KazNIVI*) 75% of the rural human population have been exposed to *Brucella* organisms. He believes 85% of exposure is to *B. melitensis*. Dr T.A. Grushina (Institute of Plague in Almaty) has evidence that 98.7% of human brucellosis in Kazakhstan is caused by *B. melitensis* (even when the disease was caused by contact with infected cattle). She believes *B. abortus* is only found in the northern *oblasts*, elsewhere in the country *B. melitensis* is responsible for clinical human brucellosis. It is possible that this is true, however, it is likely that brucellosis caused by *B. abortus* is also widespread, but in many cases not diagnosed as the symptoms are milder.

4.6.3 Control policies before independence

Before 1991, all people considered at risk (e.g. vets, shepherds, workers at state abattoirs) were vaccinated against brucellosis, despite the fact that available vaccines were not very effective. Routine vaccination and serological surveillance of livestock formed part of the brucellosis control program (N.P. Ivanov, pers. comm.). All collective cattle were vaccinated every other year with the strain 82 vaccine. There were three different

vaccination programs used for sheep and goats, depending on which part of the country (and thus at which risk) they were in. Sheep in northern *oblasts* were vaccinated with a full dose of Rev-1 every other year, whereas those in the south were either given a small dose of Rev-1 every year, with intradermal skin testing used to identify carriers, or they were given strain 19 every year. All livestock were blood sampled 4 weeks before vaccination, to test for antibodies to *Brucella* spp. Privately owned livestock were never vaccinated.

Farms were divided into two groups; ‘successful’ farms which were free of brucellosis and ‘unsuccessful’ farms that harboured brucellosis infection. On ‘unsuccessful’ farms, all collective cattle and small ruminants were bled once a month. On ‘successful’ farms collective small ruminants were bled once a year, whereas collective cattle were bled twice a year if the farm was in a southern *oblast*, and once every 3 months if it was in a northern *oblast*. The main reason for the difference in policy between the northern and southern areas is the higher prevalence of brucellosis in the north (Figure 4.20), possibly climate related as it is colder and there is less sun. If only one unit on a farm was affected, only the animals on that unit would be tested monthly, the rest of the farm would be tested as usual.

Privately owned livestock were also tested; in northern *oblasts* four times yearly, in southern *oblasts* twice yearly. All serum samples were tested using the CFT and the ‘RAYTA’ test (an agglutination test for antibodies to brucellosis). Animals that tested positive to either of the tests were slaughtered (N.P. Ivanov, pers. comm.). If a privately owned animal tested positive after a serological test, it was immediately slaughtered, and the owner given a collective animal as compensation. However, control policies were not always implemented, e.g. highly productive or pedigree animals were often not slaughtered, and sometimes not even quarantined (Rementsova, 1987).

4.6.4 Control policies after independence

After 1992 the brucellosis vaccination program collapsed due to lack of funding; vaccination is now at best sporadic (see chapter 8). Before 1996 the vaccine strain 82, used in cattle, was always bought from Moscow, whereas the sheep vaccine, Ref-1, was produced locally in Almaty (at Biokombinat). In 1997 Biokombinat closed down, and lack of funding prevented acquisition of vaccines from Moscow. The brucellosis laboratory at *KazNIVI* has now started producing its own brucellosis vaccines, but only on a small scale. The serological surveillance scheme has also suffered. According to the State Veterinary Service, all livestock are tested twice yearly, however, sources at *KazNIVI* stated privately that they did not believe this to be true, and that the official figures do not reflect the real

Figure 4.21 a

Figure 4.21 b

Fig chap4_12

situation. Senior figures at *KazNIVI* believe that the official number of brucellosis infected animals is being kept artificially low by mainly sampling animals in areas where there is known to be little brucellosis, and not sampling areas where there are known to be many positives. The reason for this is that government officials are worried about losing their job if they admit that brucellosis is a problem. The same sources also believe that the number of human cases in 1997 was around 5000 to 6000, not 2000 as was the official number registered. If this is true, it may be a form of over-reporting (i.e. over-reporting success), a continuation of the problem of misreporting so widespread in the FSU.

Even though the incidence of reported brucellosis in humans has been rising, it is still much lower than that in countries reporting similar prevalence in livestock. In Saudi Arabia, where human brucellosis is considered of major epidemiological concern (Kiel & Khan, 1989), the prevalence in livestock was determined at 2.18%, and the reported human incidence was 86 cases per 100,000 population per annum (Madkour, 1990). In Kazakhstan in 1995 there were only 14 human cases reported per 100,000, when the prevalence in livestock was 1.87% in cattle and 0.86% in small ruminants. It is surprising that the human incidence of brucellosis was 6 times less than in Saudi Arabia, when livestock prevalence was only 1.2 times less in cattle and 2.5 times less in small ruminants.

As discussed earlier (section 4.3.6), misreporting was common in the Soviet Union; it was in the interest of farmers and veterinary surgeons to report low levels of disease (over-reporting success). The centrally organised serosurveillance program for brucellosis meant that all livestock were routinely tested, which should give a degree of confidence in the government figures prior to 1991, however, it is difficult to know if the figures have been 'doctored', as there are no independent studies for comparison. The lack of independent studies must be taken into account when looking at the official statistics. With regard to FMD, although control measures for disease outbreaks were under strict government control, it is possible that some outbreaks of FMD could have gone unreported, however, as the FMD figures were top secret before 1991 they might be less likely to be 'doctored'. It is difficult to judge the reliability of the official data that is available, however the more recent data (after 1991) is likely to be less reliable than data from before 1991, due to the current lack of central involvement in veterinary care.

4.7 Other diseases of livestock in Kazakhstan

Several list B diseases are endemic in Kazakhstan, e.g. tuberculosis, rabies, hydatidosis, whilst others are sporadic, e.g. anthrax, leptospirosis, Aujeszky's disease (Morin, 1998a). Only two list A diseases, FMD and sheep pox, have been reported. The last case of rinderpest was in 1930; no other list A diseases have been reported in the country. However, the Kazakh government did not report to the OIE at all during the years 1991-1994 (T. Chillaud, pers. comm.). They reported sheep pox and goat pox in 1995 and 1996, but failed to notify the outbreaks of FMD in 1996 until October 1998 (OIE, 1998c). It is clear that the official information available is not always reliable, and there may be other list A diseases present in Kazakhstan. It was therefore decided to test all samples for rinderpest, peste-des-petites-ruminants and bluetongue, all list A diseases found in countries neighbouring, or close to Kazakhstan.

Chapter 5

Methods

5.1 Aims

The main focus of the fieldwork was to collect blood samples in order to estimate the prevalence of antibodies to FMD, brucellosis and several list A diseases in the saiga populations of Betpak-dala and Ustiurt, and to compare this with serological data collected from domestic livestock inside and outside saiga range areas. A further aim was to collect biological data on the saiga, such as male to female sex ratio, body condition of adults, size and weight of calves etc. Incisor teeth and mandibles (lower jaws) were collected from saigas in order to develop a reliable ageing method. Another aim was to collect data on the age, sex, breed, body condition, geographical location and herd management system of individual sampled livestock, in order to relate such factors to the disease status of an animal, and to enable assessment of the amount of contact between saigas and domestic livestock. Additionally, meteorological data were collected for use in a model to predict the spread of FMD virus, as well as data on past disease prevalence, control measures and monitoring schemes operating at the time of the Soviet Union.

5.2 Potential Sampling Approaches for Diseases

5.2.1 Foot-and-mouth disease

The presence of FMDV in an animal can be detected by isolation of the virus from whole blood, salivary secretions or epithelial tissue, but only in the early stages of infection (Kitching & Donaldson, 1987). Viral antigen and DNA is easier to isolate than live virus, however, these methods will only identify current or recent infection. Antibody status is a more reliable measure of risk of infection, as specific antibodies are present lifelong post-infection (R.P. Kitching, pers. comm.). A cross-sectional study can thus act as a retrospective cohort. By estimating age related prevalence of disease, it is possible to get a time series of challenge rather than just a snap shot in the short time available during an expedition. Antibodies acquired due to infection can be differentiated from those raised post-vaccination using an ELISA test recently developed at IAH (Mackay *et al.*, 1998).

5.2.2 Brucellosis

Bacteria of the *Brucellae* spp. can be isolated from whole blood or internal organs, especially from the female genital tract and placenta. These bacteria are extremely difficult to isolate unless the animal is in the early stages of infection or is pregnant (Nicoletti, 1990). The presence of antibodies specific to *Brucellae* spp., as with FMDV, indicates infection at some stage in the animal's past life, however, serological tests cannot differentiate between post-vaccinal antibodies and those acquired due to infection, or between different *Brucella* species.

5.3 Serological surveys

Serological surveys are gaining increasing importance as epidemiological tools, in particular as countries pursue eradication programmes (Donaldson, 1996). Ideally, sample size requirements should have been calculated beforehand, however, at the time of planning, very little was known about saiga disease, and available data for brucellosis and FMD were at least 30 years out of date. Hence it was impossible to choose a value for expected seroprevalence. Also, a number of diseases were being investigated, all of which were likely to have different prevalences. Hence a strategy of maximising sample size rather than optimising it to a particular disease was decided upon.

5.4 Collection Protocol

The approach used was to collect whole blood, with the intention of identifying disease specific antibodies from all animals sampled. This would allow screening for antibodies specific to rinderpest, PPR, bluetongue and EHD in addition to FMD and brucellosis. Vesicular fluid and epithelial tissue was only to be collected if an animal was exhibiting clinical signs of FMD. It was decided not to take swabs for culture from the internal organs of animals culled in autumn, because infection with brucellosis mainly occurs around calving time in spring when millions of organisms are shed during and after parturition (Nicoletti, 1990). Autumn samples were thus likely to be negative.

5.5 Timing of expeditions

The author spent a total of 20 months in Kazakhstan, over a period of 3.5 years (Table 5.1). Both expeditions to sample saiga calves were run in May. All livestock expeditions took place between spring and autumn due to the adverse weather conditions experienced in Kazakhstan. During winter many roads are closed due to heavy snowfall, it is also difficult to keep vehicles running in the extreme cold (-35 – -45°C). Permits for sampling

saigas allowed culling during specified time periods that had to coincide with the annual hunting season (October-November), with the exception of a small number of saigas sampled in spring. In 1996 the license for autumn culling did not permit culling of males over 6 months of age, and in 1997 it only allowed 7 males over 6 months of age to be culled. Licenses in the spring of 1997 and 1998 were exclusively for male saigas.

Table 5.1 Dates of field work to collect data for this study		
<i>Date</i>	<i>Region</i>	<i>Activities</i>
1st trip		
Sep – November 1996	Almaty	Learning Russian; organising the first expedition
Nov – December 1996	Central Betpak-dala	Expedition: 86 saigas sampled; pilot farm survey
2nd trip		
April 1997	Almaty	Organising the next expedition
May 1997	Central Betpak-dala	Expedition: 8 saigas, 167 saiga calves and 125 livestock sampled
June 1997	Karaganda	Organising the next expedition
June 1997	Northern Betpak-dala	Expedition: 113 livestock sampled
June – July 1997	Karaganda, Almaty	Literature search in libraries; 20 livestock sampled near Almaty; organising the next expedition, Expedition: 145 livestock sampled
August 1997	Southern Betpak-dala, South Kazakhstan <i>oblast</i>	
Sep – Oct 1997	Almaty, Karaganda	Obtaining statistics from libraries, veterinary laboratories, national statistical agency; organising the next expedition
November 1997	Central Betpak-dala	Expedition: 270 saigas sampled
December 1997	Almaty, Karaganda, Akmola	Arranging export of the samples
3rd trip		
April 1998	Almaty	Organising the next expedition
May 1998	Ustiurt	Expedition: 10 saigas, 610 saiga calves, 59 livestock sampled
June 1998	Almaty	Organising the next expedition
July 1998	Central Betpak-dala	Expedition: 496 livestock sampled
Aug 1998	Almaty	Arranging export of the samples

5.6 Sampling saigas

5.6.1 Selection of populations

The Betpak-dala saiga population is the largest and best-studied in Kazakhstan (Bekenov *et al.*, 1998), it was, therefore, chosen as the main focus of the study. Betpak-dala also contains a greater number of farms and villages and higher livestock density than the other saiga ranges (Goskomstat, 1996), possibly encouraging more contact between saigas and livestock. The Ustiurt saiga population was chosen for comparative purposes because Ustiurt contains fewer farms and is more remote from communications, hence it was assumed to have less poaching pressure. The southern part of Betpak-dala suffered from overstocking of livestock in the 1980s and early 1990s, whereas Ustiurt has always been virtually devoid of livestock (Kunaev, 1985; S. Robinson, pers. comm.).

5.6.2 Restraint of saigas for sample collection

Whole blood can be collected from live wild animals only if they are restrained. Physical restraint is extremely stressful for wild animals, especially for saigas which are naturally flighty animals. Fatal injuries during restraint are common both in the saiga and other species (personal observation; V.V. Ukrainskii, pers. comm.; Iu. A. Grachev, pers. comm.; M. Jago, pers. comm.). In an expedition connected to this project, 25 adult saigas were caught and loaded onto a truck. Upon arrival at their destination, only 100km away, 24 saigas were already dead.

In the Ural saiga population, corrals are sometimes used when hunting, but in the other two range areas the landscape is too flat to favour this method of capture (V.V. Ukrainskii, pers. comm.). The use of tranquillising drugs in the saiga is unresearched. Well recognised problems associated with chemical capture in other ungulate species are capture myopathy and prolonged sedation (Smuts & Bryden, 1973; Rossiter, 1985). The saiga antelope is hunted annually in the autumn. Collecting whole blood from animals immediately post mortem is a standard technique (Broughton *et al.*, 1970) which gives good sample quality, although it will not give a sterile sample. If the sample is rapidly frozen, bacterial growth is inhibited, and sterility thus becomes less critical (discussed in section 5.9.2)

5.6.3 Obtaining permits

The Institute for Zoology and Animal Genetics applied for its own permit to cull saiga. The Ministry of Ecology and Natural Resources of Kazakhstan issued permits for culling of saiga for scientific purposes; in November 1996 for 150 animals (no adult males), in

November 1997 for 270 animals (maximum 10 adult males). There was thus unavoidable bias against adult males. Two further permits, each to cull 10 male saigas, were issued in the spring of 1997 and the spring of 1998. The sample size in spring was thus small, but this was unavoidable.

5.6.4 Description of sampling

Saiga antelopes were shot either by professional hunters or by scientific personnel from the Institute of Zoology and Animal Genetics (of the Kazakh Academy of Sciences). During the two autumn expeditions to Betpak-dala, the team visited hunting inspectors in the *raion* centre (Dzhezkazgan) and asked for the current location of saigas. Local shepherds helped with further directions, but it still took one to two weeks to locate a group of saigas. Saigas are highly mobile, having a huge range area (45,000 km² in Betpak-dala) (Bekenov *et al.*, 1998). Once a group of saigas had been found, a base camp was set up in this area from which a vehicle was sent out to collect samples.

Animals were either shot at night using spotlights or during the day from a moving vehicle. One vehicle (Zil) with a team of 4 -5 people went out every evening at dusk, and searched for saigas using spotlights. Animals were shot with semi-automatic shotguns (Seminof). This is the method preferred by professional hunters, because a large number of animals can be shot rapidly. No more than 10 saigas were shot at any one time to prevent a time delay causing unnecessary suffering of animals that had not been killed outright. Culling during the day could be achieved if two vehicles were available. The hunters would shoot from one moving vehicle, the other vehicle was used to drive the herd. Usually fewer animals can be shot because they break up into smaller herds as they are chased. This method is also more stressful for the saigas.

Every shot animal was sampled. Collection of whole blood from the jugular vein or carotid artery took place immediately post-mortem using plain 10ml vacutainer tubes (Beckton Dickinson). Most animals were sampled during exsanguination, but in a few cases the animal was not found immediately, causing a delay of sampling and necessitating blood collection from the abdominal or thoracic cavity.

All blood samples were numbered in accordance with the numbered ear tags (Dalton Short-term Jumbotags) that were used to identify each individual animal immediately after

shooting. The time and date of shooting were noted, as were the size of the herd and the number of animals sampled. Maximum and minimum temperature and wind direction were noted daily, because FMDV can be spread by the wind. During daylight hours each animal was examined and a full post-mortem was performed either by the author or by another veterinary surgeon. Two incisor teeth were removed from all saigas for the purpose of age determination, and the entire mandible was removed in a selection of saigas of different ages for investigation into the sequence of tooth eruption. Approximate age, sex, and condition were noted, as were any gross pathological lesions and the presence of external or internal parasites. Saigas were sampled in the same manner during the spring expeditions. A total of 374 saigas were sampled (Table A4.1, Appendix 4).

5.6.5 Assigning body condition scores and age estimates

Body condition scores were assigned using the standard method developed by scientists at the Institute for Zoology and Animal Genetics (Iu.A. Grachev, pers. comm.). Assigning condition scores is subjective; the author, therefore, always did this in order to minimise between observer bias (Hilborn, & Mangel, 1997; Fowler *et al.*, 1998). Table 5.2 describes the system used. Fat around the kidneys was visually assessed, but not actually measured. Chapter 6, section 6.3.1 of this thesis describes the methods used for age estimation.

<i>Body condition score</i>	<i>Description</i>
Poor condition	Approximately <0.5cm fat around kidneys
Average condition	Approximately 0.5 –2.5cm fat around kidneys
Good condition	Approximately >2.5cm fat around kidneys

5.6.6 Constraints on sampling methods, possible bias and errors

The timing of sampling was constrained by the fact that the majority of sampling had to occur during the hunting season, which is always in the late autumn. The permits issued only allowed culling of saigas during the defined period of the hunting season.

According to the literature most disease transmission with regard to FMDV and brucellosis seems to occur in the spring, around calving (Kindyakov *et al.*, 1959; Bannikov, 1961; Postricheva, 1966; Rementsova, 1969; Starchikov, 1971; Kindyakov *et al.*, 1972; Fadeev & Sludskii, 1982; Rementsova, 1987; Sokolov & Zhirnov, 1998; Bekenov *et al.*, 1998), so this would be the ideal time to attempt isolation of live bacteria or virus. However, timing

of sampling is less relevant with respect to detection of antibodies, because these are long-lived and may be present for up to several years (R.P. Kitching, pers. comm.), regardless of when the animal was infected.

Logistical problems presented a great obstacle in the planning and execution of the expeditions, and in the return of samples to the UK. The extreme cold during the last week of the autumn expeditions (-35°C) caused the blood of saiga to coagulate very fast post-mortem. It was difficult to recover any blood at exsanguination if the animal had been dead for more than one minute. Thus towards the end of the expeditions only 1-2 saigas could be shot at any one time, slowing down the pace at which sampling could occur.

It was not possible to randomly select the place of culling, or the individual animals culled, because the saiga had to be culled where they could be found. In a herd of wild animals it is extremely difficult to randomly select an animal, and then make sure that this exact animal is culled. To achieve a reasonable sample size, animals had to be shot where and when possible. Therefore, the hunters tried to shoot as many animals as possible (up to 10 in one location), and all culled animals were sampled. It is possible that the stronger animals got away from the hunters, whereas the weaker lagged behind and were shot. This could theoretically cause a bias in favour of diseased animals, however brucellosis does not in general weaken animals, except possibly in the initial stages of infection, but this would generally be in spring, not in autumn, so brucellosis is unlikely to affect their ability to run. FMD can, in its clinical stages, cause lameness in animals, so all animals were examined carefully for lesions consistent with recent FMDV infection, none which were found. Thus for the two focal diseases the problem is minimal, however, it is possible that killing weaker animals could lead to a bias for other diseases or parasites.

When sampling we were restricted from covering the whole of the saiga range area by the cost of petrol, and by the time needed to cover such a large area. Ideally an aircraft would have been available to pinpoint the exact location of the saigas, with a further two teams using separate vehicles to ensure a higher number of samples. The window of opportunity is relatively short as saiga herds break up, to form small rutting herds, in late November / early December. Saigas were, therefore, culled mainly in one area, so there would be several entire herds in other areas that were not sampled. However, the large size of herds in October / November mean that a high percent of the population were present in one area; groups of several thousand animals are common at this time of year, although we

only observed smaller herds. However, the observation that herds are very fluid with animals commonly joining other herds, especially after rutting and calving (see below), means that any infection has good opportunity to circulate through the entire population.

Sampling several individual animals from one herd and treating them as separate datapoints, rather than treating each separate herd as one datapoint, could pose a problem of pseudo-replication (Hurlbert, 1984). This would be particularly serious in a species with a close knit herd structure, but the very fluid herd structure in saigas lessens the likelihood of bias. In particular, in the autumn, when samples were collected, the saiga are migrating and are in large groups that have formed and reformed as saigas leave the summer pastures. A study by Grachev & Bekenov (1993) demonstrated the fluidity of saiga herding behaviour using mark-recapture; they found that saigas marked in the spring in one herd were found in the winter in completely different herds, having used a number of different migration routes to reach the winter pastures. Given this herd structure, and as the study was looking at seroconversion rather than active infection, the serological status of the other members of the herd at the time of sampling can be viewed as independent of the focal individual's status. One exception would be in the event of a very recent outbreak of FMD, where the close contact between animals in one infected group would signify a greater probability of infection than that found in the rest of the (uninfected) population. However, this eventuality did not arise during our study.

In 1996 the saigas in Betpak-dala were at very low density and it was not possible to fulfil the quota given. Although saigas at this time of year are starting to form small harem herds, they have in previous years been found at much higher densities than they were either in 1996 or 1997 (V.V. Ukrainskii, pers. comm.). This may be a direct effect of constant poaching disrupting normal herd behaviour (when saigas are chased, they will split into smaller herds running in different directions), or it may be that we simply did not find the main concentration. As there was no aerial survey prior to sampling, it is theoretically possible that there was a large herd in a part of the range area that we did not cover, however, most other hunters experienced the same problem of not managing to fulfil their quotas. Few adult males were sampled, due to the licensing, and our sample may thus not be representative of the population, however, there was no prior reason to suppose that seroprevalence to FMDV would vary by gender. The percent of adult males

has decreased to around 8 – 10% in recent years due to poaching (Bekenov *et al.*, 1998), however, this is still much higher than the percentage of adult males sampled.

5.7 Sampling saiga calves

5.7.1 Reasons for sampling calves

There were two main reasons for sampling saiga calves: First, sampling could occur in the spring, which is the time of year when most disease outbreaks occur, and also when placentae can be sampled (important for isolating *Brucella* spp.). Second, that a very large number of samples could be collected in just a few days, while the calves are situated at high density in a small area. There was also no restriction on the number that could be sampled, as the calves were released unharmed. Antibody from animals less than a week of age can be assumed to be maternal antibody, i.e. the result reflects the antibody status of the breeding females in the population. Saiga antelope, like other ruminants, have a cotyledonar placenta (personal observation), and transfer of maternal antibody is through colostrum during the first few days of life. Experiments have shown that lambs born to brucellosis infected ewes are negative for antibody to brucellosis at birth, however, 15 minutes after suckling they are positive (N.P. Ivanov, unpublished data) hence sampling should reflect the prevalence of brucellosis and other infections previously experienced by the breeding females in the population.

5.7.2 Description of sampling

The method used to find young calves was to drive around the saiga range area until a herd of saigas was seen. Saigas run away when they are disturbed by a vehicle, but if there are calves too young to follow their mothers, these mothers will not run far. From their behaviour, it is obvious that there are young calves in that area.

Chemical immobilisation is not necessary to capture saiga calves, because calves less than two days old tend to lie still hiding and are usually easy to catch. If the weather is cold calves up to a few days of age can be caught, because the cold weather makes them more sluggish (Bekenov *et al.*, 1998; personal observation; Iu.A. Grachev, pers. comm.). Usually, several calves were caught at the same time, and the approximate distance (in metres) of each calf to its nearest neighbour was recorded. Physical restraint is stressful for saiga calves, but much less so than for adults. The procedure for sampling was performed quickly, with a helper restraining the calf and the author or another veterinary surgeon

collecting the samples. Each calf was weighed, and the weight noted to the nearest 50g. Its length from nose to rump was measured, to the nearest cm. The sex and approximate age was noted. Whole blood was collected aseptically from the jugular vein using a vacutainer system (Beckton Dickinson field kit for lambs), and an ear tag (Dalton Aluminium Lamb Tag) was placed in the left ear.

At the time of calving a brucellosis-infected saiga will shed millions of bacteria, most of these will be located in the placenta and placental fluids. Swabs of all newborn calves (those still wet) were taken for culture, and cotyledons were taken for culture from all fresh placentae found (7 were found). Calves were released immediately after sampling. A total of 167 calves were sampled in Betpak-dala, 610 in Ustiurt (Table A4.2, Appendix 4).

5.7.3 Age estimation of calves

Age estimation was performed by the author to prevent bias caused by the subjective age assessment of different scientists. The scale was developed by the author, in consultation with saiga experts from the Institute for Zoology and Animal Genetics. Estimations were made entirely on the appearance of the calf. Table 5.3 describes the method of age estimation. Estimations are not entirely accurate, because strong wind and sun may cause a calf's coat to dry faster than usual.

<i>Estimated age</i>	<i>Description of calf</i>
Less than 6 hours	Calf still wet
12 hours	Calf dry, but with its coat still harsh with dried placental fluid, and its navel wet
24 hours	Calf dry, with only some areas of dried placental fluid, and its navel almost dry
48 hours	Calf dry with dry navel, still slightly unsteady on its legs
72 hours	Calf dry with dry navel, not at all unsteady on its legs, only caught because it was cold

5.7.4 Constraints on sampling methods, possible bias and errors

- Timing is a major factor when sampling saiga calves, because most calves are born within a space of 5 days (Bekenov *et al.*, 1998). As they generally can only be caught when they are less than two days of age, this necessitates fast work. Once the place of

mass calving was found there was no time to search for a different herd, so all the samples in a given year are from one aggregation. However, aggregations are made up of many different herds and up to 70% of the saiga population may gather in one area, as was the case in Ustiurt, thus a large proportion of the population is represented.

- Sampling saiga calves only gives an indication of the maternal antibodies status of the saiga population, hence it excludes males, females that have aborted and those that are barren e.g. due to disease. This could lead to bias if any of the diseases have a predilection for male animals (the ones in this study do not), or if females have suffered disease in the previous year causing infertility or abortion (any of the diseases in this study could do this).
- If brucellosis is a problem in the saiga herd, it is possible that a bias could occur when newborn calves are sampled because females that have aborted (due to brucellosis) will not have a calf to be sampled, thus causing an underestimation of the real prevalence. This should be borne in mind when looking at the results. On the other hand, calves born with clinical brucellosis may be weaker, thus easier to catch than strong, healthy calves.
- If a high proportion of very young calves (that have not yet suckled) were caught, they may have no detectable antibodies because they have not yet received colostrum, thus causing possible false negatives. However, calves generally suckle within the first 30 minutes of life if the dam is not disturbed (Tsapliuk, 1982). Also, few calves estimated as under the age of 6 hours were caught, and this age-group had a higher prevalence of antibodies to brucellosis than the older calves (discussed in section 7.3.3).
- A very short time period was available for sampling, it was, therefore, deemed important to sample as many calves as possible. Thus random sampling was not attempted, instead all caught calves were sampled. Many calves were not caught because they ran away. An unknown number would have not been found because of their ability to hide and melt into the surrounding terrain. These biases are unavoidable, and impossible to quantify.

- The expedition to Betpak-dala was delayed by several days due to problems with bank transfers, thus many saigas had already calved when we arrived in Betpak-dala. We also did not find the main herd and, therefore, did not manage to sample as many animals as in Ustiurt.

5.8 Sampling domestic livestock

5.8.1 Aims

The main focus of the fieldwork was to collect blood samples from domestic livestock in order to estimate the prevalence of antibodies to *Brucellae* and FMD, bluetongue, EHD, rinderpest and PPR viruses. Farms were selected from within the range of the Betpak-dala and Ustiurt saiga populations, and outside of saiga range, for comparative purposes.

5.8.2 Obtaining permits

Sampling was restricted by the need to obtain official permits. The Central Veterinary Committee in Almaty issued a general permit for sampling of livestock and data collection from farms in Kazakhstan, however, individual permits had to be obtained for every *raion* visited. These could only be obtained in the *oblast* centre, not in Almaty.

5.8.3 Selection of study farms

Betpak-dala was the main focal area of the study, so most farms selected for the study were in this region. Seventeen villages (former *sovkhoses*) were visited along three separate transects in the Betpak-dala saiga range area (Figure 5.1). For comparative purposes, one village was visited in the Ustiurt saiga range area, and three villages located outside saiga range area. In addition, shepherds grazing livestock near the saiga calving areas were visited. These livestock came from three villages; Zhailma (Sarysu *raion*), Chu (Suzak *raion*) and Zhenis (Zhana-arkin *raion*), which were visited on other expeditions.

Virtually all the farms using land in Betpak-dala were situated along one of the three transects, so a high proportion of farms (73.9% [17/23]) was sampled. Thus the sample should be representative even though the farms were selected for their contrasts. It was considered important to sample domestic livestock in each seasonal range of the saiga.

The northern transect visited farms located in the saiga summer range, or which sent livestock into their range. Four farms were visited; Amantau, Arshelinski, Taldesay (all in Nurin *raion*) and Tkenekta, (in Tengiz *raion*). The middle transect followed the road

between Dzhezkazgan and Karaganda, on the boundary of the steppe and the semi-desert zones (the spring / summer range of the saiga). Due to financial constraints, only four farms could be visited. The villages Sarysu and Muibulak (Dzhezdin *raion*) were selected for study because of their large size. They are representative of a group of sheep-raising *sovkhozes* founded in the 1960s in semi-arid areas where grain growing is impossible. These areas had until that time only been used as transit areas for stock going to summer pastures further north. Druzhba (Zhana-arkin *raion*) was chosen as a contrast because grain can be grown there, and it was a cattle-raising *sovkhov*. Zhenis (Zhana-arkin *raion*) was chosen because it is the only farm in the *raion* to still have a collective structure.

In the south of Betpak-dala, most villages are scattered along a transect following the river Chu and the northern line of the Moinkum sands. This area encompasses the saiga winter grounds, and is on the boundary between semi-desert and desert. Livestock from this area

Fig 5.1

migrate across Betpak-dala to their northern pastures in Karaganda *oblast* in the summer. Every village along the west-to-east transect, except Tasdy, was visited, up to the settlement of Ulan Bel' in the east. Nine villages were visited; Zhailma, Kalinina, Chiganak, Kamkaly, Sarysu (Sarysu *raion*, Dzhambul *oblast*), Ulan Bel' (Moinkum *raion*, Dzhambul *oblast*), Chu, Suzak, Zhuantobe (Suzak *raion*, South Kazakhstan *oblast*). These *raions* encompass much of the Moinkum desert and southern Betpak-dala, which is the saiga winter range, and the area in which forage shortages are most likely to occur, as discussed in chapter 2.

In Ustiurt the density of farms and livestock is much lower than in Betpak-dala. Four sovkhoses were selected using a random number table. However, due to logistical problems, only one farm was sampled. Outside the saiga range area, three farms were visited; two in the southern part of South Kazakhstan *oblast*, and two in Almaty *oblast*. Two of the farms had experienced an outbreak of FMD and it was important to find out which measures had been taken to contain the disease. This could have caused bias with respect to FMD data, therefore the data were analysed with and without these farms included. The aim was to get a general idea of which control measures are implemented

when outbreaks of major diseases occur in Kazakhstan; this would be informative in predicting the course of an epidemic if an outbreak should occur in the saiga range.

During the study period, an outbreak of FMD occurred on a farm in Almaty *oblast*. The farm was visited so the author could personally observe the control measures implemented. Samples of blood and vesicular epithelium from a cow exhibiting clinical symptoms of FMD were obtained, however, it was not possible to conduct any interviews or sample any other animals without official permission. The other farm sampled in Almaty was selected because of its close proximity to Almaty, and because sampling was allowed without an official permit, allowing a single day spent sampling without having to organise a whole expedition.

5.8.4 Selection of herds / flocks

Due to the large size of the farms, many of them around 80,000 hectares, it was not possible to sample every herd on the farm. The plan of randomly selecting herds to sample was not possible in practice, because villagers and shepherds refused permission to sample their livestock when approached directly, and instead directed us to go and see the veterinary surgeon. This happened on all farms, forcing us to always contact the local veterinary surgeon or animal-technician, who on most occasions was willing to help us. An explanation of the process of random selection was given, however, not a single veterinary surgeon could understand the point of this, and all insisted that they choose the herds / flocks to be sampled. We did manage to insist that animals kept in different areas of the farm were sampled, including those collectively owned and those owned by co-operatives or peasant farmers who had left the state farm. Thus political issues on the farms meant that it was not possible to randomly choose herds for sampling, however it was recorded whether animals were kept in the village or on the *zhailo* (summer pasture).

5.8.5 Selection of individual animals

Camels were not sampled as there are very few of them in the study area, and they are not thought to be of great importance in the epidemiology of brucellosis in Kazakhstan (N.P. Ivanov, pers. comm.). Cattle, sheep and goats were sampled. Within a herd / flock containing less than 20 animals, all animals were sampled. In larger herds / flocks, 20-50 animals were caught by the owner. On no farm was there a race or extra gates / pens available to help select individual animals and enable random sampling. The owner was, therefore, asked to catch “any” animal, and the author personally supervised the capture exercise to

ensure there was no obvious systematic bias e.g. in favour of animals in good condition. The age and sex of sampled animals are depicted in Figures 5.2 and 5.3 and Tables 5.4 a-c.

Table 5.4 The number of livestock sampled by age and sex

a) in the Betpak-dala area

<i>Age in years</i>	<i>Cattle</i>		<i>Sheep</i>		<i>Goats</i>	
	<i>Male</i>	<i>Female</i>	<i>Male</i>	<i>Female</i>	<i>Male</i>	<i>Female</i>
0 - 6 months	19	24	57	41	14	17
7 - 12 mths	6	9	3	0	0	1
13 - 18 mths	11	15	20	23	9	8
19 - 24 mths	2	11	15	42	5	12
2 - 3 years	9	52	34	117	8	38
4 - 5 years	1	39	10	86	6	11
6 - 8 years	0	26	1	14	1	5
> 8 years	1	15	0	14	0	0
Total	49	190	140	337	43	92

Figures 5.2 – 5.3

b) The number of livestock sampled in Ustiurt by age and sex

<i>Age in years</i>	<i>Cattle</i>		<i>..... ..Sheep</i>		<i>Goats</i>	
	<i>Male</i>	<i>Female</i>	<i>Male</i>	<i>Female</i>	<i>Male</i>	<i>Female</i>
0 – 6 months	1	0	5	2	1	0
7 – 12 mths	0	1	0	0	0	0
13 – 18 mths	0	2	0	0	0	0
19 – 24 mths	0	2	0	3	0	0
2 – 3 years	0	0	0	9	0	1
4 – 5 years	0	1	1	28	0	0
6 – 8 years	0	2	0	0	0	0
Total	1	8	6	42	1	1

c) The number of livestock sampled outside saiga range area by age and sex

<i>Age</i>	<i>Cattle</i>		<i>..... ..Sheep</i>		<i>Goats</i>	
	<i>Male</i>	<i>Female</i>	<i>Male</i>	<i>Female</i>	<i>Male</i>	<i>Female</i>
0 – 6 months	1	2	0	0	0	0
7 – 12 mths	0	1	0	0	0	0
13 – 18 mths	0	0	0	0	0	0
19 – 24 mths	0	0	1	2	0	0

2 – 3 years	0	3	0	2	0	0
4 – 5 years	0	10	1	9	0	0
6 – 8 years	0	13	0	2	0	0
>8 years	0	1	0	0	0	0
Total	1	30	2	15	0	0

5.8.6 Description of sampling and body condition scoring

Whole blood was collected aseptically by venepuncture of the jugular vein. A vacutainer system was used (Beckton Dickinson 10ml plain sterile vacutainer blood-collecting tubes and 1 inch 16 gauge sterile needles). Samples of vesicular epithelium were collected from any animal exhibiting clinical signs of FMD. Cattle were restrained by a rope tied around their horns, which was then tied to a post. Sheep and goats were turned up, placed on their side and restrained on the ground by a farm worker. The availability of the RBPT allowed on-farm screening of the samples for brucellosis, and the owners were informed of the results. The owner or shepherd was asked the age of each animal, and this was checked for reliability by the appearance of its teeth. The time and date of sampling, type of ownership, place of birth, vaccination status and disease history were noted.

Body condition scoring in cattle was based on the method described by Webster (1993). This visual assessment is used to score cattle according to the amount of subcutaneous fat and muscle in the loin area around the lumbar vertebrae, and in the area between the pelvis and the tailhead. For sheep and goats, the method was based on the body condition scoring system advocated by the Meat and Livestock Commission (MLC, 1983). Manual assessment was used to score the animals according to the prominence of the spinous and transverse processes of the lumbar vertebrae, the amount of muscular and fatty tissue underneath the transverse processes, and the fullness of the eye muscle and its fat cover. The author modified both scoring systems from five point to three point systems, in order to minimise sampling error. There were three condition scores; ‘poor’, ‘average’ or ‘good’.

5.8.7 Constraints on sampling methods, potential bias and errors

Logistical problems, in particular the time taken to obtain official permits, presented a great obstacle in the planning and execution of the expeditions. The time of year that sampling could occur was constrained by the extreme weather conditions of the steppe, which make transport extremely unreliable during winter and early spring. The saiga expeditions had to be in the spring and autumn, leaving only the summer for the livestock

expedition. According to the literature, brucellosis is most commonly transmitted around parturition (generally late winter and early spring) (Nicoletti, 1990), so this would be the ideal time to attempt isolation of live bacteria, however this was not possible.

Although villages were not randomly selected, such a high proportion (73.9% [17/23]) of villages in Betpak-dala was selected that this is unlikely to have caused a serious bias. There may have been some bias in the selection of herds if the veterinary surgeon on the farm chose herds that he believed to be disease free, or perhaps on the contrary, selected herds he wished to be tested for brucellosis. However, the general lack of veterinary medicines, anthelmintics and vaccinations on farms meant that veterinary care was minimal for all stock, regardless of the status of the owner. All livestock kept on the farm were observed to have virtually identical management systems. They were housed in their owner's yard at night, and during the day grazed on the common pasture near the village if their owners lived in the village, otherwise they grazed on or near land owned by the co-operative / peasant farmer.

Errors could have been made by the owners when estimating the ages of their livestock, but as this was checked by inspecting their teeth any errors are likely to have been only in older animals, and are unlikely to have much of an effect on the results. Operator error, such as mislabelling tubes may have occurred. The possibility of this happening was minimised by labelling the tube before the sample was taken, and at the end of the day by checking that the numbers on the tubes were in linear order, and were identical to the identification numbers recorded on the sample sheets.

5.9 Conservation and analysis of samples

5.9.1 Conservation and transport

After collection, blood was left to clot at ambient temperature for 10-15 hours, then centrifuged (IEC MediSpin) at 2700 rpm for 10 minutes. The serum was inspected visually for signs of disease, e.g. jaundice. Many of the sera from saiga calves in Ustiurt were noted to be cloudy; a possible indication of a high triglyceride count (one of the main constituents of fats and oils). This was noted down, but not investigated further. Serum was extracted using sterile polythene 3ml Pasteur pipettes and placed in two sterile 2ml polypropylene biofreeze vials (Costar), then frozen in a freezer at a temperature of -18° C or in liquid nitrogen at -192° C. The duplication of samples was to enable them to be sent to both the IAH and the VLA, for tests to be performed on the samples. A RBPT was

performed on the serum left in the original tube, using a negative and positive control (calf serum, supplied by the VLA) with every test to ensure the antigen was intact. Swabs were immediately put into modified Amies charcoal medium, and frozen in liquid nitrogen. Cotyledons were put in plain 5ml tubes and frozen without conservants. Vesicular epithelial tissue was suspended in a glycerine and phosphate buffer with added antibiotics (Kitching & Donaldson, 1987). All samples were kept in liquid nitrogen until they were packed for transport to the UK.

During the expeditions, mandibles were clearly labelled, and kept in a cardboard box to allow them to dry out. The incisor teeth were kept in plastic test tubes, which were numbered with indelible ink. On arrival in Almaty, the mandibles and teeth were boiled for 30 minutes to destroy any infectious organisms, and all remaining fascia was removed. They were then left to dry for 24 – 48 hours, and repacked. For shipping, samples were securely packed according to the guidelines issued by the IAH (Kitching & Donaldson, 1987). On arrival at Heathrow airport the samples were placed in a freezer at -20° C to await collection by staff from VLA and IAH. On arrival at each respective laboratory the samples were noted still to be frozen.

5.9.2 Constraints on conservation and transport methods, possible bias and errors

During the first expedition in November 1996, the freezer broke on the journey to Betpakdala, and the sera had to be kept at ambient temperature (between +5° and -30° C). On the expedition in May 1997, a freezer made specifically for use in a car was taken, however, it too broke a few days into the expedition. The ambient temperature ranged from +20° to +35° C, therefore, the sera were placed in a thermos in buckets filled with cold water (+10° to +15°C) from a deep well every few hours. During the livestock expedition in August 1998, a greater than anticipated number of samples were collected, and the liquid nitrogen evaporated. All the samples thawed out, and were kept at ambient temperature (+20° to +30° C) for two days until we could buy more liquid nitrogen.

The storage problems experienced may have affected the quality of the samples, decreasing the number of positives. However, IAH has received thousands of samples from all over the world, many of which have been stored at ambient temperature in hot countries for weeks, and the general opinion is that the storage of these samples would be unlikely to cause a failure to detect antibodies using the ELISA test (J. Anderson, pers.

comm.). The results from the RBPT performed on fresh samples were compared with the ELISA results from VLA to ensure similarity of the results for brucellosis.

Labelling errors could occur when sera were transferred into the biofreeze vials. Such errors were minimised by labelling the vials before transferral. Two people were involved in transferral, and they both checked that the labelling was accurate. All transfers occurred in sequential order, making it easier to spot mistakes. No more than one farm was sampled on a single day, decreasing the risk of samples from different farms being mixed up.

To minimise errors, mandibles and teeth were labelled immediately upon removal from the animal, however, it is possible that mislabelling could have occurred. This is most likely to have occurred with the teeth, as these had to be taken out of the labelled test tubes to be boiled. The risk of replacing teeth in the wrong tubes was minimised by only boiling 10 pairs of teeth at a time, in a pan divided into 10 separate compartments.

5.9.3 Sample analysis

Analysis of sera and tissue samples for FMD

All the samples from saigas and domestic livestock were tested by the author at the World Reference Centre for Foot-and-Mouth Disease, IAH, Pirbright, Surrey. The standard test used there for detection of antibodies to FMDV is the liquid-phase blocking sandwich ELISA developed by Hamblin *et al.* (1986). The author was trained to perform this test by staff at the IAH, using the protocol described by Hamblin *et al.* (1986). The ELISA uses antigen specific to one virus type, therefore, all samples had to be tested separately for antibodies to virus types A and O. Samples were tested in duplicate, and any with borderline results were re-tested. All media used was made up by technicians at the institute. Positive samples were tested again, using the virus neutralisation (VN) test for confirmation according to the standard procedure at the IAH (Donaldson *et al.*, 1996; Golding *et al.*, 1976). Positives were also tested using a new ELISA, developed by Mackay *et al.* (1998), which differentiates between antibodies raised by vaccination and those by infection. This ELISA is not specific to virus type; it will identify non-structural antibodies raised to any FMDV type. Both these tests were performed by staff at the IAH.

Analysis for brucellosis

Samples were tested for antibodies to *Brucellae* spp. by staff in the Brucella Research Section, VLA, Surrey, using ELISA and CFT according to standard procedures described by Corbel and Macmillan (1996) and Greiser-Wilke *et al.* (1991).

Analysis for other diseases

The author tested the samples for antibodies specific to rinderpest, PPR, bluetongue, and EHD at the IAH. A monoclonal antibody based competitive ELISA was used, which is available as a field kit made up at the IAH. The tests are identical for all four viruses, with the exception of the antigen and monoclonal antibodies, which vary for each virus. The test protocol and reagents used for rinderpest and PPR is described by Anderson & McKay (1994), for bluetongue by Anderson (1984) and for EHD by Thevasagayam *et al.* (1996a).

5.10 Data collection on farm management and animal health

5.10.1 Aims

The main aim was to collect data on the age, sex, breed, condition, vaccination status, geographical location and herd management system of the individual animals that were blood-sampled, in order to relate such factors to the disease status of the animal, and to enable an assessment of the amount of contact between saigas and domestic livestock.

5.10.2 Design of questionnaires

The author designed the veterinary questionnaires, using standard social research techniques (Moser & Kalton, 1971). The questionnaires for the livestock owners were designed by the author in co-operation with a colleague, Sarah Robinson, working on a related project (Biological and Social Aspects of Land Degradation on the Steppes, and Effects on the Saiga Antelope, INTAS project 96-2056). In order to increase the efficiency of data collection, data for the two studies were collected simultaneously.

A preliminary questionnaire was tested by the author during a pilot study, which took place during the saiga expedition in November 1996. It became clear during the pilot study, that a formal questionnaire was inappropriate. It was, therefore, decided to replace this with a semi-structured interview. The initial questionnaire (Appendix 3) was used as a base to compile a list of questions with the intention of encouraging the interviewees to talk about the subject in question.

5.10.3 Administration of questionnaires

The author administered the questionnaire to key personnel on the farm: the local veterinary surgeons, shepherds, private farmers and individual livestock owners. A colleague (Sarah Robinson) conducted the interviews with the administration, and with some of the shepherds and private farmers. All interviews were conducted in Russian, without the presence of an interpreter. In order to check for factual accuracy, a farmer would be asked how many cows, calves, bulls etc. he had, then how many cattle he had in total. Inevitably, these numbers would not add up, thus this method failed to ensure reliability of the data collected. Total livestock numbers were therefore not used in the analysis, with the exception of herd / flock size, which the author could count herself.

During the first of the livestock expeditions it was realised that most data acquired from the farm accountant on collective stock (where it remained) and private stock, plus human population figures, were available for individual farms from the *raion* offices, and therefore many farm data were collected in this way. Regional veterinary laboratories were visited to obtain records of the disease status on individual farms.

5.10.4 Collection of data from raion centres

Government veterinary laboratories in *raion* centres were visited, and all data available on FMD and brucellosis, such as results from serological surveys, disease outbreaks, vaccinations, were obtained for individual farms (where available) and for the *raion* as a whole. Staff were interviewed about official vaccination programs and serological surveillance for brucellosis operating in the *raion*. Government statistical offices were also visited, and data were obtained on the human population on each farm, the type of land on the farm, the number of livestock kept and livestock mortality (where available).

5.10.5 Constraints on data collection methods, possible bias and errors

Most interviewees rapidly became bored with answering questions. This was a serious limitation on the time that could be spent effectively obtaining information. Furthermore, if the owner of the livestock was not present, his wife or children had to be interviewed; in which case these sometimes gave conflicting information. A possible source of error was the fluency of the interviewers in Russian. Both interviewers spoke conversational Russian, however, neither were fluent in the language. Errors caused by misunderstandings may, therefore, have occurred. This was minimised by a Russian member of the team

(non-English speaking) being present, who would clear up any obvious misunderstandings caused by the interviewee using unfamiliar words, by explaining things in simple Russian.

Interviewees may have misinformed or given misleading information, causing errors in the data. As discussed in chapter 4, during the time of the Soviet Union, a ceiling was placed on the number of private livestock individuals could keep. Although this system no longer exists, there is now a tax levied. As private livestock must be registered at the *Selsoviet* (village council) to have a veterinary certificate issued, there are records of the number of private livestock on each farm. However, these figures may not be accurate, and at the statistical offices it was only possible to obtain data on the total number of private livestock on each farm, not the number of livestock kept by individual owners. It was thus not possible to countercheck the accuracy of the data obtained.

On every farm enterprise visited there was an accountant who still kept incredibly detailed records of collective stock numbers by age, and recording births, deaths, the number bought, sold or bartered etc. This would imply that the figures for collective stock could be accurate, which is backed up by the fact that on farms where we checked the accountant's stock numbers, these matched exactly with those found at the *raion* centre. In the past it was generally collective livestock that were present in the pastures away from the state farm centre, thus these were more likely to contact saigas than the privately owned animals.

5.11 Data management and analysis

The data were entered into dBASE III plus Version 1.0 (Ashton Tate) and analysed using Epi Info Version 6.04b (Dean, A.D., Dean, J.A., Burton, A.H. & Dicker, R.C., 1991, Stone Mountain, Georgia, USA) animals Microsoft Excel 2000™. Maps were scanned into the computer, then edited using Microsoft Paint™.

Chapter 6

Saiga ecology

6.1 Introduction

In this chapter I outline the results of analyses of the age, weight, sex and body condition of the saiga antelopes sampled during the fieldwork. Comparisons were performed to investigate any differences between saiga populations in these ecological parameters, and possible reasons for these differences are discussed. The accurate ageing of saiga antelopes is an important issue, both for the interpretation of the results of this study, and for future ecological and epidemiological work. Thus the chapter also includes a comparison of the effectiveness of a number of ageing techniques, based on an analysis of the data collected during this study. These techniques include those currently used by local scientists for saiga antelopes and those employed for other ungulate species. On the basis of this comparative analysis, recommendations are made for future work.

6.2 Methods of age estimation

6.2.1 Age estimation of ungulates

Ungulates can be given an estimated age by a variety of methods, such as the tooth eruption pattern, tooth wear, tooth sectioning technique, incisor height, mandible length, eye lense weight, horn size, hind leg to body weight ratio (Rasmussen *et al.*, 1982; McCullough & Beier, 1986; Hrabec & Koubek, 1989; Brown & Chapman, 1991a; Brown & Chapman, 1991c; Angibault *et al.*, 1993; Moore *et al.*, 1995; Kierdorf & Becher, 1997). Some of these methods are less accurate (Moore *et al.*, 1995; Hewison *et al.*, 1999).

In domestic ungulates such as sheep, the tooth eruption pattern is well-described and easy to use. It accurately ages sheep from the age of 1 year and 3 months (when the first, most central, permanent incisors erupt) to the age of 2 years and 9 months (when the fourth, most lateral, permanent incisors erupt) (Williams, 1988). In many wild ungulate species (e.g. Javan rusa deer (*Cervus timorensis russa*) red deer, fallow deer, roe deer), eruption of all permanent teeth is complete by the age of 18 – 36 months, so ageing beyond this limit is more difficult (Brown & Chapman, 1991a; Brown & Chapman, 1991b; Bianchi *et al.*, 1997; Moore *et al.*, 1995; Ratcliffe & Mayle, 1992).

The tooth-wear method, usually based on the extent of wear on the mandibular teeth, has been used for age estimation in moose, saiga antelopes and red deer (Passmore *et al.*, 1955; Bannikov, 1961; Brown & Chapman, 1991c). However, it is not an accurate method (Hewison *et al.*, 1999). Recently, Kierdorf & Becher (1997) showed that measuring of enamel hardness and taking this into account to modify the wear scores / indices improved

the accuracy of age estimation. In both moose and reindeer, a tooth sectioning technique (TST) has given more accurate results than the tooth-wear method (Sergeant & Pimlott, 1959; Reimers & Nordby, 1968). Laws (1952) developed a method of age determination in seals by preparing thin sections of the canine teeth and examining the regular sequence of growth layers in the dentine (annuli). This method was adopted by Sergeant and Pimlott (1959) to age moose, using incisor teeth. They suggested that there is a seasonal sequence of deposition of the annuli, and considered their accuracy to be plus or minus 1 year for younger animals, and 2 years for older animals. They therefore put the animals in age classes, e.g. 5 - 6 years, 6 - 8 years, rather than attempting to accurately estimate the age.

According to Ratcliffe and Mayle (1992), precise assessment of the age of ungulates with a complete set of permanent teeth is only possible using TST. This has been applied to roe deer in which the age to the nearest month can be estimated if the date the animal was culled is known (Aitken, 1975; Ratcliffe & Mayle, 1992). However, Moore *et al.* (1995) compared different techniques for age estimation of fallow deer, and found that incisor height, molar height, molar wear and annuli in dental cementum were directly comparable techniques. Of these four methods, incisor height was the most appropriate method for the study population and accurately aged almost 90% of males.

6.2.2 Age estimation of saigas

Male saiga can be reliably aged by the size and shape of their horns up until the age of 18 months (Figure 6.1). After 18 months only a rough estimate can be given. Young horns are shorter and straighter with black tips; the older the animal gets the more lyrate the horns become and the tips become less black. (Bannikov *et al.*, 1961; Sokolov & Zhirnov, 1998). As saigas are all born in May, animals up to a year old can be reliably aged if the date of culling is known, simply because they are clearly juveniles (Bekenov *et al.*, 1998).

The sequence of tooth eruption can be used to age both males and females up to the age of 19 months. According to Bannikov *et al.* (1961) there is no difference between male and female saigas in the timing or sequence of tooth eruption. This assumption was used by the author and her colleagues. Above the age of 19 months, Bannikov *et al.* (1961) advocated the tooth wear method, using the molar teeth. Using this method he grouped animals into age categories of 3, 4, 5 – 6, 7 – 08, and 9-10 years (Figure 6.2).

Fig 6.1 and 6.2

Fig chap6_1

Sokolov & Zhirnov (1998) found it difficult to get good results using the TST, as they found the annuli were often not precisely defined. In their study, they found only 23.8% of incisor teeth showed clearly visible annuli (N = 68). A further 33.3% were good enough for analysis, 36.5% were analysed with difficulty, and in 6.3% the annuli were not possible to differentiate.

6.3 Age estimation used in this study for culled saigas

6.3.1 Age estimation by visual assessment

A local technician from the Institute of Zoology (A. Grachev) with many years' experience working with saigas made visual estimations based on his personal experience, using horn size and shape, the eruption of the last molars and the general appearance of the animal. As all saigas are born in May, animals culled in November would be 6 months, 18 months, 30 months old etc. Similarly, animals sampled in spring would all be of integer age. In saigas the last molar teeth erupt at 8 – 10 months of age, thus six-month-old saigas can be differentiated from those 18 months or over. Males were given an estimated age by the shape and size of their horns. In the spring Betpak-dala expedition, all the sampled males (6) were thought to be one year of age, and the 2 sampled females were thought to be over one year of age. In the Ustiurt expedition, 5 of the 10 sampled males were thought to be one year old, one was estimated as 2 years of age, one as 3 years of age and two were estimated as 4 years of age. Table 6.1 depicts the visually assessed age ranges of the animals sampled during the Betpak-dala autumn expeditions.

Table 6.1 Estimated age and sex of saigas sampled in Betpak-dala, using the visual assessment method. The lack of adult males sampled in 1996 was because the licence did not permit culling of males over 6 months of age

<i>Age estimates</i>	<i>Male</i>		<i>Female</i>		<i>Total</i>	
	<i>Number</i>	<i>%</i>	<i>Number</i>	<i>%</i>	<i>Number</i>	<i>%</i>
Nov-Dec 1996						
<i>6 months</i>	21	24.4	13	15.1	34	39.5
<i>≥18 months</i>	0	-	52	60.5	52	60.5
<i>Total</i>	21	24.4	65	75.6	86	100
Nov 1997						
<i>6 months</i>	89	33	75	27.7	164	60.7
<i>≥ 18 months</i>	7	2.6	99	36.7	106	39.3
<i>Total</i>	96	35.6	174	64.4	270	100

6.3.2 Age determination by analysis of tooth eruption and tooth wear

The technique of analysis by tooth eruption and tooth wear (TEWT) was performed on mandibles (lower jawbones) in collaboration with Dr. Rolf Langvatn at the Norwegian Institute for Nature Research (NINA), Trondheim, Norway. No information about the individual samples (such as sex, body condition) were available to Dr. Langvatn. The total material consisted of mandibles from 57 of the saigas sampled during the study. Age assessment was performed using estimates based on tooth eruption pattern and wear (Gruzdev & Pronyaev, 1994; Langvatn, 1997). The whole procedure was then repeated without reference to the previous results, so that there were two independent estimates.

Thirty-eight (67%) of the estimates were identical using the two different methods, 10 (18%) varied by only 1 year, and 9 (16%) varied by 2 years. The youngest animal that was given age estimates varying by two years was estimated as either 3 or 5 years old. It seems to be more difficult to give accurate estimates in older animals. This ties in with the results of previous studies on other species (Bianchi *et al.*, 1997; Ratcliffe & Mayle, 1992).

The repeatability of the TEWT was assessed using regression analysis, which showed highly significant correlation; $r^2 = 0.91$, $p < 0.001$ (***) between the two estimates, as was expected (Figure 6.3). The slope of the regression was 1.08 (95% confidence interval 0.99 – 1.18), thus there was no evidence for systematic bias in age estimation between the two independent estimates. The largest difference between estimates was 2 years, and this only occurred in older animals (estimated as over 4.5 years). The suggested age using this technique was taken as the mean of the two estimates. The age range spanned from 0.5 to 10.5 years.

6.3.3 Age determination by the tooth sectioning technique

The first incisor is the largest of the four incisiform teeth and erupts first; it was therefore, chosen for age determination. The two first incisors were removed from 180 saigas. A further set of 57 incisor pairs was removed from the saiga mandibles that had been analysed using the TEWT technique. Microscopic slides were produced from sections of the roots of the incisor teeth according to a procedure described by Reimers & Nordby (1968). Dr. Langvatn performed the examinations, because the author did not have access to the facilities necessary to section teeth. Annuli in the dental cementum were counted and the age was determined controlling for time of eruption of the permanent first incisor

(Gruzdev & Pronyaev, 1994). The whole procedure was repeated a second time, using a different section from the same tooth, such that there were two different estimates.

To save time during the expedition, samples of incisor teeth were not taken from 137 saigas that were judged to be 6 months of age by visual age assessment. A further 5 samples were either not taken by mistake, or lost during transport. These 5 saigas had their age recorded using the visual assessment as >18 months.

Fig 6.3 and 6.4

Fig_chap6_2

145 (61%) of the duplicated estimates were identical, 78 (33%) estimates differed by one year, 11 (5%) estimates differed by two years, and 3 (1%) differed by three years. The difference of 3 years was caused once by a mistake in reading the first section, while the other two were in older animals, aged as 4.5 or 7.5 years, and 8.5 or 11.5 years. The repeatability of the TST was assessed using regression analysis, which showed highly significant correlation between the two estimates ($r^2 = 0.88$, ***), as was expected (Figure 6.4). The slope of the regression was 0.93 (95% confidence interval 0.89 – 0.98). The first estimate (TST 1) slightly underestimated the age compared with the second estimate (TST 2), but there was no evidence of serious bias. The suggested age using this technique was taken as the mean of the two estimates.

6.3.4 Comparison between the TST and TEWT methods

Correlation between the techniques

Fifty-seven saigas were aged using both techniques. Thirty-nine (68%) of the estimates were identical using the two different methods, 12 (21%) varied by only 0.5 years, and 5 (9%) varied by 1 or 1.5 years. Only 1 (2%) varied by 2 years. This was an adult animal, aged as 4 or 6 years.

The correlation between the two techniques was assessed using regression analysis (Figure 6.5). This showed highly significant correlation ($r^2 = 0.97$, ***). The slope of the regression was 1.04 (95% confidence interval 0.989 – 1.096). The estimate from the TEWT compared with the estimate from the TST showed no evidence of bias. It is interesting to note that there was better correlation between the two different techniques (TST and TEWT) than there was between the two estimates using the same technique. This is probably because the means of the values have been used.

In general, the more reliable of any two methods is the one that is least variable between trials, i.e. the one with the smallest confidence interval. In this case both methods gave excellent results. In this study, the confidence interval for the TST was lower than for the TEWT, however the strength of correlation for the TEWT was higher ($r^2 = 0.91$), than for

the TST ($r^2 = 0.88$). Nevertheless, the fact that there were more samples for the TST meant that this might be the reason it had a narrower confidence interval. To obtain confidence intervals for the TST samples that were comparable to the TEWT ones, five sets of 57 samples were picked at random from the 237 TST analysed samples, and a regression analysis performed on each set. All five sets gave much higher confidence intervals than the regression analysis for all the samples tested by TST, and they were all in the range of the equivalent confidence interval of the TEWT (Table 6.2). Thus it was concluded that there was no significant difference between the two methods in terms of their variability between trials.

Table 6.2 Output from regression analysis performed on the results from 5 sets of 57 randomly selected samples tests by TST. Conf int = confidence interval, *** = $p < 0.001$

<i>Samples</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Conf int</i>	r^2	<i>Significance</i>
<i>Random1</i>	0.79	0.96	0.16	0.89	***
<i>Random2</i>	0.87	1.12	0.25	0.83	***
<i>Random3</i>	0.80	1.00	0.20	0.85	***
<i>Random4</i>	0.85	1.01	0.16	0.91	***
<i>Random5</i>	0.79	0.96	0.17	0.89	***
<i>All TST</i>	0.89	0.98	0.09	0.88	***
<i>All TEWT</i>	0.99	1.18	0.19	0.91	***

Discussion

As shown above, there was no significant difference between the results achieved by either of the laboratory techniques. Considering there was better correlation between age estimates using two different techniques (TST and TEWT) than there was between the estimates using the same technique (TST or TEWT), it seems preferable to aim to use both methods. The greater reliability gained by using them both might offset the extra time and effort needed to perform both techniques rather than just one.

Table 6.2

Fig 6.5

Fig chap6_3

In some studies accuracy in ageing of animals may not be essential, in which case performing a single technique may be adequate. The disadvantage to using the TEWT is that it is more time-consuming to collect mandibles than incisor teeth, and the weight and bulk of the mandibles make transport more expensive. However, the TST is a more time consuming technique and it requires facilities for sectioning the teeth, microscopes to read the annuli and well trained laboratory technicians. Considering the good correlation between TST and TEWT, either could be used.

6.3.5 Comparison of laboratory findings with visual estimates

Correlation between laboratory findings and visual estimates

First a direct comparison of the results was carried out, then the laboratory data were cleaned to remove mistakes and to make them consistent with analysis results. In the first comparison, the proposed age was taken as the mean of the estimates obtained by TST and TEWT (where these were done) or as the TST estimate if TEWT was not done, in order to enable comparison with the visual estimates. The correlation between these estimates and visual methods was assessed using regression analysis. This showed good correlation ($r^2 = 0.54$, $N=84$, ***). The slope of the regression was 0.49 (95% confidence interval 0.39 – 0.59), however the relationship was not linear, therefore the regression is not valid. The laboratory analysis consistently overestimates the age compared to the visual assessment, but the differences became proportionally smaller as age increases. Table A4.3 (Appendix 4) shows the age estimates by visual assessment and laboratory analysis.

Next the suggested age, based on the laboratory analysis, was altered to take into account the date the animal was culled, such that all animals culled in autumn were aged 0.5 years, 1.5 years, 2.5 years etc. If the difference between the TST and TEWT estimates was only 0.5 years, the age was taken as the non-integer estimate. Where only the TST had been performed, and the mean estimate was of integer age, the age was taken as 0.5 less than the mean estimate. This was done because the proposed laboratory estimates tended to over-

estimate the ages in comparison with the visual estimates (see above). If the animal was sampled in spring, the mean was rounded to the nearest whole number. On one occasion where one of the duplicated TEWT estimates was thought to have been a mistake, the age was determined to be that of the mode of the three other estimates (TEWT and TST). On the two occasions where one of the duplicated TST estimates was thought to have been a mistake, the age was determined to be that of the other estimate, and not the mean of the two.

The correlation between the two methods was re-assessed using regression analysis. This showed good correlation ($r^2 = 0.62$, $N=84$, ***). The slope of the regression was 0.57 (95% confidence interval 0.47 – 0.67), however, this relationship was also not linear, therefore the regression was not valid. The suggested age based on the laboratory analysis was still higher than the visual estimate (Figure 6.6), although the difference was less after the laboratory-based suggested ages were adjusted to take into account the date of culling.

Discussion

The visual assessment of males between the age of 6 months and one year is likely to be accurate, as the horns of 6-month-old and 1-year-old animals are difficult to mistake for those of older males, leading to the conclusion that the age determination by laboratory methods is less reliable than visual assessment for this age group of males. However, for males under two years of age the difference between the estimates does not exceed one year, therefore, it seems that the laboratory method is accurate within one year either side, as is demonstrated in Table A4.4 (Appendix 4). Over the age of one year the visual method is likely to become much more unreliable, however, there is still a maximum of only 2 years difference between the laboratory and visual estimates.

Fig 6.6

Figures chap 6_4

Female saigas are more difficult to age by the visual method, as they have no horns. As all females were culled in autumn, it was theoretically possible to differentiate visually between those aged 6 months and those aged 18 months or over by using the tooth eruption method. If the back molars had not erupted, the animal was 6 months old, if they had erupted it was \geq 18 months. If carefully done, this method is accurate, however it is easy to make mistakes under field conditions. A bias could have occurred in either direction, depending on whether the technician performing the visual age assessment failed to notice the last molars, thus underestimating the age, or erroneously thought they were present, thus overestimating the age.

It is thus difficult to know which of the methods (visual and laboratory) is the most accurate for female saigas. Considering the number of mistakes made among the juvenile males using the laboratory method, it is possible that the 24 female saigas aged as over 1 year by the visual assessment method and aged 0.5 years by the laboratory method, actually were 1.5 years old (Table 6.3). It is also possible that the 15 females aged 1.5 years by the laboratory method actually were 0.5 years, as observed by the visual method. I would have thought the former was more likely under field conditions.

Our sections were analysed blind by the investigator; who did not know which animals had been culled in spring and which in autumn. However, using the techniques described here he managed without mistake to differentiate between the 8 saigas sampled in May (1 year of age) and the 229 sampled in November (0.5 or 1.5 years of age). This gives a measure of confidence in the laboratory assessments.

Table 6.3 Female saigas aged <3 years by laboratory methods (taking into account date culled), compared to age estimated by visual assessment. Age by laboratory methods is in rows, age by visual assessment in columns

	<i>Age by visual assessment</i>				<i>Total</i>
	<i>6 months</i>		<i>? 18 months</i>		
	<i>Number</i>	<i>%</i>	<i>Number</i>	<i>%</i>	
<i>Laboratory</i>					
<i>6 months</i>	16	40.0	24	60.0	40
<i>1.5 years</i>	15	32.6	31	67.4	46
<i>2.5 years</i>	3	10.7	25	89.3	28
<i>Total</i>	34	29.8	80	70.2	114

The current analysis has looked for internal consistency within one technique, and agreement between techniques. It does not quantify the precision or any bias in estimating the actual age of the sampled saiga. For this reason, this study needs to be complemented by an experiment trying these methods out on known-age animals.

The final assessment of the saigas in the study was arrived at as follows: All saigas aged 6 months by the visual method were assumed to be this age, even if the laboratory estimate was different. The suggested age of the other sampled saigas was taken as the age suggested by the laboratory method (as defined in section 6.3.5).

6.3.6 *The age profile of saigas in the study*

Mean age of sampled saigas was 1.7 years (N=369, median age 0.5, range 0.5 – 10.5, SD=2.0). The ages of sampled males and females are depicted in Table 6.4. Sampled females were significantly older than males (Mann-Whitney U = 9066, ***). The mean age of females was 2.3 years, (median = 1.5, N=236, range 0.5 – 10.5, SD=2.3), whereas the mean age of males was 0.7 years, (median = 0.5, N=133, range 0.5 – 5, SD=0.7). The study sample did not constitute a natural herd, and there was an unquantifiable bias against adult males because of the licensing situation.

Table 6.4 Suggested age and sex of the sampled saigas

<i>Age in yrs</i>	<i>No.males</i>	<i>% males</i>	<i>No.females</i>	<i>% females</i>	<i>Total</i>
<i>0.5</i>	111	50.3	110	49.7	221

<i>1</i>	11	84.6	2	15.4	13
<i>1.5</i>	5	13.2	33	86.8	38
<i>2.5</i>	1	3.8	25	96.2	26
<i>3.5</i>	0	-	15	100	15
<i>4</i>	4	100	0	-	4
<i>4.5</i>	0	-	15	100	15
<i>5</i>	1	100	0	-	1
<i>5.5</i>	0	-	16	100	16
<i>>6</i>	0	-	25	100	25
<i>> 1.5 years</i>					
<i>(age not estimated)</i>	0	-	5	100	5
<i>Total</i>	133	35.6	241	64.4	374

6.4 Ecological results - adult saigas

6.4.1 Population structure

Mean age of saigas sampled in the two autumn hunts in Betpak-dala was 1.7 years (N=351, median age 0.5, range 0.5 – 10.5, SD=2.1). The oldest saiga in our study was a female estimated to be 10.5 years old (the four individual age estimates ranged from 8.5 – 11.5 years). To date the oldest recorded saiga in a hunted population is a 9.5-year-old female, caught with an ear-tag marking the year of birth (Fadeev & Sludskii, 1982).

Between 1965 and 1970, 18,556 saiga calves in Kazakhstan were marked with ear tags stating the year and location of tagging. Of these 1,408 were culled during autumn hunts between 1965 and 1975, giving scientists reliable information on their ages (Fadeev & Sludskii, 1982). Mean age of these saigas was 2.1 years (N=1408, median age 1.5, range 0.5 – 7.5, SD=1.2), which was significantly higher (Mann-Whitney U = 143926, ***) than in our study 1.7 years (N=369, median age 0.5, range 0.5 – 10.5, SD=2.0). However, the difference in mean age was only 0.4 years, well within the margin of error (1-2 years) of our age-estimation techniques, thus it may have no actual biological significance.

There was a significantly higher (Yates corrected $\chi^2=28.7$ df=1, ***) proportion of adults aged over 6 years in our study than in the study by Fadeev & Sludskii (Table 6.5). This is surprising, as one would expect fewer animals in this age category, due to the increased poaching in recent years. However, the sample size is very low, and may thus not be demonstrating accurate changes in demography. Additionally, differences in sampling

methods, and the fact that our age-estimation is not accurate could have introduced biases that are difficult to quantify.

Table 6.5 The age of saigas sampled in autumn hunts in this study, compared to those sampled by hunters in 1965 – 1975 (Fadeev & Sludskii, 1982). Median age was 0.5 years for this study, 1.5 years for the Fadeev & Sludskii study. $df = 1$, $\chi^2 =$ Yates corrected χ^2

<i>Age in years</i>	<i>This study</i>		<i>Fadeev & Sludskii (1982)</i>		χ^2	<i>p</i>
	<i>Number</i>	<i>%</i>	<i>Number</i>	<i>%</i>		
<3 years	285	81.2	1189	84.4		NS
3.5 – 5.5	46	13.1	204	14.5		NS
>6 years	20	5.7	15	1.1	28.7	***
Total	351	100	1408	100		

Table 6.6 shows the age and sex ratios of saigas culled in Kazakhstan between 1989 and 1993. The proportion of adult males in our study fell within the range obtained in Kazakhstan overall during the period 1989 – 1993, however our study had a greater proportion of juveniles than was recorded in Kazakhstan during that period. Unfortunately, the crudeness of the data precluded further statistical analysis.

Table 6.6 Age and sex ratios of saigas sampled in two different studies. a) this study; b) a study of commercial capture (Bekenov *et al.*,1998). The data from the Betpak-dala population and for all of Kazakhstan are the ranges for the years 1989 – 1993, and were obtained from animals captured commercially using corrals. ‘male’ = adult males; ‘female’ = adult females; ‘juvenile’ = both sexes, less than 1 year old

	<i>% male</i>	<i>% female</i>	<i>% juvenile</i>
<i>a) This study (Betpak-dala only)</i>	3.3	36.0	60.7
<i>b) Betpak-dala 1989 – 1993</i>	5.3 – 11.6	47.6 – 67.7	22.7 – 46.6
<i>a) This study (overall)</i>	5.9	35.0	59.1
<i>b) Kazakhstan 1989 – 1993</i>	2.3 – 17.9	37.3 – 78.0	18.1 – 46.4

6.4.2 Body condition of sampled saigas

A higher proportion of female saigas (32.8%) was in ‘good’ body condition compared to males (24.1%), and a lower proportion of females (15.7%) was in ‘poor’ body condition compared to males (19.5%), however, the differences were not statistically significant.

Older animals were in significantly better condition than younger animals (N= 369, Kruskal-Wallis H = 42.0, df=2, ***) (Table 6.7). This holds true also when males and females are analysed separately, although the results for the males (Kruskal-Wallis H = 27.1, df=2, ***) were not as highly significant as for the females (Kruskal-Wallis H = 6.9, df=2, *), because of the smaller sample size.

Table 6.7 The effect of age on the body condition score of saigas. Mean age is given with the 95% confidence interval. The difference in condition is significant (Kruskal-Wallis H = 42.0, df=2, ***)

<i>Condition</i>	<i>Sample size</i>	<i>Mean age</i>	<i>Age range</i>	<i>Std. Dev.</i>
Poor	63	1.1 ± 0.4	0.5 – 8.5	1.74
Average	198	1.5 ± 0.3	0.5 – 9.5	1.88
Good	108	2.4 ± 0.4	0.5 – 10.5	2.26

The proportion of saigas in ‘good’ body condition was lower in autumn than in spring, while the proportion in ‘poor’ body condition was three times as high in autumn as in spring. Due to the small sample size in spring, these results are not statistically significant, however, they are rather surprising, as one would expect saigas to be in better condition in autumn, after summer grazing. The summer of 1997 was very dry in Kazakhstan, so considering that the majority of saigas were sampled in the autumn of 1997 it is possible that they were in a poorer than usual condition because of the summer drought. It is also possible that there could be some observer bias, although this was minimised by the same person always performing the condition scoring. The mean body condition score of animals sampled in Betpak-dala the autumn of 1996 was significantly higher than those sampled in the autumn of 1997 (Kruskal-Wallis H = 5.6, df=1, *). Table 6.8 relates year to body condition score.

Table 6.8 The relation between year and body condition score of saiga antelopes. The differences in body condition between 1996 and 1997 were statistically significant ($\chi^2=12.7$, df=2, **)

<i>Condition</i>	<i>1996 (autumn)</i> <i>% (n=84)</i>	<i>1997 (autumn)</i> <i>% (n=267)</i>
Poor	18.6	17.3
Average	37.2	58.4
Good	44.2	24.3
Total	10	100

6.5 Ecological results - saiga calves

6.5.1 Sex ratio of saiga calves

In saiga calves, the sex ratio of embryos and neonates is generally close to 1:1 (Fadeev & Sludskii, 1982; Bekenov *et al.*, 1998). Fadeev & Sludskii (1982) noticed that in years when the saiga population was at its peak (1972 and 1974) the sex ratio of embryos was in favour of males, and when it was at its lowest (1976 – 1978) was in favour of females. However, on analysis of data from 1983 – 1997, the slight fluctuations observed in the sex ratio had no clear link with climatic conditions or population size (Bekenov *et al.*, 1998). During this period the highest proportion of newborn males recorded was 59.8% (Betpak-dala, 1988). The highest proportion of newborn females recorded was 55.3% (Ural, 1992). The method used in all these studies was to cull pregnant saigas, and sex the embryos found *in-utero*. Hence there was unlikely to be any bias towards one particular sex.

In addition to the 167 calves caught for the purpose of this study in Betpak-dala, a further 27 were caught at the same time for another, linked study (Experimental brucellosis in the saiga antelope, Ivanov *et al.*, 1999). The only data obtained from these 27 calves was their sex; therefore, they have only been included in the analysis of sex ratio. In Betpak-dala there was no statistically significant difference between the numbers of male and female calves sampled. In Ustiurt significantly more males were caught (Yates corrected $\chi^2 = 6.3$, $df=1$, **). The difference between the sex ratio of calves found in Betpak-dala in 1997 and that found in Ustiurt in 1998 was significant ($\chi^2 = 7.05$, $df=1$, **).

Table 6.9 Comparison of saiga calf sex ratio in Betpak-dala and Ustiurt. The difference between the two populations was significant ($\chi^2 = 7.05$, $df=1$, **)

	<i>No. males</i>	<i>% males</i>	<i>No. females</i>	<i>% females</i>	χ^2	<i>p</i>
<i>Betpak-dala</i>	85	43.8	109	56.2		NS
<i>Ustiurt</i>	336	55.1	274	44.9	6.3	**

6.5.2 Estimated ages of saiga calves

The mean age of captured saiga calves was 25.6 hours (range 6 – 72, $SD=13.4$, mode = 24). Table 6.10 depicts the age ranges of sampled calves in the two populations. The number of calves caught aged 48 – 72 hours is much lower than calves aged 24 hours because they get increasingly more difficult to catch as they grow older. The low number of calves under 24 hours may be an artefact of the classification system, where calves under 24 hours which were born in very windy and sunny circumstances may appear older

and, therefore, classified as 24 hours of age. It may also be that the team reached the calving area a few days late, and there were fewer calves than expected under 24 hours.

<i>Age in hours:</i>	<6	12	24	48	72	<i>Total</i>
<i>Betpak-dala</i>						
Male	9	10	28	36	0	83
Female	14	13	39	18	0	84
<i>Total</i>	23	23	67	54	0	167
<i>Ustiurt</i>						
Male	28	47	198	59	4	336
Female	26	34	173	39	2	274
<i>Total</i>	54	81	371	98	6	610

The reason the sample size was so much higher in Ustiurt than in Betpak-dala is that the concentration of saigas found in Ustiurt was of about 200,000 - 250,000 animals, whereas in Betpak-dala a total of 12,000 - 15,000 saigas were seen in the study area. Many of these had already calved, and the calves were already too old to catch. The estimated age of saiga calves was significantly higher both when the temperature at dawn was lower (Kruskal-Wallis $H = 66.6$, $df=5$, ***), and when the temperature at midday was lower (Kruskal-Wallis $H = 50.9$, $df=5$, ***) (Figure 6.7). This is probably because calves are more sluggish in cold weather (personal observation), and thus easier to catch. However, it is possible calves could dry sooner in colder weather.

6.5.3 Weight of saiga calves

Figure 6.8 shows the frequency distribution of the weight of saiga calves. Calves born in Betpak-dala were significantly heavier ($t=4.6$, $df=246$, ***) than those born in Ustiurt, as seen in Table 6.11. The results were confounded by the fact that the mean age of calves sampled from Betpak-dala was greater than in Ustiurt, and older calves are generally heavier. A generalised linear model of the association between weight, age and location born was constructed, with age treated as an ordinal variable due to the problems of accurate ageing discussed above. As was expected from the univariate analysis, the model showed that weight was highly significantly related to age ($F = 31.2$, $df = 4$, ***), and to location ($F = 16.0$, $df = 1$, ***). The interaction term was not significant.

Figure 6.7 Mean age in hours of calves with changing temperature at dawn. The estimated age of saiga calves was significantly higher when the temperature at dawn was lower (Kruskal-Wallis $H = 66.6$, $df=5$, ***)

Table 6.11 Comparison of the weight in grams of saiga calves in Ustiurt and Betpak-dala (t=4.6, df=246, ***)

	<i>Sample size</i>	<i>Mean weight</i>	<i>Range</i>	<i>Std Dev.</i>
<i>Betpak-dala</i>	167	3480 ± 68	1950 – 4700	448
<i>Ustiurt</i>	610	3303 ± 32	1850 – 4700	406
<i>Total</i>	777	3341 ± 30	1850 – 4700	421

The mean weight of male calves was significantly higher than that of female calves in both the populations (Table 6.12), however in Betpak-dala these results were confounded by the fact that male calves were significantly older than female calves. In Ustiurt, where the sample size was much larger, there was no significant difference in age between the sexes. Thus weight may vary with the sex of the calf. This has also been found in previous studies. Fadeev & Sludskii (1982) reported that newborn male calves weigh on average 3.9kg (range 3.5 – 4.5), and newborn female calves weigh 3.6kg (range 3.1 – 4.2). Bannikov *et al.* (1961) reported mean birth weights of 3.3kg (range 2.0 – 4.4) in males and 3.1kg (range 1.9 – 4.4) in females. According to Fadeev & Sludskii (1982), male and female calves grow at roughly the same rates. Our data fit well with the data from Bannikov *et al.*, and are slightly lower than those of Fadeev & Sludskii.

Fig 6.7

Fig 6.8

Figures chap6_5

Fig 6.8

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	<i>Sample</i>	<i>Mean</i>	<i>Range</i>	<i>Std. Dev.</i>	<i>T</i>	<i>df</i>	<i>P</i>
<i>weight</i>							
<i>Betpak-dala</i>							
Male	83	3621 ± 87	2600 – 3700	404			
Female	84	3340 ± 96	1950 – 3400	448	4.3	164	***
<i>Ustiurt</i>							
Male	336	3384 ± 42	2400 – 4700	395			
Female	274	3204 ± 47	1850 – 4600	398	5.6	581	***
<i>Total</i>							
Male	419	3431 ± 39	2400 – 4700	407			
Female	358	3236 ± 43	1850 – 4700	414	6.6	752	***

6.5.4 Length of saiga calves

The overall mean length of saiga calves was 618.5 cm (range 500 – 740, SD=35.6). In Betpak-dala, the mean length of saiga calves was 616 cm (range 500 – 690, SD=37), in Ustiurt it was 619 cm (range 500 – 740, SD=34), but this difference was not statistically significant. In Betpak-dala, male calves were significantly longer than female calves ($t=2.1$, $df=165$, *). Figure 6.9 shows the frequency distribution of the length of calves. In Ustiurt the mean length of male calves was greater than that of female calves, however, the difference was not statistically significant (Table 6.13).

	<i>Sample size</i>	<i>Mean length</i>	<i>Range</i>	<i>Std. Dev.</i>	<i>T</i>	<i>df</i>	<i>P</i>
<i>Dev.</i>							
<i>Betpak-dala</i>							
Male	83	622 ± 8	510 – 690	35			
Female	84	610 ± 8	500 – 690	37	2.1	165	*
<i>Ustiurt</i>							
Male	336	621 ± 3	500 – 740	34			
Female	274	617 ± 4	510 – 620	34			NS
<i>Total</i>							
Male	419	621 ± 3	500 – 740	34			
Female	358	615 ± 4	500 – 720	35	2.2	752	*

Figures chap6_6

Figure 6.9

Fig 6.10

6.5.5 Comparison of age, weight and length of saiga calves

Regression analysis was used to assess the correlation between weight and length (Figure 6.10). This showed significant correlation ($r^2 = 0.34$, ***); longer calves were heavier as would be expected, however r^2 was low because the relationship was variable. A generalised linear model of the association between length and age showed that length was highly significantly related to age ($F = 20.3$, $df = 4$, ***). Another generalised linear model was constructed, relating weight of saiga calves with the age, length, gender, and location born (Table 6.14). Generally the biological correlates were as expected. The interaction terms for age and location, and for age, location and gender were significant. This showed that length was related to weight; heavier calves were larger, as found above. Interestingly there was an effect of location; male calves and calves born in Betpak-dala were heavier at a given length than were female calves or those born in Ustiurt.

Table 6.14 Generalised linear model relating weight of saiga calves with the age, length, gender, and location born

Factor	F	df	p
Model	41.8	18	***
Intercept	4.6	1	*
Length	367.1	1	***
Estimated age	18.2	4	***
Location	31.0	1	***
Gender	4.1	1	*
Estimated age * location	3.9	3	**
Estimated age * gender	1.4	4	NS
Location * gender	0.3	1	NS
Estimated age * location * gender	4.9	3	**

6.5.6 Newborn saiga calves

A total of 51 calves of the 777 sampled (6.6%) were still wet when they were found, indicating that they were newborn. The proportion of newborn calves sampled in Betpak-dala was significantly greater than the proportion sampled in Ustiurt (Yates corrected $\chi^2=13.8$, $df=1$, ***). In Betpak-dala 22 (13.2%) newborn calves were sampled, in Ustiurt 29 (4.8%) were sampled. Overall, the mean weight for newborn males (3150 grams, range 2400 – 4150, $SD=440$) was greater than that for females (3071 grams, range 1850 – 3750, $SD=464$), although this was not significant. The lack of significance could have been due to the low sample sizes.

Newborn calves in Betpak-dala were significantly heavier than in Ustiurt ($t=2.6$, $df=47$, *), despite the fact that there was no statistically significant difference between the numbers of newborn calves of each gender sampled in the two locations. The mean length (Table 6.15)

was also greater for calves born in Betpak-dala, however this difference was not statistically significant. Within Betpak-dala, the mean weight of male calves (3267 grams, range 2800 – 3950, SD=374) was actually lower than that of female calves (3288 grams, range 2900 – 3750, SD=253), although the difference was not statistically significant. The mean length of male newborn calves (611cm, range 560 – 670, SD=39) in Betpak-dala was greater than that of female newborn calves (599cm, range 540 – 670, SD=39), however, the difference was not statistically significant.

	<i>Sample size</i>	<i>Mean</i>	<i>Range</i>	<i>Std. Dev.</i>
<i>Weight</i>				
Betpak-dala	22	3280 ± 125	2800 – 3950	300
Ustiurt	29	2981 ± 183	1850 – 4150	504
<i>Length</i>				
Betpak-dala	22	604 ± 16	540 – 670	38
Ustiurt	29	591 ± 71	520 – 670	38

Bannikov *et al.* (1961) reported mean length of newborn females as 552cm (range 500 – 670), and of males as 594cm (range 520 – 670). This range is within the range found in our study, however the mean lengths that they found were slightly lower than our results. The actual data from Bannikov *et al.* are not available; therefore statistical tests could not be performed.

The presence of cloudy serum (in domestic livestock usually related to hyperlipidaemia) was noted in 142 out of 581 samples in Ustiurt. Cloudy serum often indicates high amounts of triglycerides in the blood. In newborn calves the occurrence of cloudy serum was significantly less prevalent than in older calves (Yates corrected $\chi^2=5.4$, $df=1$, *). Cloudy serum was noted in only 3.4% (1 of 29) of newborn calves, whereas it was noted in 24.3% (141 of 581) of older calves. From these results it seems that the presence of high amounts of triglycerides in the serum may be normal in young calves, presumably as a result of the very high fat content in saiga milk (Bannikov *et al.*, 1961). It is less likely to be found in newborn calves, possibly as these have not yet suckled. There was no correlation between the gender, weight or length of calves and the presence of cloudy serum. In the future it would be interesting to compare the incidence of cloudy serum in the Ustiurt and Betpak-dala populations.

6.5.7 Proximity to the nearest neighbour

The mean distance of sampled calves to their nearest neighbour was 29 metres, (range 5 – 900, SD=53.5). Calves born in Betpak-dala were found significantly further apart (Kruskal-Wallis H=18.2, df=1, ***) than those born in Ustiurt (Table 6.16). This is an important analysis to carry out because saiga antelopes are gregarious, so that distance to nearest neighbour could be a measure of disturbance, of population density or possibly of the dominance status of the dam. Given that the Betpak-dala population is known to suffer greater human disturbance than the Ustiurt one, it is possible that our results are detecting this.

Table 6.16 Comparison of the proximity to their nearest neighbour of saiga calves in Ustiurt and Betpak-dala. Proximity (distance) to nearest neighbour is given in metres, with the 95% confidence interval

	<i>Sample size</i>	<i>Mean distance</i>	<i>Range</i>	<i>Std. Dev.</i>
Betpak-dala	167	53 ± 18	10 – 900	119.9
Ustiurt	610	22 ± 1	5 – 40	12.0
All calves	777	29 ± 4	5 – 900	53.5

6.6 Interaction with humans

All interviewed farmers and shepherds in the saiga range area of Betpak-dala were asked if they had seen saigas near their village. This information is important, because of transmission of disease between livestock and saigas. Of the people interviewed, 100% (N= 50) had seen saiga less than 500 metres from their village, although less often during the last few years due to a perceived reduction in saiga numbers. Of these, 34% (17 of 50) had seen saigas drink at watering places used by domestic livestock. Table 6.17 details the year in which interviewees perceived the reduction in saiga numbers to begin. 18% of respondents had first noticed fewer saigas appearing in their area in 1993, and 10% had first noticed it in 1994. The one person who had noticed a reduction in 1985 believed this to be the result of a coal mine opening at that time, causing the saigas to move away from the area. He believed that numbers had dropped further since 1992 due to poaching.

Table 6.17 The year in which interviewed farmers and shepherds perceived the reduction in saiga numbers began

<i>Year</i>	<i>Number of responses</i>	<i>% of responses</i>
1985	1	2
1992	4	8
1993	9	18
1994	5	10
1995	3	6
No opinion	28	56
Total	50	100

Thus all the respondents except one believed that the reduction in the saiga population had begun after the independence of Kazakhstan (1991), but previous to the years during which this survey took place (1997 – 1998). One poacher, a young man, was interviewed. He explained that he had started poaching at the age of twelve, but only a few animals a year, to bring meat home for his family on special occasions. When the mine where he worked closed down in 1994 he lost his job and poaching is now his only source of income. He kills about 100 male saigas per month, and splits the proceeds with his partner. He is paid 26 US\$ per kg saiga horn by a middle-man who comes to the village once a month to collect the horns. There are approximately 6 horns to a kilogram, so he earns about US\$ 8.5 per saiga he kills. In a country where the average wage is 60 US\$ a month this is good money.

6.7 Discussion

It is interesting to note that our results showed no significant difference between the birth weight of male and female calves, but within a few days of life male calves become significantly heavier than female calves. This lack of significance between the weight of newborn (wet) calves of differing gender could be due to the low sample size, but if it is a true effect, it may indicate that males grow faster than females. In Betpak-dala, the length of newborn male calves was significantly greater than that of female calves, however, the difference between newborn (wet) males and females was not statistically significant. This lends further credit to the argument that newborn male calves may grow faster than newborn female calves, although the lack of significance could again be due to the low sample size.

In the Ustiurt population, significantly more male than female calves were caught. It is possible there could have been a sex bias in capture, e.g. if male calves were stronger, it is possible they were more likely to escape capture. However, the methods used to catch

calves in Ustiurt and Betpak-dala were identical; thus the relative difference between the two locations is likely to be caused by a true effect. In ungulates such as red deer, a strong influence of birth weight on subsequent mating success has been found for males, such that dominant females in good condition can maximise their lifetime reproductive success by biasing their offspring's sex ratio towards males (Hewison & Gaillard, 1999). The influence of birth weight on subsequent reproductive success is less pronounced for female offspring in red deer, so that females in poor condition are more likely to maximise their lifetime reproductive success by biasing their offspring sex ratio towards females. It is possible that a similar effect could be in operation for saiga antelopes, because they too have males that defend harems of females, with a large amount of variability in reproductive success among males, and much less among females. The higher proportion of males in Ustiurt could be explained in a number of ways, for example local environmental conditions being exceptionally good in the particular year under study. However, the effect of female condition on offspring sex ratio is not fully understood, and no work has been done on reproductive success rates in saigas. Thus any explanations of the differences in sex ratios between the two populations must remain speculative until further work is carried out.

The significant difference in weight of newborn (wet) calves in Betpak-dala and Ustiurt (newborn calves in Betpak-dala were significantly heavier) may be a true effect, as there was no statistically significant difference between the gender ratio of newborn calves in the two locations. This analysis indicates that the calves sampled in Betpak-dala actually may have had higher birth weights than those born in Ustiurt, rather than being heavier simply because they were caught older. This could for example be caused by more favourable climatic conditions in 1997, when the Betpak-dala population was sampled.

The study found a greater proportion of juvenile animals and a smaller proportion of adult females in Betpak-dala than was recorded from animals captured commercially between 1989 and 1993. However, no valid conclusions can be drawn from this, as the data from both the studies are likely to be biased. This study is biased because of the non-random sampling technique and low sample size, the 1989 – 1993 study because of hunting regulations; there was a quota each year, incorporating a set ratio of adult males to adult females to juveniles that could be culled (generally 30 – 40% juveniles). Due to the inherent biases in sampling techniques and in some cases low sample sizes, this study is

more likely to have missed significant effects than to have found them. However, the collection methods were the same in Ustiurt and Betpak-dala, thus any differences between these two locations are likely to be valid.

6.8 Summary

- There was good correlation between the two laboratory based ageing techniques (TST and TEWT). Both gave excellent results, with low variability between trials. However, the study needs to be complemented by a trial on known age animals.
- Older saigas were in significantly better condition than younger animals.
- A generalised linear model demonstrated significant relation between the weight of saiga calves and the age, length, gender, and location born.
- In the Ustiurt population, significantly more male than female calves were caught.
- Newborn calves in Betpak-dala were significantly heavier than in Ustiurt.
- All interviewees in the villages of Betpak-dala had seen saigas less than 500 metres from their village, although less often during the last few years due to a perceived reduction in saiga numbers, which was thought to be connected to higher poaching rates in Betpak-dala.

Chapter 7

Results from the serological survey of saiga antelopes

7.1 Introduction

This chapter discusses the results from the serological studies and clinical examinations of the saiga antelopes sampled during the field expeditions. Viral and bacterial diseases are discussed separately, and the different serological tests employed are compared.

7.2 Viral diseases

All the samples from saiga antelopes tested negative for antibodies to FMD, rinderpest, PPR, bluetongue and EHD viruses. None of the sampled animals had signs of recent clinical disease (males develop stunted horns after FMD infection, this was not observed, neither were vesicles in the oropharynx or coronary band). The sample size required to be 95% confident that a disease is not present at or below a prevalence of 1% in a population of 300,000 animals was calculated using the following formula (Martin *et al.*, 1987):

$$1) \quad n = [1 - (1 - a)^{1/D}] [N - (D - 1)/2]$$

where n is the required sample size

a = probability (confidence level) of observing at least one diseased animal in sample
when the disease affects at least D/N of the population

D = number of diseased animals in population

N = population size

According to the above formula, a sample size of 297 would be required. This study sampled 531 saigas in Betpak-dala and 620 saigas in Ustiurt; thus we can say with >95% confidence that if FMD had been present in either the Betpak-dala or Ustiurt saiga population, we would have found it.

In domestic livestock there was a highly significant difference between the prevalence of vaccination inside and outside of saiga range areas (Yates corrected $\chi^2=27.7$, $df=1$, ***). Outside saiga range, 45.8% of livestock were vaccinated, compared to only 15.6% inside saiga range areas (Table 7.1). The difference was caused mainly by the

cattle, of which 64.5% outside compared to 24.6% inside saiga range areas were vaccinated (Yates corrected $\chi^2=19.4$, $df=1$, ***). The difference between small ruminants was not significant. The high proportion of vaccinated cattle outside saiga range area may have been an artefact of the fact that on one of the three farms sampled outside saiga range area, there had been an outbreak of FMD a year previously, and all the cattle in the village had been vaccinated in response to the outbreak. However, when this farm was excluded from the analysis, the difference was still significant (Yates corrected $\chi^2=5.5$, $df=1$, *). This may have been because these farms were located in the oblasts targeted by the official government vaccination program (Table 4.1).

Both within and outside saiga range areas, seroprevalence among cattle was significantly higher than among small ruminants (Table 7.1). However, the difference was significantly greater within the saiga range areas (Yates corrected $\chi^2=20.0$, $df=1$, ***) than outside (Yates corrected $\chi^2=10.3$, $df=1$, **). Saiga antelopes are more likely to contact small ruminants, commonly kept at pastures away from the village, than cattle, which are often kept close to the village.

Table 7.1 Seroprevalence to FMDV among livestock within and outside of saiga range areas. Seroprevalence among cattle within saiga range areas was significantly lower than outside saiga range areas (Yates corrected $\chi^2=19.4$, $df=1$, ***)

	<i>Sample size</i>	<i>% seropositive</i>	<i>Yates χ^2</i>	<i>Df</i>	<i>P</i>
<i>Cattle</i>					
Within saiga range area	248	24.6			
Outside saiga range area	31	64.5	19.4	1	***
<i>Small ruminants</i>					
Within saiga range area	662	12.2			
Outside saiga range area	17	11.8	0.1	1	NS
			Fisher exact		NS
<i>All livestock</i>					
Within saiga range area	910	15.6			
Outside saiga range area	48	45.8	27.3	1	***

There was no significant difference between seroprevalence to FMDV of livestock sampled in Betpak-dala and Ustiurt (Table A5.1, Appendix), however, when small ruminants were analysed separately, seroprevalence in Ustiurt was significantly higher than in Betpak-dala ($\chi^2=3.9$, $df=1$, *). When tested independently, sheep and goats showed

no significant difference between Betpak-dala and Ustiurt, nor did cattle. This may have been due to the low sample numbers in Ustiurt.

71% cattle and 86.3% of small ruminants in this study were found to be susceptible to FMDV, thus vaccination seems less common than it was before independence. If an FMD epidemic occurs, there is a very high risk of transmission from livestock to saigas. The reason why vaccination of cattle in the saiga range area is significantly less than outside saiga range area may be that the areas sampled outside saiga range are all in the border areas to Uzbekistan and Kyrgyzstan (the ‘at risk’ zone), and are therefore more likely to have been vaccinated against FMDV.

7.3 Brucellosis

7.3.1 Comparison of serological tests

All samples were surveyed using two serological tests; the Rose Bengal Plate Test (RBPT) and the enzyme-linked immunosorbent assay (ELISA). The complement fixation test (CFT) was used to verify any positives. The ELISA showed 1.7% positive whereas the RBPT showed 1.5% positive. The difference between the results of the two tests was significant (Fisher exact test, ***). All the ELISA positive results were confirmed with the CFT. The one RBPT positive that was ELISA negative was also negative on CFT. The actual observations are depicted, in Table 7.2. Animals positive to two separate tests were classified as true reactors. The overall proportion of reactors was 1.7% (20/1151). Thus the RBPT has a slight but significant tendency to produce false negatives (or the ELISA has a slight but significant tendency to produce false positives).

Table 7.2 Comparison of the results from two serological tests: ELISA and RBPT

RBPT	ELISA		Total
	Number negative	Number positive	
Number negative	1130	4	1134
Number positive	1	16	17
Total	1131	20	1151

7.3.2 Brucellosis in adult and juvenile saigas

The prevalence of antibodies to *Brucella* spp. in saiga antelopes aged >6 months old was 2.1% (8/374). This is within the range of 1.3 – 2.9% previously reported in saigas (Postricheva, 1966; Rementsova *et al.*, 1969; Rementsova, 1987). Among sampled females there were 2.5% (6/241) reactors compared to 1.5% (2/133) of males. A higher

seroprevalence in females was expected due to the nature of the disease, however, the difference between males and females was not statistically significant. There was a statistically significant effect of age on the prevalence of antibodies to *Brucella* spp. (Kruskal-Wallis H=3.9, df=1, *); the mean age of seronegative animals was 1.7 ± 0.2 years (range 0.5 – 10.5, SD 2.0, N=361), the mean age of seropositive animals was 3.5 ± 1.9 years (range 0.5 – 7.5, SD 2.7, N=8). This was expected as older animals have had a longer exposure to the risk of infection. In juvenile saigas (6 months of age) the prevalence of brucellosis was 1.5% (3/221), lower than in adult saigas where the prevalence was 3.5% (5/153), however, the difference was not statistically significant. Reactors are shown by age-class and sex in Table 7.3.

Table 7.3 *Brucella* reactors by age-class (adult or juvenile) and gender. The differences are not statistically significant

	<i>Number positive</i>	<i>Sample size</i>	<i>% positive</i>
<i>Adult males</i>	0	22	0
<i>Adult females</i>	5	131	3.8
<i>Juvenile males</i>	2	111	1.8
<i>Juvenile females</i>	1	110	0.9
<i>Total</i>	8	374	2.1

There were 3 juvenile reactors, which is higher than expected, as sexually immature animals are more resistant to infection. However, if they receive a high enough dose they can be infected (A.A.P. Macmillan, pers.comm.). In cattle, it is generally considered that maternal antibodies wane by 6 months, thus maternal antibodies are unlikely to be the cause of a positive reaction in a 6-month-old saiga (A.A.P. Macmillan, pers.comm.). Calves may be born congenitally infected with brucellosis (Nicoletti, 1990). Most calves will self-cure, but some may remain infected and seroconvert at a later date. This may be the most likely reason for the seropositivity seen in juveniles.

All samples (N=10) in the Ustiurt population were negative, whereas in the Betpak-dala population the overall prevalence was 2.2% (N=364). However, due to the low sample size in Ustiurt the difference is not statistically significant. There was no relationship between the condition of the animal and the presence of detectable antibodies to *Brucella* spp.

7.3.3 *Brucellosis in saiga calves*

The overall seroprevalence of antibodies to *Brucella* spp. in saiga calves was 1.5% (12/777). All samples in the Ustiurt population (N=610) were negative, whereas in the Betpak-dala population the overall prevalence was 7.2% (12/167). The difference between the two populations was statistically highly significant (Fisher exact ***). Gender had no effect on seroprevalence among calves in Betpak-dala, which was 7.2% (N=83) among male calves and 7.1% (N=84) among female calves. Although calves in Betpak-dala had higher seroprevalence (7.2 %) than adult females (3.8%), the difference was not statistically significant.

The mean weight of seropositive calves was significantly lower than that of seronegative calves ($t = 2.96$, $df = 14$, *). The mean length of seropositive calves was also lower than that of seronegative calves, but the difference was not significant (Table 7.4). Domestic cows and ewes with brucellosis may give birth to weak calves and lambs (Amiraeiev *et al.*, 1986; A.A.P. Macmillan, pers.comm.); it is possible this occurs in saigas also.

Table 7.4 Test result for antibodies against brucellosis in saiga calves in Betpak-dala.

The mean is given with the 95% confidence interval. DF=1

	<i>Sample size</i>	<i>Mean</i>	<i>Range</i>	<i>Std. Dev.</i>	<i>KW</i>	<i>P</i>
<i>Age in hours</i>						
Negative	155	27 ± 2	6 – 48	15.3		
Positive	12	33 ± 10	6 – 48	16.8	1.4	NS
<i>Distance in metres</i>						
<i>to nearest neighbour</i>						
Negative	155	56 ± 18	10 – 900	114		
Positive	12	25 ± 1	20 – 30	2.6	0.01	NS
<i>Adult group size</i>						
Negative	155	147 ± 14	1 – 300	86		
Positive	12	90 ± 34	10 – 200	60	5	*
<i>Weight in g.</i>						
Negative	155	3503 ± 70	1950 – 4700	446		
Positive	12	3175 ± 206	2500 – 3800	364	6.7	**
<i>Length in cm</i>						
Negative	155	617 ± 6	500 – 690	36.5		
Positive	12	602 ± 21	510 – 650	37.4	1.8	NS

7.3.4 Bacteriology

Bacteriae of the *Brucella* spp. were not isolated from any of the biopsies or swabs taken from placentae and newborn saiga calves. This may have been an artefact of the low sample size; in Betpak-dala only 7 fresh placentae were found, and swabs were taken from

only two seropositive calves (only two seropositive calves were wet). The lack of culture positive samples may also reflect the difficulty in isolating *Brucella* organisms (Nicoletti, 1990). As differentiation of *B. abortus* and *B. melitensis* is not possible by serology alone, either organism could have been the cause of the serological results. *B. melitensis* has been isolated from saigas previously, and as this is the main pathogen of sheep and goats, species that more commonly share pastures with saigas than cattle do, it seems more likely that the antibodies were raised against infection with *B. melitensis*.

7.3.5 Discussion

When both adults and calves were included, the overall prevalence of antibodies to brucellosis was 3.8% (N=531) in the Betpak-dala population, whereas in the Ustiurt population it was zero (N=620). This difference in prevalence between the two saiga populations sampled was highly significant (Yates corrected $\chi^2=21.6$, df=1, ***). According to official statistics for the years of the study, brucellosis was present among domestic livestock in both Ustiurt and Betpak-dala. The density of livestock was higher in Betpak-dala than in Ustiurt, hence the zero prevalence in Ustiurt may reflect a lower saiga – livestock interaction in Ustiurt compared to Betpak-dala. Although the official statistics have not been verified by independent studies, they are unlikely to be overestimating the prevalence of brucellosis, considering the current political climate.

Other authors have reported seroprevalence of antibodies to *Brucella* spp. in 1.3 – 2.9% of saiga (Postricheva, 1966; Rementsova *et al.*, 1969; Rementsova 1987). This study sampled 610 saigas in Ustiurt; enough samples to state with 99% confidence that brucellosis would have been detected if the disease was present at a prevalence of 1% in a population of between 50,000 and 500,000 animals (see section 7.2).

7.3.6 Comparison with livestock

All samples from domestic livestock in Ustiurt were negative, however, the sample size was small, and all were taken from a single farm and were thus not representative of the whole area. Seroprevalence to *Brucella* spp. in Betpak-dala was significantly higher in saigas than in small ruminants (Yates corrected $\chi^2=6.2$, df=1, *), however, the difference between cattle and saigas was not significant, nor was the overall difference between domestic livestock and saigas (Table 7.5). Considering that saigas are more likely to contact small ruminants than cattle, the higher seroprevalence among saigas compared to

small ruminants may reflect the fact that saigas are more susceptible to infection than domestic livestock are. In Betpak-dala more cattle and small ruminants were positive than in Ustiurt or outside saiga range, however, the differences are not statistically significant, because of the low sample size in the other areas. According to official statistics, the seroprevalence of brucellosis among cattle in Chalkar *raion* (the only *raion* in Ustiurt sampled in this study) was 4.1% in 1998 (see Table 8.16). The higher seroprevalence of saigas in Betpak-dala may thus be due to greater saiga – livestock contact in this area, rather than a higher prevalence of the disease among domestic livestock in Betpak-dala.

Table 7.5 Comparison of the seroprevalence to *Brucella spp.* of saiga and livestock in Ustiurt, Betpak-dala and outside saiga range area. % positive = percent seropositive

	<i>Ustiurt</i>		Betpak-dala		<i>Outside saiga range</i>	
	% pos	Sample	% pos	Sample	%	Sample
Cattle	0	9	5.9	239	3.2	31
<i>Small ruminants</i>	0	50	1.3	612	0	17
<i>Saiga</i>	0	610	3.8	531	-	0

7.4 Summary

The results discussed in this chapter are as follows:

- No evidence of infection with FMDV was found in saigas; the entire saiga population may be susceptible.
- Seroprevalence to FMDV of domestic livestock located within the saiga range was significant lower than outside the saiga range area. With only 15.6% of livestock within the saiga range area immune to FMD, there seems to be a high risk of FMDV transmitting to saiga antelopes in the event of an epidemic near saiga range areas.
- No evidence of infection with epizootic haemorrhagic disease, peste-des-petites-ruminants or rinderpest viruses was found in saigas. This was expected, as there have been no reports of these diseases among saigas. The zero seroprevalence to bluetongue was highly unexpected, considering the high seroprevalence found in domestic livestock (see chapter 9, section 9.2).

- Seroprevalence to *Brucella* spp. in saigas was within the range previously reported, thus it seems that there has been no discernible increase in the prevalence of brucellosis among saigas after the independence of Kazakhstan, in contrast to the situation among domestic livestock. This may be related to the dramatic drop in livestock numbers, and the likely consequent decrease in contact between saigas and domestic livestock.
- The mean weight of calves seropositive to *Brucella* was significantly lower than that of seronegative calves. It is possible that female saiga antelopes with brucellosis may give birth to weak calves.
- *Brucella* organisms were not isolated from any samples taken, thus it was not possible to identify the species of *Brucella* responsible for infection.
- No evidence of infection with brucellosis was found in the Ustiurt saiga population. The significantly higher prevalence in Betpak-dala may be because the saiga – livestock interaction is greater there than in Ustiurt.
- The significantly higher seroprevalence to brucellosis of saiga antelopes compared to small ruminants may reflect the fact that the saiga is more susceptible to infection, as has been suggested in the literature.

Chapter 8

Results from the serological survey of livestock – FMD and brucellosis

8.1 Introduction

This chapter details the results of the serological surveys for antibodies against FMDV and *Brucella* spp in livestock. Factors important for the epidemiology of the diseases were identified, and key factors were chosen for hierarchical modelling. Additionally, the results from the questionnaire survey of veterinary surgeons are presented.

8.2 Foot-and-mouth disease

8.2.1 Prevalence of antibodies to foot-and-mouth disease virus

Antibodies against FMDV were detected in 29.0% (81/279) of cattle, 13.8% (75/542) of sheep and in 5.8% (8/137) of goats. The difference in prevalence of antibodies between the species is highly significant ($\chi^2=44.3$, $df=2$, ***). Cattle had significantly higher prevalence of antibodies than small ruminants (sheep and goats classed together) (Yates corrected $\chi^2=38.2$, $df=1$, ***). With cattle excluded from the analysis, seroprevalence among sheep was significantly higher than among goats (Yates corrected $\chi^2=5.8$, $df=1$, *).

8.2.2 Prevalence of antibodies related to infection with foot-and-mouth disease virus

Of the livestock that were seropositive, only 8 were recorded positive to the ELISA test for antibodies to non-structural proteins, indicating that these were the only animals that had been exposed to infection with FMDV (Mackay *et al.*, 1998). This result is validated by the fact that these animals were all cattle from a village that had experienced an outbreak of FMD a year previously. Thus 2.9% of all tested cattle had experienced infection with FMDV, whereas none of the small ruminants (sheep and goats) had. The livestock that were recorded positive against FMDV types A or O, but were negative to the ELISA for non-structural proteins, were most likely vaccinated against FMDV, rather than infected. Thus Table 8.1 presents evidence of previous vaccination, rather than infection with FMDV. In animals under the age of 6 months, antibodies are most likely to be maternal, caused by infection or vaccination of the dam, however these have all been grouped together in the analyses. Among cattle, 14.0% of animals were under 6 months of age, in sheep 13.6% were less than 6 months old, and in goats 18.2% were under 6 months. A second complete analysis excluding all animals under the age of 6 months was performed.

The results were similar as to when animals of all ages were included, therefore, only these results have been included.

8.2.3 Factors affecting the prevalence of antibodies against FMDV

Origin of animal

Of the 958 livestock sampled, 71 (7.4%) had been bought in, the rest had been born on the farms where they were sampled. There was almost no difference between cattle and sheep; 7.5% of cattle and 8.9% of sheep had been bought in (Table A5.2, Appendix 5), however, the proportion of goats bought in was significantly less ($\chi^2=8.73$, $df=2$, *); 1.5%.

There was a highly significant difference (Yates corrected $\chi^2=13.8$, $df=1$, ***) between the seroprevalence of animals born on the farm, and those bought in from elsewhere (Table 8.1). The results from sheep and goats (small ruminants) have been pooled, because the epidemiology of the diseases under study are similar in these species, and under Kazakh animal husbandry practices they are kept together. Seroprevalence among bought-in cattle was significantly higher (Yates corrected $\chi^2=10.3$, $df=1$, **) than those born on the farms, as it was among small ruminants (Yates corrected $\chi^2=3.87$, $df=1$, *). This may be because animals must have veterinary certificates issued before sale, and animals are usually vaccinated against diseases common in the region at the time of examination. The data could be confounded by the fact that 11 of the 71 animals that had been bought in came from the farm that had experienced an outbreak of FMD. However, excluding these from the analysis still gave a statistically significant difference between those born on a farm and those bought in (Yates corrected result: $\chi^2 = 14.1$, $df =$ ***). Thus, there was evidence that animals which were bought-in were more likely to have been vaccinated, however, 66.2% (47 /71) of bought-in animals had no detectable antibodies to FMDV. This could be because vaccines given were ineffective, or it could indicate that veterinary control over the transport of livestock was inadequate.

<i>Origin</i>	<i>No. positive</i>	<i>% positive</i>	<i>Sample size</i>	χ^2	<i>DF</i>	<i>P</i>
<i>Cattle</i>						
Born on farm	68	26.4	258			
Bought-in	13	61.9	21	10.3	1	**
<i>Small ruminants</i>						
Born on farm	11	22.0	50			
Bought-in	72	11.4	629	3.9	1	*
<i>Total</i>						
Born on farm	140	15.8	887			
Bought-in	24	33.8	71	13.8	1	***

Vaccination status

Only 1.9 % (18/958) of livestock were believed to have been vaccinated against FMDV during the previous two years (Table 8.2). These animals were from the same village, but had two owners. The difference in seroprevalence between those thought by their owners to have been vaccinated, and those thought not to have been vaccinated was significant (Yates corrected $\chi^2=7.8$, $df=1$, **). Of the supposedly vaccinated livestock, 55.6% had no detectable antibodies. This could be because the vaccine was ineffective, or the antibodies may already have declined to undetectable levels, or the owners were wrong. Of the livestock believed by their owners not to have been vaccinated against FMDV, 16.6% (156/940) had antibodies to FMDV. Only 5 of these animals had acquired antibodies by infection, and these were all from a single peasant farm, where the owner had recently acquired them and knew nothing of their previous medical history. Assuming that the ELISA for non-structural proteins is accurate, the other 151 animals had owners that either did not know their vaccination status, or gave misleading information. These animals were owned by 41 different owners, indicating that 75.9 % of owners (N=54) were ignorant about the vaccination status of their livestock. It is possible that livestock had been vaccinated more than 2 years previously, and retained a detectable level of antibodies. On several occasions, I noticed that farmers would say the veterinary surgeon had vaccinated their livestock, but did not know against which diseases.

Table 8.2 Seroprevalence to FMDV in two groups of livestock; vaccinated and unvaccinated (based on information from the owner regarding vaccination of the livestock against FMDV within the last 2 years)

<i>Owner info.</i>	<u><i>Absence of antibodies</i></u>		<u><i>Presence of antibodies</i></u>		<i>Total</i>
	<i>Number</i>	<i>%</i>	<i>Number</i>	<i>%</i>	
Not vaccinated	784	83.4	156	16.6	940
Vaccinated	10	55.6	8	44.4	18
Total	794	82.9	164	17.1	958

Body condition

Overall, 7.5% of sampled livestock were assessed in poor body condition, 72.4% were average and 20.4% were in good body condition (Table A5.3, Appendix 5). Figure 8.1 illustrates the difference between the species. The mean condition score of males was 2.3 ± 0.07 , females was 2.1 ± 0.04 , although this difference is statistically significant (Kruskal-

Wallis $H=55.3$, $df=1$, ***), it is unlikely to be of biological significance as the scoring system was in 3 categories. Figure 8.2 compares the condition score of males and females.

Fig_chap8_1

Figure 8.1

Figure 8.2

Table 8.3 The association between age and body condition score of cattle. Mean age is given in years with the 95% confidence interval. The difference in condition is significant (Kruskal-Wallis $H = 11.6$, $df=2$, **)

<i>Condition</i>	<i>Sample size</i>	<i>Mean age</i>	<i>Age range</i>	<i>Std. Dev.</i>
Poor	19	6.4 ± 2.4	0.25 – 18.0	5.3
<i>Average</i>	220	3.5 ± 0.4	0.08 – 14.0	2.9
<i>Good</i>	40	4.8 ± 0.9	0.33 – 11.0	2.9

Table 8.4 The association between age and body condition score of small ruminants. Mean age is given with the 95% confidence interval. The difference in condition is significant (Kruskal-Wallis H = 18.7, df=2, ***)

<i>Condition</i>	<i>Sample size</i>	<i>Mean age</i>	<i>Age range</i>	<i>Std. Dev.</i>
Poor	53	4.6 ± 0.8	0.25 – 15.0	2.9
<i>Average</i>	474	3.0 ± 0.2	0.04 – 14.0	2.1
<i>Good</i>	152	3.0 ± 1.4	0.04 – 12.0	2.4

Breed of animal

Of the sampled cattle, 20 (7.2%) were Kazakh breed, 143 (51.2%) Bely galov and 116 (42.6%) local mixed breed. Of the sampled sheep, 47 (8.7%) were Edilbayev, 60 (11.1%) Karakul and 435 (80.2%) local mixed breed. Of the sampled goats, 47 (34.3%) were Angora and 90 (65.7%) local mixed breed. There was no significant difference between breeds with respect to the prevalence of antibodies to FMD virus. As seroprevalence indicates vaccination status rather than infection, it was not expected that there would be a significant difference between breeds, unless certain breeds are considered more valuable.

Gender

The sampled population consisted of 25.4% male (243/958) and 74.6% females (715/958). Figure 8.3 illustrates the sex ratios sampled by species. Seroprevalence was relatively higher among females (18.2%) than among males (14.0%), though not significantly. Vaccination policies do not vary according to the gender of the animal, thus a gender-related seroprevalence was not expected.

Type of ownership

The sampled population consisted of 85.9% privately owned (823/958) and 14.1% collectively owned (135/958) livestock. Figure 8.4 illustrates the proportion of privately owned livestock by species. Collectively owned small ruminants were kept in significantly smaller herds than privately owned small ruminants (Kruskal-Wallis H=7.3, df=1, **); mean herd size for the former was 82 ± 10 (N=123, range 30 – 250), for the latter it was

150 ± 13 (N=556, range 10 – 500). The most likely reason for this is that the managers of farm enterprises split up large flocks for ease of management, whereas private farmers may not be able to afford the extra costs of hiring another shepherd.

Figure 8.3

(fig_chap8_2)

Figure 8.4

Overall, 16.3% of collectively owned animals and 17.3% of privately owned animals were seropositive to FMDV. There was no significant difference between or within the species. It might have been expected that fewer private livestock would have been vaccinated on cost grounds. The fact that there was no difference between collectively owned and privately owned livestock may be an indication that at least some private farmers are vaccinating their livestock, or it may just reflect the fact that during the privatisation process, collective livestock were distributed among the farm workers and thus many 'private' animals were in fact 'collective' a year or two before the study.

Year of sampling

Figure 8.5 shows the proportion of cattle, sheep and goats sampled in 1997 and 1998. The difference in the proportion of these species sampled varies significantly between the years ($\chi^2=9.2$, $df=2$, *). Seroprevalence to FMD was significantly higher ($\chi^2=4.0$, $df=1$, *) in 1998 (19.3% [107/555]) than in 1997 (14.1% [57/403]). The higher seroprevalence in 1998 was unexpected, as these samples were collected in Aktiubinsk and Dzhezkazgan *oblasts*, where there have been no state-run vaccination programs against FMD in recent years. In the village of 'Zhenis', where livestock were sampled both in 1997 and in 1998, there was an increase in seroprevalence from 10.3% (3/29) to 23.2% (19/82), however this difference was not statistically significant.

Systemic disease

Figure 8.6 shows the proportion of cattle, sheep and goats that had clinical signs of systemic disease. The χ^2 test was invalid because of the small sample size, however when sheep and goats were combined into one category, the difference between these small ruminants (2.9% diseased) and cattle (0.4% diseased) was statistically significant (Yates corrected $\chi^2=5.0$, $df=1$, *). Significantly more livestock sampled in 1998 showed signs of systemic disease than those sampled in 1997 (Yates corrected $\chi^2=10.8$, $df=1$, **). Of livestock sampled in 1997, only 0.2% (1/403) showed signs of systemic disease, compared to 3.6% (20/555) in 1998. However, the diseased animals sampled in 1998 (12 sheep and 8 goats) were all from the same herd.

Seroprevalence to FMD among livestock showing signs of systemic disease was higher (19.0%, N=21) than among the healthy livestock (17.1%, N=937). The difference was not statistically significant. Although the results suggest that there is no correlation between systemic disease and FMD status, the lack of correlation could have been caused by the low sample size of diseased animals and clustering of diseased animals.

Figure 8.5

Figure 8.6

(fig_chap8.3)

Herd size

Cattle were kept in significantly smaller herds than small ruminants (Kruskal-Wallis $H=370$, $df=1$, ***). Mean herd size for cattle was 22 ± 3 ($N=279$, range 1 – 10), for small ruminants it was 138 ± 11 ($N=679$, range 10 – 500). Seropositive animals were kept in herds or flocks that were significantly smaller than the seronegative animals (Kruskal-Wallis $H=8.7$, $df=1$, **). Mean herd size for seropositive animals was 90 ± 20 ($N=164$, range 2 – 500), for seronegative animals it was 107 ± 9 ($N=794$, range 1 – 500). When analysed separately, cattle and small ruminants showed no statistically significant difference with regard to herd / flock size, suggesting that the difference is because cattle were kept in smaller herds than small ruminants, and cattle were significantly more likely to be seropositive.

Age

Seropositivity among older animals was significantly higher (Kruskal-Wallis $H=20.4$, $df=1$, ***) than among younger animals (Table 8.5). Figures 8.7 – 8.9 show age-serological profiles, relating age to antibody status. This analysis did not separate out animals under the age of 6 months, in which antibodies are most likely to be maternal, caused by infection or vaccination of the dam. When animals under the age of six months were removed from the analysis, there was still a statistically significant effect of age on the prevalence of vaccination (Kruskal-Wallis $H=9.7$, $df=1$, **). This may be related to two factors; younger animals may not yet have been vaccinated because of their age, and additionally were born after the vaccination programs in Kazakhstan fell apart (from 1992 onwards).

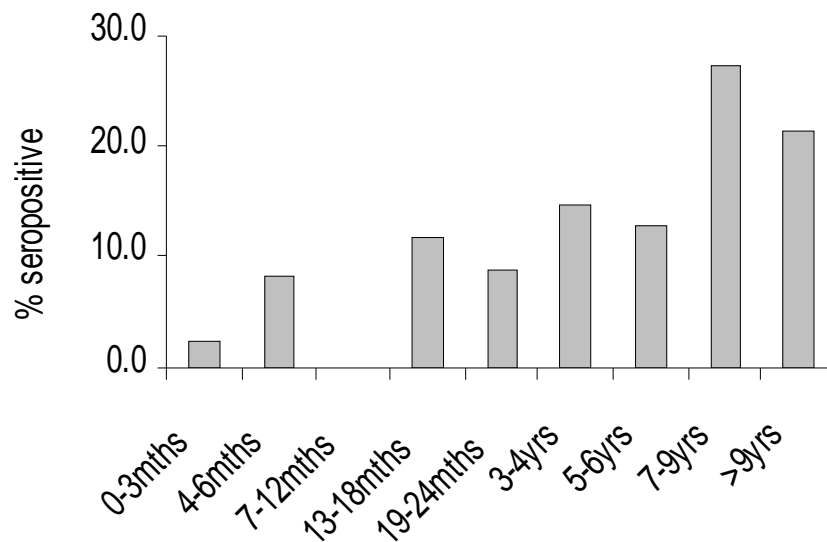
Summary of the factors affecting the seroprevalence to FMDV

Four factors were identified as significantly affecting the prevalence of antibodies against FMD virus, however, when looking at these results it must be taken into account that due to the sampling techniques they could be biased, as was discussed in chapter 5:

- Species: seroprevalence among cattle was significantly higher than among sheep, which was significantly higher than among goats.
- Origin: bought-in livestock were significantly more likely to have antibodies to FMD virus than farm-born livestock were.
- Year of sampling: animals sampled in 1998 were significantly more likely to be seropositive than those sampled in 1997.
- Age: older animals were significantly more likely to be seropositive.

Table 8.5 Seroprevalence to FMDV in livestock by age. Mean age is given in years with the 95% confidence interval. (KW = Kruskal-Wallis H)

	<i>No. sampled</i>	<i>Mean age</i>	<i>Range</i>	<i>Std.Dev</i>	<i>KW</i>	<i>P</i>
<i>Cattle</i>						
Seronegative	198	3.7 ± 0.5	0.08 – 18.0	3.29		
Seropositive	81	4.5 ± 0.7	0.25 – 14.0	3.07	7.0	**
<i>Sheep</i>						
Seronegative	467	3.1 ± 0.2	0.04 – 15.0	2.3		
Seropositive	75	4.0 ± 0.6	0.25 – 14.0	2.7	6.8	**
<i>Goats</i>						
Seronegative	129	2.7 ± 0.3	0.25 – 9.0	2.0		
Seropositive	9	3.0 ± 1.5	1.25 – 8.0	2.3	0.07	NS
<i>Small ruminants</i>						
Seronegative	596	3.0 ± 0.3	0.04 – 15.0	2.23		
Seropositive	83	3.9 ± 0.6	0.25 – 14.0	2.63	7.6	**
<i>All livestock</i>						
Seronegative	794	3.2 ± 0.2	0.04 – 18.0	2.55		
Seropositive	164	4.2 ± 0.4	0.25 – 14.0	2.86	20.4	***



8.2.4 Spatial variation

The effect of oblast and raion

Oblasts and *raions* are administrative areas, with *raions* being sub-divisions of *oblasts*. As veterinary policy varies between *oblasts* and between *raions* within *oblasts*, it was expected that there would be a significant difference in the seroprevalence to FMDV between them.

Figure 8.7 (fig_chap8.4)

Figure 8.8

Figure 8.9

As discussed in chapter 4, the central veterinary committee in each *oblast* has the freedom to decide in which *raions* livestock should be vaccinated, depending on the local circumstances.

The animals sampled in different *oblasts* varied significantly with respect to several of the factors identified as important predictors of FMD status. There was a significant difference between *oblasts* with regard to the number of livestock sampled ($\chi^2=20.5$, $df=5$, ***, Figure 8.10) and breed type. There was also a significant difference between *oblasts* with respect to the proportion of species sampled ($\chi^2=39.6$, $df=10$, ***, Figure 8.11). Figures 8.12 – 8.13 illustrate the proportion of private livestock and the proportion of bought-in livestock, which also varied significantly between *oblasts*. Tables 8.6 and 8.7 show how the mean age of cattle and small ruminants vary significantly between *oblasts*. These differences between the samples are taken into account by carrying out a multivariate analysis of the data, with the key factors included (described in section 8.2.6).

Table 8.6 Mean age of sampled cattle in different *oblasts* (Kruskal-Wallis $H=27.1$, $df=5$, ***). Mean age is given in years with the 95% confidence interval

<i>Oblast</i>	<i>Sample size</i>	<i>Mean age</i>	<i>Range</i>	<i>Std. Dev.</i>
Almaty	12	5.4 ± 1.1	3.0 – 9.0	2
South Kazakhstan	36	7.9 ± 0.9	0.08 – 14.0	2.8
Dzhambul	36	8.3 ± 0.9	0.5 – 11.0	2.9
Dzhezkazgan	151	11.3 ± 0.5	0.08 – 18.0	3.4
Karaganda	35	11.4 ± 1.1	0.33 – 13.0	3.4
Aktiubinsk	9	8.7 ± 1.9	0.17 – 8.0	2.9

Table 8.7 Mean age of sampled small ruminants in different *oblasts* (Kruskal-Wallis $H=19.0$, $df=5$, **). Mean age is given in years with the 95% confidence interval

<i>Oblast</i>	<i>Sample size</i>	<i>Mean age</i>	<i>Range</i>	<i>Std. Dev.</i>
Almaty	8	4.2 ± 1.0	2.0 – 7.0	1.5
South Kazakhstan	79	2.6 ± 0.3	0.08 – 7.0	1.5
Dzhambul	90	2.6 ± 0.4	0.25 – 7.0	1.7
Dzhezkazgan	374	3.2 ± 0.3	0.04 – 15.0	2.7
Karaganda	78	3.7 ± 0.5	0.17 – 11.0	2.1
Aktiubinsk	50	3.1 ± 0.4	0.25 – 4.0	1.4

From the information available on vaccination programs in recent years, one would expect the *oblasts* of Dzhambul and South Kazakhstan to have the highest seroprevalence, because these border to Kyrgyzstan and Uzbekistan and have been targeted for vaccination. In this study, however, *Almaty oblast* had the highest prevalence (30%), although South Kazakhstan *oblast*

Fig_chap8_5

Fig 8.10

Figures 8.11

Figures 8.12 and 8.13

was not far behind (22.6%). What was surprising was the low percent of vaccinated animals in Dzhambul *oblast* – only 11.1%, despite ongoing (official) vaccination programs. The results from Almaty and Aktiubinsk *oblasts* may be affected by the fact that samples were taken from livestock in only one village. Table A5.4 (Appendix 5) demonstrates that there is a greater variation in FMD status between *raions* than there is between *oblasts*, although this is expected, as the *oblast* data are means of the *raion* data. Seroprevalence varied from 0 – 80% between *raions*, and from 7.1 – 30% between *oblasts*. Figure 8.14 illustrates the spatial aspect of the results.

As discussed in section 8.2.1, a much higher proportion of cattle than small ruminants had antibodies to FMDV. The antibody status of cattle varied from 2.9% to 52.8% between *oblasts*, that of sheep varied from 0 to 22.9%, and that of goats from 0 to 10.1% (Table 8.8). A high percent of cattle in Aktiubinsk *oblast* (33.3%) were seropositive, despite the *oblasts* not being officially targeted in FMD vaccination campaigns of recent years. The result is likely to have been confounded by the small sample size (only 9 animals), and the fact that all the cattle were bought-in. The high prevalence of seropositive sheep in Aktiubinsk *oblast* (22.9%) is surprising, but this may again be explained by the fact that

the sheep had been bought-in, rather than born on the farm. In Karaganda *oblast* the proportion of cattle with antibodies was extremely low compared to the others. This may be because Karaganda *oblast* has no international borders, and is therefore not considered to be at risk, however the higher proportion of seropositive sheep is therefore unexpected.

Table 8.8 The seroprevalence to FMD in livestock by *oblast*. % pos = % seropositive

<i>Oblast</i>	<i>Cattle</i>		<i>Sheep</i>		<i>Goats</i>	
	% pos	Sample	% pos	Sample	% pos	Sample
Almaty	50.0	12	0.0	8	-	0
South Kazakhstan	52.8	36	12.3	57	0.0	22
Dzhambul	27.8	36	5.0	60	3.3	30
Dzhezkazgan	27.8	151	15.4	305	10.1	69
Karaganda	2.9	35	10.9	64	0.0	14
Aktiubinsk	33.3	9	22.9	48	0.0	2
Total	29.0	279	13.8	542	5.8	137

The effect of village, farm and owner

Within each *raion* there were differences in seroprevalence between villages, but these differences were not significant. Within each village there were large differences between seroprevalence of livestock belonging to different owners, but a χ^2 test could not be carried out because of the low sample sizes. The variation in seroprevalence between different villages was great, ranging from 0 to 80%. The main source of the variation lies in the cattle, which varied from 0 to 80%. The highest seroprevalence in sheep in any one village

Fig_chap8_6

Fig 8.14

was 22.9%, in goats 14.3%. In several villages, livestock belonging to only one owner were sampled; thus it is difficult to assess whether the observed variation between villages was caused by differences in the villages, or differences between owners. Table A5.5 (Appendix 5) shows the variation between owners.

8.2.5 Comparison with official data

According to official statistics, about 200,000 cattle were vaccinated in 1997, all in Dzhambul and South Kazakhstan *oblasts*. The combined number of cattle in these two *oblasts* in 1997 was 507,700; thus 39.4% of the cattle population in these *oblasts* were

vaccinated. The results from this study showed 40.3% seroprevalence to FMDV from these two *oblasts* combined. This fits well with the information provided by the official data, and is a good indication that the results achieved in this study are reliable, although the lack of independent verification of the official statistics means they cannot completely be relied upon. There was no official plan to vaccinate livestock in any other *oblasts* in 1997, and there are no statistics available for the number of animals vaccinated against FMD in these other *oblasts*. On the expedition in 1998, animals were sampled exclusively from villages in the former Dzhezkazgan *oblast* (currently part of Karaganda *oblast*). This *oblast* was not affected in the 1998 FMD epidemic; it therefore had no officially organised vaccination program. However, we found that 27.8% of cattle and 14.4% of small ruminants were seropositive, indicating that they had been vaccinated.

8.2.6 Modelling a hierarchical structure

A hierarchical model was developed in collaboration with a colleague, Dr C. O’Callaghan (University of Warwick). The data were read into SAS (SAS Institute, Cary, NC, USA) and onward into MLwiN (Multilevel Models Project, Institute of Education, London, UK) and produced a model hierarchy as follows: *oblast*, *raion*, village, farm, owner.

Data distribution

The colinearity and paucity of data at the upper levels of the hierarchy made it extremely difficult to apportion variation over the many different levels of observation. For example, looking at the distribution of *raions* within *oblasts* (Table 8.9), for two *oblasts* only one *raion* was sampled, while in three *oblasts* only two *raions* were sampled. In consequence it is very difficult to estimate the amount of variation between *raions* within *oblasts*.

Further, at these “upper” levels of the hierarchy pyramid, it can be difficult to estimate the variation because of few observations. A similar situation existed with farms and owners; of the 35 farms sampled, on only 7 were samples taken from more than one owner. This made it virtually impossible to estimate the variation between owners within farms.

<i>Oblast</i>	<i>Raion</i>	No. villages	No. farms	No owners
Almaty	Talgar	1	1	2
South Kazakhstan	Otrar	1	1	1
South Kazakhstan	Saryagach	1	1	1
South Kazakhstan	Suzak	3	5	5

Dzhambul	Moinkum	1	1	1
Dzhambul	Sarysu	5	6	11
Dzhezkazgan	Dzhezdin	2	7	17
Dzhezkazgan	Zhana-arkin	2	8	11
Karaganda	Nurin	3	3	3
Karaganda	Tengiz	1	1	1
Aktiubinsk	Chalkar	1	1	1
Total		21	35	54

Model development

Several hierarchical models were developed, using MLwiN software. Initially a three-level (*raion*, farm, animal) variance components model of FMD sero-status was fitted, using a logistic link, and where the animal-level variation was modelled under the binomial assumption, but where the potential for overdispersion was accounted for by fitting an extra-binomial parameter ($k \cdot \Omega_e$). The *raion* (Ω_v) and Farm (Ω_u) level variances were estimated under the assumption of normality. Estimation was by means of Restricted Iterative Generalised Least Squares (RIGLS) using a second-order Taylor Expansion and a Penalised Quasilikelihood (PQL) methodology. Table 8.10 gives the parameter estimates.

In this intercept-only model, the intercept was estimated as -1.642 , the inverse logit transformation of this gave a prevalence estimate of 16.2%. Further, the extra-binomial variance parameter was estimated as 1.004. If the animal-level variance was absolutely binomial, this value would be equal to 1. Since it was virtually equal to 1, it was concluded that there was no binomial overdispersion. Since maximum likelihood methods could not be used to assess whether or not random effect parameters were significant to model fit, linear contrasts were assessed using approximate Wald-based estimates. The *raion*-level variance estimate was tested under the null hypothesis that it was equivalent to 0 (i.e. that there was no significant variation between *raions* when farm and animal-level variance was accounted for). The result showed that there was no significant variation at the *raion*-level in this model ($\chi^2=2.5$, $df=1$, $p=0.1$), therefore this level was removed from the model.

Table 8.10 Parameter estimates for a three-level hierarchical model (*raion*, farm, animal). The intercept represents the mean prevalence of FMD among cattle

<i>Parameter</i>	<i>Estimate</i>	<i>SE</i>
<i>Random effects</i>		
Extra-binomial variance	1.004	0.336

Farm level variance	0.318	0.194
<i>Raion</i> level variance	0.817	0.519
<i>Fixed effects</i>		
Intercept	-1.642	0.336

Another three-level model was developed (*oblast*, farm, animal). In this case, the *oblast*-level variance estimate was less than that observed for the *raion* (above model), and the farm-level variance had increased. The significance of the *oblast*-level variance was assessed, and the result showed that there was even less evidence of significant variation at the *oblast*-level ($\chi^2=0.5$, $df=1$, $p=0.8$). This is not surprising because we already concluded that there was no evidence of significant variation between *raions*. The *oblast*-level was therefore also removed from the model.

Every level above farm in the hierarchy was tested in this way, and all were found to be not significant in a variance components only model. It was therefore deemed appropriate to reduce the model to a two-level model (farm, animal). In this model, the farm-level variance estimate was significant ($\chi^2=7.5$, $df=1$, $p=0.006$), showing significant clustering of responses by farm.

Additional fixed effects were added to this model, which had been shown to be of importance in determining FMD status in the analysis above. These were: species, origin (bought in or farm-bred), sex and age. Unsurprisingly, the estimate for the farm-level variance decreased in magnitude once these effects had been accounted for. Just considering the parameter estimates and their standard errors, it appeared that sheep and goats were less likely to test positive than cattle, that there was no significant difference by gender, that there was a linearly increasing risk with age and that animals which were born on the farm were less likely to test positive than those which were bought-in. These results matched those found in the univariate analyses earlier in the chapter. Despite all of this, there still appeared to be a significant farm-level variance (i.e. a clustering of similar responses by farm).

It may be unreasonable to assume a linear increase in probability of testing positive with age; therefore a quadratic term (age^2) was added and assessed. The outcome was that the linear component increased in magnitude (from 0.1 to 0.3) and that there was a significant negative quadratic effect. This indicated that a combination of linear and quadratic age

terms might be an acceptable functional form over the range of observation. The same age profile was modelled for each species with differing intercept values, i.e. assuming parallel age:seroprevalence relationships. Age:species interaction terms were included and tested for significance, to assess the parallel lines assumption. However, there was no evidence to suggest that, after the difference in intercepts was controlled for, there was any significant difference in the age relationships for any species.

Gender also did not appear to be a significant covariate ($\chi^2=0.98$, $df=1$, $p=0.32$), and was also eliminated from further consideration. The possibility that there could be a significant age profile difference between those animals born on the farm and those purchased was also tested for by means of an interaction term, however there was again no significant difference in the age-profiles ($\chi^2=0.29$, $df=2$, $p=0.87$).

Model results

Parameter estimates for the final, most parsimonious model are listed in Table 8.11. These showed that there was significant farm-level clustering ($\chi^2=4.66$, $df=2$, $p=0.03$). Sheep were significantly less likely to test positive than cattle, irrespective of age ($\chi^2=14.2$, $df=1$, $p<0.001$). Goats were significantly less likely to test positive than cattle, irrespective of age ($\chi^2=14.4$, $df=1$, $p<0.001$). Goats were less likely to test positive than sheep, irrespective of age ($\chi^2=3.6$, $df=1$, $p=0.057$). Animals born on the farm were less likely to test positive than those bought-in, irrespective of age ($\chi^2=7.54$, $df=1$, $p=0.006$). There was a significant linear and quadratic relationship between the probability of a positive test and age, irrespective of species or origin ($\chi^2=15.8$, $df=2$, $p<0.001$).

Table 8.11 Parameter estimates for the most parsimonious hierarchical model, with two levels (farm, animal). Among the fixed effects, the intercept represents the mean prevalence of FMD among cattle, ovine represents the difference in mean prevalence between cattle and sheep, and caprine represents the difference in mean prevalence between cattle and goats. Origin represents the difference in mean prevalence between animals bought in and those born on the farm. The age and age² parameters cannot be interpreted independently, as age gives the linear relationship between prevalence and age (in years) and the quadratic parameter the quadratic relationship between prevalence and age. The tests for significance are Wald-type linear contrast tests, not standard significance tests

<i>Parameter</i>	<i>Parameter estimate</i>	<i>SE</i>	<i>P</i>
<i>Random effects</i>			
Farm level variance	0.372	0.173	0.03
Animal level variance	0.977	0.045	
<i>Fixed effects</i>			
Intercept	-0.452	0.569	
Ovine	-0.816	0.217	<0.001
Caprine	-1.588	0.418	<0.001
Origin	-1.474	0.537	0.006
Age	0.276	0.085	<0.001
age ²	-0.014	0.007	<0.001

Testing the adequacy of the distributional assumptions in the model

The assumptions of binomial distribution / normality of errors were tested by means of the standardised residuals. At the farm-level, positive residuals were those farms with more positive tests than predicted and negative residuals were those farms with fewer positive tests than predicted. The normal-probability plot of standardised *versus* normalised residuals at the farm-level yielded a straight line (Figure 8.15), indicating that the distributional assumptions were being met at the farm-level. The assessment was repeated for the animal-level residuals, demonstrated in Figure 8.16. In this case, the line did not approximate $y = x$, and there were several high positive residuals (i.e. greater than 3). The model over-predicted the number of positive animals. However, from examination of leverage, it was clear there were no values exhibiting undue influences, hence the overall model fit based on analysis of residuals was deemed adequate, such that the tests of significance for the fixed effects observed were considered valid.

Fig_chap8_7

Fig 8.15

Fig 8.16

Thus the multiple variable model confirmed the previous univariate results that important factors affecting FMD status were species, age and the origin of the animal. Factors that were not important in determining FMD status included the type of farm (private or collective). It was found that there was no significant clustering at the *oblast*, *raion* or village levels, but only at the farm level. However, because the data were limited, with a number of farms only having data collected from one owner, it was not possible to discern the farm level from the owner level. Thus we cannot be sure whether the observed clustering was actually at the farm level or at the level of the individual owner. Nonetheless, the results are interesting because in view of the official vaccination policy of

Kazakhstan, one would expect clustering by *oblasts*, with higher seroprevalence in Dzhambul and South Kazakhstan *oblasts*, where there have been ongoing vaccination programs in recent years. Based on these results, it would seem that the most important factor in vaccination status is the individual farmer, who ultimately makes the decision whether to vaccinate his livestock or not. However, the non-random sampling technique may also have influenced the results. Further work using a randomised stratified sampling regime would be useful to validate this result, as the result could well have important policy implications concerning the level at which the government should target any vaccination policies.

8.3 Brucellosis

8.3.1 Prevalence of antibodies to Brucella spp.

In domestic livestock the overall seroprevalence of antibodies to *Brucella* spp. was 2.4% (23/958). Seroprevalence in cattle was 5.4% (15/279), sheep 1.3% (7/542), goats 0.7% (1/137). When combining the data from sheep and goats, seroprevalence in these small ruminants (1.2%) was significantly lower (Yates corrected; $\chi^2 = 13.1$, $df = 1$, ***) than in cattle (5.4%). Unfortunately, it is not possible by serological tests alone to identify whether an animal has been infected with *B. abortus* or *B. melitensis*. In sheep and goats the most likely cause was *B. melitensis*, however in cattle it could be either *B. melitensis* or *B. abortus*, considering the reported problem of *B. melitensis* infection in cattle in Kazakhstan, discussed in chapter 3.

8.3.2 Factors affecting the prevalence of antibodies to Brucella spp.

Origin of animal

83.1% of livestock bought in by their owners were reported as vaccinated against brucellosis, in contrast to only 6.3% of livestock born on the farm. This difference was highly significant ($\chi^2=360$, $df=1$, ***), and was significant when cattle and small ruminants were analysed separately (Table 8.12). However, none of the bought-in animals (cattle or small ruminants) actually had detectable antibodies to *Brucella* spp. It seems likely that the owners were told upon purchase that the animals had been vaccinated against brucellosis, however either they were misinformed, or the vaccines used were routinely ineffective.

Table 8.12 The effect of the origin of the animal on the owners' perception of vaccination status against brucellosis. Among both cattle and small ruminants the difference is statistically significant (cattle: Fisher exact ***; small ruminants: $\chi^2=332$, df=1, ***)

<i>Origin of animal</i>	<i>Cattle</i>		<i>Small ruminants</i>	
	<i>Vaccinated</i>	<i>Sample size</i>	<i>Vaccinated</i>	<i>Sample size</i>
Born on farm	5	258	6.8	629
Bought-in	42.9	21	100	50

Vaccination status

Seroprevalence amongst livestock may reflect either infection or vaccination. The ELISA test cannot differentiate between the two. 5.8% (N=257) of cattle and 1.4% (N=586) of small ruminants thought not to have been vaccinated against brucellosis during the past two years were seropositive, indicating that they had probably experienced infection. Interestingly, all the livestock (22 cattle and 93 small ruminants) which had been vaccinated recently (according to their owners), were seronegative, indicating that they had either not been vaccinated, or the vaccine was ineffective.

Body condition

With respect to body condition, a higher proportion of small ruminants categorised as 'thin' (1.9%, N=53) were seropositive than those categorised as 'average' (1.3%, N=474) or 'good' (0.7%, N=152). In cattle, the proportion seropositive increased as body condition improved, with seroprevalence of 0 (N=19) among animals in 'poor' body condition, 4.5% (N=220) among animals in 'average' body condition and 12.5% (N=40) in 'good' body condition; however the results were not statistically significant.

Breed of animal

There was no significant difference between breeds with respect to the prevalence of antibodies to *Brucella* spp. This was expected, as the organisms are not known to have specific breed predilections.

Gender

There was no significant difference in seroprevalence by gender in small ruminants or cattle, however, a noticeably higher proportion of females (6.1%, N=228) than males (2.0%, N=51) were seropositive. According to the literature, brucellosis is more prevalent in female ruminants (Nicoletti, 1990), as infection most commonly occurs around

parturition, when males are often kept away from the pregnant females. In Kazakhstan many animals are kept in the owner's backyard at night, thus it may not be convenient to keep the males separated. This could be a cause of the difference between males and females being non-significant, otherwise it could be due to the low sample size of males.

Type of ownership

5.6% (N=267) of privately owned cattle had antibodies to *Brucella* spp., whereas seroprevalence among the collectively owned cattle was zero (N=12), but the difference was not statistically significant. In small ruminants the difference between privately and collectively owned animals was small. Privately owned small ruminants were kept in significantly larger flocks than collectively owned small ruminants (discussed in section 8.2.3), and even those kept in small flocks were generally pastured on the common land, thus contacting a large number of animals. This would have increased their risk of infection with brucellosis, however our results showed no evidence for this. The current similarity between the proportion of seropositive animals among private and collective small ruminants presumably reflects the similarity between their husbandry systems, now that collective livestock are no longer owned by the state, and do not receive better health care than privately owned livestock.

Year of sampling

There was very little difference with respect to the year of sampling, however in Zhenis village, sampled both in 1997 and 1998, the proportion of animals with antibodies to *Brucella* spp. was much higher in 1998 than in 1997 (Table 8.13). This was particularly evident in cattle, where 23% were positive in 1998 compared to zero in 1997. The results were not statistically significant, and may result from the small sample size in 1997. Brucellosis is endemic in Kazakhstan, and a great variation in seroprevalence between years was not expected.

Systemic disease

None of the 20 sheep with clinical signs of systemic disease were seropositive, and there was no relationship between the incidence of systemic disease and seroprevalence to brucellosis. Brucellosis generally manifests itself through abortion (Carter & Chengappa, 1991), thus it was not expected that seropositive livestock would have worse health status.

Table 8.13 The effects of the year of sampling on the seroprevalence of antibodies to *Brucella* spp. of livestock in all villages, and in Zhenis village

Year of sampling	Cattle		Small ruminants	
	% positive	Sample size	% positive	Sample size
<i>All villages</i>				
1997	5.5%	128	1.8%	275
1998	5.3%	151	0.7%	404
<i>Zhenis</i>				
1997	0%	9	0%	20
1998	23%	22	2%	60

Herd size

Seropositive livestock were kept in herds or flocks that were smaller than the seronegative animals (Kruskal-Wallis $H=13.3$, $df=1$, ***). Mean herd size for positive animals was 30 ± 11 ($N=23$, range 4 – 100), for seronegative animals it was 106 ± 8 ($N=935$, range 1 – 500). When analysed separately, cattle and small ruminants showed no statistically significant difference with regard to herd / flock size, suggesting that the difference was caused by confounding; cattle were kept in smaller herds than small ruminants, and cattle were significantly more likely to be seropositive.

Age

The proportion of livestock with antibodies to *Brucella* spp. was significantly higher in older animals (Figures 8.17 and 8.18). The most probable cause of this was that older animals have been exposed to the infective agent for longer. From Table 8.14 it is obvious that this effect comes mainly from the cattle. The difference in mean age between seropositive and seronegative animals was greater among cattle, probably because the mean age of cattle was significantly greater than the mean age of small ruminants (Kruskal-Wallis $H=8.2$, $df=1$, **). No cattle in the age groups 4 months to 4 years were positive; this was to be expected as cows are quite resistant to infection before they become pregnant (Nicoletti, 1991) and most of the cows we saw in Kazakhstan reached maturity at about 3 years of age. No sheep or goats under the age of 12 months were positive. Sheep in Kazakhstan breed at 18 months of age (personal observation), thus at this time they are more likely to acquire infection. When animals under the age of six

months (i.e. those that may have had maternal antibodies) were removed from the analysis, seroprevalence among older animals was still significantly higher (Kruskal-Wallis $H=9.0$, $df=1$, **).

Fig_chap8_8

Fig 8.17

Fig 8.18

Table 8.14 Age related prevalence of antibodies to *Brucella* spp. in livestock. Age is in years, with the 95% confidence interval

	<i>Sample size</i>	<i>Mean age</i>	<i>Range</i>	<i>Std. Dev.</i>	<i>KW</i>	<i>p</i>
<i>Cattle</i>						
Seronegative	264	3.8 ± 0.4	0.08 – 18.0	3.23		
Seropositive	15	5.9 ± 1.4	0.08 – 11.0	2.86	7.4	**
<i>Small ruminants</i>						
Seronegative	671	3.1 ± 0.2	0.04 – 15.0	2.29		
Seropositive	8	4.4 ± 2.0	1.25 – 10.0	2.88	1.6	NS
<i>Total</i>						
Seronegative	935	3.3 ± 0.2	0.04 – 18.0	2.61		
Seropositive	23	5.4 ± 1.2	0.08 – 11.0	2.89	12.5	***

Summary of the factors affecting the seroprevalence to brucellosis

Two factors were found to significantly affect seroprevalence to brucellosis:

- Species: seroprevalence among cattle was significantly higher than among small ruminants
- Age: older animals were significantly more likely to be seropositive

8.3.3 *Spatial variation*

The effect of *oblast* and *raion*

The proportion of all livestock with antibodies to *Brucella* spp. varied from 0 to 6.2% between *oblasts*. In cattle the variation was greater (0 to 14.3%) than in small ruminants (0 to 2.6%) as detailed in Table 8.15.

Table 8.15 The occurrence of antibodies to *Brucella* spp. by *oblast*

<i>Oblast</i>	<u><i>Cattle</i></u>		<u><i>Small ruminants</i></u>	
	<i>% seropositive</i>	<i>Sample size</i>	<i>% seropositive</i>	<i>Sample size</i>
Almaty	0	12	0.0	8
South Kazakhstan	2.8	36	1.3	79
Dzhambul	2.8	36	2.2	90
Dzhezkazgan	5.3	151	0.8	374
Karaganda	14.3	35	2.6	78
Aktiubinsk	0	9	0.0	50
Total	5.4	279	1.2	679

Figures 8.19 and 8.20 illustrate the spatial aspect. Between *raions* the proportion varied from 0 to 6.8% (Table A5.6, Appendix 5). Cattle varied from 0 to 20%, and small ruminants varied from 0 to 6.7% between *raions*.

The effect of village, farm and owner

The proportion of all livestock with antibodies to *Brucella* spp. varied from 0 to 6.2% between village. The variation was greater among cattle (0 to 30%) than among small ruminants (0 to 20%). The variation between different villages seemed greater than that

Fig_chap8_9

Fig 8.19

Fig 8.20

between different *raions*, although this was expected, as the *raion* data are means of the village data. It seems that the main place of variation in seroprevalence may be at the level of the village (except in Karaganda, where all four sampled villages had seropositives). Nine of the 21 sampled villages had seropositive animals. In several villages, livestock belonging to only one owner were sampled; thus it was difficult to assess whether the observed variation between villages was caused by differences in the villages, or differences between owners. Table A5.7 (Appendix 5) shows the seroprevalence of livestock by owner. The results suggested significant variability between units at each of the spatial scales, but hierarchical modelling could not be done because the number of seropositives was too low.

8.3.4 Comparison with official data

Livestock were tested in 11 different *raions*; for 7 of these, official data were obtained from the regional veterinary committee for the relevant year. In three *raions* (Sarysu, Dzhezdin and Chalkar), seroprevalence in this study was lower than the official data suggested, but not significantly (Table 8.16). In Suzak *raion*, the seroprevalence was equal (both zero). In Moinkum *raion*, no cattle were sampled in our study, thus no comparison was possible. Thus the results obtained from this serological study of cattle fit quite well with the official data in these *raions*. In Zhana-arkin and Nurin *raions*, seroprevalence in our study was significantly higher than the official data suggested. It is possible that livestock in villages that we did not sample had a very low prevalence of antibodies to *Brucella* spp., and that we by chance sampled the farms with higher seroprevalence. Alternatively, there may be a problem with the official data in these *raions*; thus they should be re-checked.

In Dzhezdin *raion*, there had been no serological testing of small ruminants since before 1995, thus there were no results with which to compare. In Sarysu *raion*, no small ruminants were tested in 1997, therefore the official data from 1996 were used. Among small ruminants, seroprevalence in this study was the same or consistently higher in all *raions* than the official data suggested (with the exception of Moinkum), however only in one *raion* (Zhana-arkin) was the difference statistically significant (Table 8.17). The

results obtained from this serological study of small ruminants thus fit quite well with the official data. However, due to our low sample sizes in several *raions* and the entailing lack of statistical ‘power’ (Thompson *et al.*, 2000) it is possible we could have missed significant effects. It must be borne in mind that this study may have produced biased results due to the non-random selection of farms and herds, and that any such biases are unquantifiable. In addition, no independent studies of brucellosis had been performed in Kazakhstan with which to compare the official data, which, as discussed in chapter 4, are likely to be biased towards underestimating the real prevalence of brucellosis.

Table 8.16 Comparison of the results from this study with official data for brucellosis prevalence in cattle in different *raions*

Year	Raion	Sample size		% positive		Stat.sig. Fisher exact
		Official data	This study	Official data	This study	
1997	Suzak	327	17	0	0	NS
1998	Moinkum	160	0	0	-	NS
1997	Sarysu	2153	36	0.6	0	NS
1997	Dzhezdin	5700	59	1.1	0	NS
1997	Zhana-arkin	5400	92	0.5	8.7	***
1997	Nurin	21700	25	3.8	20.0	**
1998	Chalkar	556	9	4.1	0	NS

Table 8.17 Comparison of the results from this study with official data for brucellosis prevalence in small ruminants in different *raions*. ^ indicates that the official data is for 1996, the year before this study took place

Year	Raion	Sample size		% positive		Fisher exact
		Official data	This study	Official data	This study	
1997	Suzak	94	70	0	1.4	NS
1998	Moinkum	1203	10	0.7	0	NS
1997 [^]	Sarysu	2328	79	1.1	1.3	NS
1997	Dzhezdin	No testing	203	-	0	
1997	Zhana-arkin	1000	171	0	1.8	**
1997	Nurin	3500	63	1.5	1.6	NS
1998	Chalkar	100	50	0	0	NS

8.4 Observations on animal health and vaccination schemes in villages

The veterinary surgeon or animal technician, if there was one present, was interviewed on all farms. A pre-written questionnaire (Appendix 3) was used as a base for this semi-structured interview. On farms where no veterinary surgeon or animal technician was

present it was not possible to gather any information on animal health or vaccinations used. These interviews allowed comparisons between official policy, reported policy at the farm level and the results of the serological survey carried out on these farms. This provided qualitative insights into the spatial scale at which variation in vaccination policy occurred, to complement the statistical analyses performed in sections 8.2 and 8.3 above.

8.4.1 Aktiubinsk oblast

In the village of Zhanakonys, vaccines against FMD were ‘used a long time ago’. A vaccine against anthrax and rabies was provided by the state up until two years before the study (1995), but since then no vaccines had been provided free of charge by the state. There was therefore no vaccination ‘policy’, although most farmers vaccinated against anthrax. The veterinary surgeon did not know which other vaccines, if any, were used by individual farmers. No anthelmintics were used, as farmers could not afford them.

8.4.2 Karaganda oblast

Tengiz raion

In the village of Tkenekta, vaccination against brucellosis ceased in 1987. Vaccines against sheep pox and anthrax were provided by the state free of charge for yearly vaccinations. They vaccinated all sheep against sheep pox in 1996 and 1997. No drugs had been used since 1995, as they could not afford to pay for them, however the farm enterprise had experienced no increase in mortality of sheep or horses during the last 5 years.

Nurin raion

In the village of Amantau, all yearling sheep were vaccinated against brucellosis between 1992 and 1996. At the time of the study, vaccines against anthrax, sheep pox, pasteurellosis and listeriosis were provided by the state free of charge every year.

In the village of Arshelinski, vaccines against anthrax, blackleg (caused by the bacterium *Clostridium chauvoei*), sheep pox and tuberculosis were provided free of charge by the state. All collective sheep were vaccinated annually against brucellosis until 1991. In 1992 a serological survey of sheep revealed 35 sheep seropositive to brucellosis, but these were not clinically ill. Despite this, vaccination has not been resumed due to the costs involved.

In Taldesay village, all female yearling sheep were vaccinated against brucellosis until 1995, when the vaccines were no longer provided free of charge. In 1996 and 1997 they vaccinated against anthrax, blackleg and sheep pox, as these were provided by the state. Five sheep were diagnosed with brucellosis in 1995, none in 1996 or 1997.

8.4.3 South Kazakhstan oblast

Saryagach raion

On the peasant farm visited in Shardarinski village, all livestock were vaccinated against anthrax; additionally cattle were vaccinated against brucellosis at the age of 7-8 months. All sampled livestock (only cattle were sampled) were negative to brucellosis. Half of them had serological evidence of FMD infection, and 80% had evidence of FMD vaccination. The cattle had all been bought in from a farm in a nearby *raion* (Pakhtaral *raion*) where no outbreaks of FMD have been recorded since 1988. This indicates that not all outbreaks of FMD are officially recorded. However, it is possible that the cattle had been infected in the 1988 FMD outbreak in Pakhtaral *raion*, although the owner believed the cattle to be 6 years of age and thus not born until 1991.

Otrar raion

The farm at Akkum received vaccines against anthrax, brucellosis, FMD, TB and rabies free of charge up to and including the year of the study, 1997. Other vaccines used routinely, until 1996, were against blackleg, pasteurellosis, listeriosis and salmonellosis. Brucellosis vaccines had been used for over 10 years. In 1996, three sheep (out of 1,000) were diagnosed with brucellosis, and were slaughtered immediately. In 1997, 200 private sheep were tested; all were free of brucellosis. There had been outbreaks of FMD at this farm recently (see section 8.4.7 for details).

Suzak raion

On Chu farm, all livestock were vaccinated against anthrax, collective cattle against FMD and collective sheep against sheep pox. All female yearlings (sheep and cattle) and collectively owned breeding males were vaccinated against brucellosis every year. There had not been a case of brucellosis for five years. Anthelmintics were used regularly, but antibiotics were very expensive and were only used when deemed essential. Farmers paid the veterinary surgeon about 100 *tenge* (USD 0.7) per animal vaccinated, but the vaccines were free from the *raion* veterinary centre.

At Suzak farm, all livestock were vaccinated against anthrax, and female yearling sheep were vaccinated against brucellosis. There had been no cases of brucellosis in recent years, neither did this study find any animals seropositive to brucellosis. Although located in South Kazakhstan *oblast*, which was targeted in the FMD control program, the veterinary surgeon and owner of the sampled livestock stated that FMD vaccines were not used, and that the sampled livestock had not been vaccinated against FMD. However, 20.0% of cattle and 6.7% of sheep did have antibodies to FMD.

At the farm Zhuantobe, all livestock were vaccinated against anthrax, all sheep were vaccinated against sheep pox, and all one year old sheep against brucellosis. The vaccines were provided by the state; the farmers could not afford to buy any.

8.4.4 Dzhambul oblast

Sarysu raion

According to the veterinary surgeon in charge of the veterinary committee of Sarysuraion, the state provided vaccines against the following diseases free of charge to all farms in the region; anthrax, brucellosis, foot-and-mouth disease, sheep pox, blackleg, leptospirosis, and rabies. Privately owned cattle were usually not vaccinated against brucellosis. Collective cattle were vaccinated against brucellosis at 5-6 months, after which breeding animals were vaccinated every year. They were tested annually before vaccination according to the “plan” which was decided centrally. Breeding-sheep were tested just before vaccination against brucellosis, then again after 3 weeks and after 8 months to check the antibody titre.

The veterinary surgeon at Chiganak farm informed me that the farm had not received vaccines against brucellosis or FMD during the previous 3 years, and had therefore stopped vaccinating. Prior to 1994, livestock were routinely vaccinated against brucellosis. The year of the study (1997) there had been 300 abortions among sheep, but no causal agent was identified.

The state provided anthrax, sheep pox and rabies vaccines to Kamkaly farm. Until 1992 livestock were regularly vaccinated against brucellosis and FMD. The veterinary surgeon insisted that farmers would buy these and other vaccines if they could afford it. Occasionally they would buy antibiotics, but these were expensive.

On Kalinina farm, only collective livestock were vaccinated; against FMD (all cattle once yearly), brucellosis (breeding cows and ewes) and sheep pox (all collective sheep). Brucellosis had not been diagnosed on the farm since 1986, and there had been no outbreaks of FMD during the same period.

8.4.5 Dzhezkazgan oblast

According to the veterinary committee in Dzhezkazgan *oblast*, a total of 4,600 cows, but no sheep, were vaccinated against brucellosis in their *oblast* in 1997. In the spring of 1997, vaccines against anthrax were provided by the *raion* centres to farms, such that all cattle, sheep and horses in the *oblast* could be vaccinated. In Dzhezdin *raion* no private livestock were tested for brucellosis in 1997 due to a lack of funding. State veterinary surgeons at the veterinary committee in the *oblast* centre were interviewed in 1998 (after Dzhezkazgan became part of Karaganda *oblast*). According to them, the official vaccination policy for brucellosis was that female calves were vaccinated at 3 – 5 months of age with strain 19, then again at 10 months of age with strain 82, then before the first calving and every 2 years thereafter with strain 82.

Zhana-arkin raion

In the village of Druzhba, livestock were routinely vaccinated against anthrax, but farmers had no money to buy other vaccines. Cases of anthrax and TB had been seen sporadically, but brucellosis and FMD had not been diagnosed in recent years. Farmers found it was difficult to get hold of medicines and anthelmintics and even when they were available they were prohibitively expensive.

In the village of Zhenis, the veterinary surgeon stated that all livestock were vaccinated against anthrax and black leg. Sheep were vaccinated against brucellosis at the age of 6 months. Prior to vaccination they were blood-sampled to ensure they were brucellosis-free. Cattle were tested for brucellosis every year, but they were not vaccinated. FMD vaccines had not been used since 1993.

In the village of Sarysuskii, livestock were vaccinated against anthrax and blackleg. Farmers would treat a good breeding male (and occasionally a sick cow, if she was

valuable) with antibiotics, but other animals were slaughtered. Farmers paid the veterinary surgeon about 40 *tenge* (USD 0.3) per animal vaccinated, but the vaccines were free from the *raion* veterinary centre. Farmers had to pay themselves if they wanted their livestock tested for brucellosis.

8.4.6 Almaty oblast

Talgar raion

Luchivastok farm kept a dairy herd. According to the veterinary surgeon all livestock were vaccinated against anthrax and blackleg every year. Sheep were also vaccinated against salmonellosis. Collectively owned cattle were vaccinated against brucellosis at 6 months of age, and heifers were vaccinated again at 12 months. Sheep were not vaccinated against brucellosis, but all livestock were bled twice yearly and tested for brucellosis. Privately owned animals were vaccinated only if the owner paid.

8.4.7 Action in the case of an FMD outbreak

In 1991 there was an outbreak of FMD at Akkum farm (Otrar *raion*, South Kazakhstan *oblast*), and more recently, there was an outbreak in October/November 1996, which lasted about one month. Clinical disease was observed in about 30 – 40% of livestock (cattle, sheep and camels). It was first noticed in cattle; the farmer believed the outbreak originated from an infected cow brought onto the farm. All affected animals were treated with antibiotics, the premises were disinfected and the entire farm quarantined for 24 days. All animals were re-vaccinated against FMD (prior to the outbreak, all livestock, private and collective including camels, had been vaccinated annually). Since the outbreak, all livestock have been vaccinated every 6 months. In 1997 no vaccines were bought, as they could not afford them.

In August 1998, an outbreak of FMD occurred among privately owned cattle in the village of Uzunagach (Dzhambul *raion*, Almaty *oblast*). I was able to observe directly the actions taken to control the outbreak. The entire village was quarantined for 34 days. Both roads leading into the village had roadblocks set up, manned by “veterinary police officers”. At the roadblocks a long, shallow pit had been constructed and filled with disinfectant to prevent virus being further spread, e.g. by the tyres of cars.

Sixteen cattle (32%) of the population of 53 in the village exhibited clinical symptoms of FMD. Samples of blood and vesicular epithelium from a cow exhibiting clinical symptoms of FMD were obtained, however it was not possible to conduct any interviews or sample any other animals without official permission. I was informed that the animals were being treated with antibiotics, and that all ruminant livestock in the village had been vaccinated with a bivalent (type A and O) FMD vaccine. No affected animals were slaughtered. Samples sent to the FMD laboratory at *KazNIVI* were analysed for the presence of virus, where FMD serotype O was diagnosed by CFT. Samples were also sent to the IAH, where live virus was not isolated, but analysis by PCR revealed antigen from FMDV serotype A, which was given the name *A/Kazakhstan/2/98*. The type A PCR gave a band, this cDNA fragment was sequenced. Phylogenetic analysis of part of this sequence revealed that the *A/Kazakhstan/2/98* virus is closely related to FMD type A viruses which occurred in the former USSR between 1963 and 1993 (approximately 5% different). Figure 8.21 shows this genetic relationship, and the virus' distant relationship (18.5% different) with the new Iranian virus strain. The fact that two different serotypes were identified from the same sample was a cause for concern, indicating that the laboratory techniques for identification of FMDV in Kazakhstan were not entirely reliable, or it is possible that the infection was mixed.

Figure 8.21 Genetic relationship between *A/Kazakhstan/2/98*, viruses isolated from the former USSR, and the new Iranian virus strain (World Reference Centre for Foot-and-Mouth disease).

8.4.8 Discussion

From Table 8.18 it is clear that there was a large difference in vaccination policies between villages. The major findings of the survey are listed as follows:

- In 8 (47%) of the 17 villages discussed above, the general vaccination policy had changed since independence because farmers were no longer provided with all vaccines from the state.
- The information provided by local veterinary surgeons did not correspond well with the information obtained from state veterinary surgeons in the *raion* centres.
- Livestock owned by peasant farmers or by villagers on a farm enterprise were rarely vaccinated against any disease other than anthrax, because the owner had to pay for vaccination.
- In 16 (94%) of the villages visited, livestock were thought by their owners not to have been vaccinated, yet a proportion of the livestock had vaccinally-induced antibodies to FMDV. Thus many veterinary surgeons and livestock owners seemed unaware of the immune status of their animals.
- In 9 (53%) of the villages, serological evidence to brucellosis was found. This was not unexpected, as the villagers themselves were aware that brucellosis was endemic in the area.

- Most veterinary surgeons were under the impression that farmers would like to buy vaccines and antibiotics for their livestock, but could not afford to do so.
- Few farmers used anthelmintics, because they were too expensive.
- In 6 (35%) of the villages, brucellosis had been diagnosed in humans.
- Outbreaks of FMD in the villages visited had been controlled by quarantine and on one farm by disinfection of the affected premises. In the village I visited there was no sign of disinfection within the village, only at the roadblocks. Affected animals were treated, not slaughtered.
- The fact that two laboratories identified different FMD serotypes from the same sample was a cause for concern, indicating that the laboratory techniques for identification of FMDV in Kazakhstan may not be entirely reliable.

This survey has highlighted the fact that vaccination against diseases of major economic and public health importance are no longer being performed in many parts of rural Kazakhstan. The reason seems to be lack of funding. This could lead to epidemics of major diseases, which may constitute a serious problem for Kazakhstan's future economy.

Fig_chap8.9d

The situation is likely to worsen in the next few years as the proportion of unvaccinated livestock increases. Brucellosis is a disease of particular concern as it is a serious health problem for humans. A number of the villages reported cases of brucellosis in humans, which could well be linked to recent reductions in livestock vaccination.

8.5 Summary

In this chapter I have analysed data on the seroprevalence of the two focal diseases in our study (FMD and brucellosis) in livestock. I used univariate statistics to assess the significance of individual factors, and for FMD used a hierarchical modelling approach to assess the level at which the most variability in seroprevalence could be explained. The results were compared to those found in the literature and in the official statistics for Kazakhstan, and any differences were discussed. The analysis showed that:

- Seroprevalence to both FMDV and brucellosis was significantly higher among cattle than among small ruminants, and among older than younger livestock.
- Bought-in livestock and livestock sampled in 1998 were significantly more likely to have antibodies to FMDV.

- There was no significant clustering of FMD seropositives at the *oblast*, *raion* or village levels, only at the farm level.
- The results for FMD fitted well with the information provided by the official data. For brucellosis, seroprevalence in some *raions* was similar to the official figures, but in other *raions* the official seroprevalence was significantly lower.

Chapter 9

Results from the serological survey of livestock – bluetongue, epizootic haemorrhagic disease, peste-des-petites-ruminants and rinderpest

9.1 Introduction

This chapter details the results of the serological surveys for antibodies against bluetongue, epizootic haemorrhagic disease, peste-des-petites-ruminants and rinderpest viruses. Factors important for the epidemiology of the diseases were identified, and then key factors were chosen for hierarchical modelling.

9.2 Bluetongue

9.2.1 The prevalence of antibodies to bluetongue

In domestic livestock the overall seroprevalence of bluetongue was 23.2% (222/958). Seroprevalence in cattle was 25.4% (71/279), sheep 21.4% (116/542), and goats 25.5% (35/137). Livestock in Kazakhstan were not vaccinated against bluetongue, thus antibodies are likely to be from infection with the virus (or maternal antibodies in youngsters).

9.2.2 Factors affecting the prevalence of antibodies against bluetongue virus

The effect of age

The proportion of livestock with antibodies to bluetongue virus was significantly higher in older animals (Table 9.1), probably because these had been exposed to the infective agent for a longer period of time.

Table 9.1 Age related prevalence of antibodies to bluetongue virus in livestock.

	<i>Sample size</i>	<i>Mean</i>	<i>Range</i>	<i>Std. Dev.</i>	<i>KW</i>	<i>P</i>
<i>age</i>						
<i>Cattle</i>						
Seronegative	208	3.5 ± 0.4	0.08 – 18.0	3.1		
Seropositive	71	5.1 ± 0.8	0.25 – 16.0	3.4	13.9	**
<i>Small ruminants</i>						
Seronegative	528	3.0 ± 0.2	0.04 – 14.0	2.3		
Seropositive	151	3.6 ± 0.4	0.04 – 15.0	2.2	10.4	*
<i>Total</i>						
Seronegative	736	3.1 ± 0.2	0.04 – 18.0	2.6		
Seropositive	222	4.1 ± 0.4	0.04 – 16.0	2.7	24.1	***

The age distribution of seropositive animals was classical, with high seroprevalence in very young animals (due to maternal antibodies), prevalence falling as maternal antibodies wane, then increasing again as animals were exposed to virus (Figures 9.1 – 9.3).

Figchap9_1

Figure 9.1

Figure 9.2

Origin of animal

Seroprevalence was relatively lower among bought-in livestock (15.5%, N=71) than those born on the farm (23.8%, N=887), although not significantly so. Among cattle, 26% (N=258) of farm-born and 19% (N=21) of bought-in animals were seropositive. Among small ruminants, 21.6% (N=629) of farm-born and 14% (N=50) of bought-in were seropositive. As livestock in Kazakhstan were not vaccinated against bluetongue, there was no reason to expect bought-in livestock to have higher seroprevalence.

Table 9.2 The relation between body condition and seroprevalence of antibodies to bluetongue virus. Body condition score has a statistically significant effect on seroprevalence; in cattle $\chi^2 = 6.9$, $df = 2$, * in small ruminants $\chi^2 = 9.3$, $df = 2$, **

<i>Condition score</i>	<i>% positive</i>	Cattle		<i>Small ruminants</i>	
		<i>Sample size</i>	<i>% positive</i>	<i>Sample size</i>	<i>% positive</i>
Poor	11.8	19	34	53	
Average	24.1	220	19.2	474	
Good	40	40	27.6	152	

Body condition

Body condition had a statistically significant association with seroprevalence (Figure 9.4). In small ruminants the effect was as expected; seroprevalence among animals in 'poor' body

Fig 9.3

Fig_chap9_1a

Table 9.2

Chap_fig9_2

Fig 9.4

Fig 9.5

condition was higher, probably due to the significant relationship between body condition and age, with older animals generally in poorer condition (discussed in chapter 8). Surprisingly, the effects were opposite in cattle (Table 9.2), with seroprevalence lower among animals in 'poor' body condition and higher among those in 'good' body condition. This may be related to the fact that the mean age of animals classed as in 'poor' or 'good' body condition was higher than that of animals classed in 'average' body condition.

Breed

There was a significant difference between breeds of cattle and sheep with respect to the prevalence of antibodies to bluetongue virus (Figures 9.5 – 9.7). The difference between breeds of goat was not significant. Among both cattle and sheep, seroprevalence was significantly lower among the ‘local mixed breed’. Native breeds of sheep have, in several African countries, been reported to be more resistant to disease than imported European breeds (Daniels *et al.*, 1995). It was thus expected that there might be a breed-related difference in seroprevalence.

Fig 9.6

Fig 9.7

Gender

There was no significant difference in seroprevalence by gender; 21.0% of males and 23.9% of females were seropositive. According to the literature, susceptibility to bluetongue virus does not vary with gender (Verwoerd and Erasmus, 1994; Daniels *et al.*, 1995), hence this result was expected.

Type of ownership

Seroprevalence among the collectively owned cattle was lower (8.3%) than among privately owned cattle (26.2%), however the difference was not statistically significant, and may be an artefact of the low sample size (N=12). Among small ruminants seroprevalence was 25.2% (N=123) among collectively and 21.6% (N=556) among privately owned animals; this difference was also not statistically significant. Any difference between private and collective livestock might have been caused by the proximity of the pasture to water, as the midges need water to survive (Sellers, 1991).

Year of sampling

Sheep sampled in 1997 had a significantly higher seroprevalence than those sampled in 1998 (Table 9.3), whereas cattle had a significant higher prevalence in 1998. This interesting inter-species difference in seroprevalence between the years 1997 and 1998 may reflect differing demography, including a difference in the abundance of the vector.

Table 9.3 The effects of year of sampling on the seroprevalence of antibodies to bluetongue virus in cattle and small ruminants. ? is Yates corrected, df=1

Year	Cattle				Small ruminants			
	% positive	Sample size	?	p	% positive	Sample size	?	p
All farms								
1997	19.5	128			24.7	275		

1998	30.5	151	12.7	***	20.5	404	16.1	***
<i>Zhenis</i>								
1997	55.6	9			80	20		
1998	36.4	22	0.34	NS	11.7	60	30.9	***

In the village of ‘Zhenis’, both cattle and small ruminants sampled in 1997 had a higher seroprevalence than those sampled in 1998, however the difference was significant only among sheep, possibly due to the low sample size of cattle in ‘Zhenis’ village. The difference may reflect the fact that the animals sampled in 1997 were at their summer pastures in Betpak-dala, which could be located in more mosquito-ridden areas than the common pasture land around in the village where sampling took place in 1998.

Systemic disease

Seroprevalence to bluetongue was higher in animals with clinical signs of systemic disease (28.6%) than in healthy animals (23.1%), however, the difference was not statistically significant. There were no cattle with signs of systemic disease. When small ruminants were analysed separately, the difference between sick (30% seropositive, N= 20) and healthy (22% seropositive, N= 659) animals was still not significant, but this may have been due to the small sample of sick animals.

Herd size

Seropositive livestock were kept in herds or flocks that were larger than the seronegative animals, but the difference was not statistically significant. Mean herd size for seropositive animals was 112 ± 19 (N=22, range 3 – 500), for seronegative animals it was 102 ± 9 (N=736, range 1 – 500). Seroprevalence was higher among both cattle and small ruminants kept in larger herds / flocks, but when analysed separately, they showed no statistically significant difference with regard to herd / flock size.

Summary

- The proportion of livestock with antibodies to bluetongue virus was significantly higher in older animals, probably because these had been exposed to the infective agent for a longer period of time.
- Among both cattle and sheep there was a breed-related difference in seroprevalence; it was significantly lower among the ‘local mixed breed’.

- Small ruminants sampled in 1997 had a significantly higher seroprevalence than those sampled in 1998. The difference in seroprevalence between the years 1997 and 1998 may reflect a yearly difference in the abundance of the vector, or the location where the animals were sampled.

9.2.3 Spatial variation

The effect of oblast and raion

Seroprevalence among livestock varied significantly between *oblasts* ($\chi^2=36.2$, $df=5$, ***); from 5 – 43.5% (Figure 9.8). It was highest in South Kazakhstan; the most southern of the *oblasts* surveyed. In cattle seroprevalence varied from 8.3% to 54.3%; the highest was in Karaganda *oblast*, the furthest north of the *oblasts* surveyed (Table 9.4). The seroprevalence among small ruminants varied from 0 – 53.2% between *oblasts*. It was highest in South Kazakhstan; the most southern of the *oblasts* surveyed. Although there was a significant difference in seroprevalence between *oblasts*, seropositives were found in every *oblast*, indicating that bluetongue is widespread throughout Kazakhstan.

Table 9.4 Prevalence of antibodies to bluetongue virus by *oblast*. For small ruminants the difference between *oblasts* is statistically significant ($\chi^2 = 54.7$, $df = 5$, ***), for cattle the difference is not statistically significant

<i>Oblast</i>	<i>Cattle</i>		<i>Small ruminants</i>	
	% positive	Sample size	% positive	Sample size
<i>Almaty</i>	8.3	12	0	8
<i>South Kazakhstan</i>	22.2	36	53.2	79
<i>Dzhambul</i>	36.1	36	17.8	90
<i>Dzhezkazgan</i>	19.2	151	20.6	374
<i>Karaganda</i>	54.3	35	11.5	78
<i>Aktiubinsk</i>	11.1	9	14	50

Fig_chap9_3

Figure 9.8

Table 9.4

Fig_chap9_4

Figure 9.9

Seroprevalence also varied significantly between *raions* ($\chi^2=43.9$, $df=10$, ***); from 0 to 61%. (Table A5.8, Appendix 5). It was highest in the three *raions* Otrar, Suzak and Saryagach, all in South Kazakhstan (Figure 9.9). In cattle, the difference between *raions* was significant ($\chi^2 = 24.1$, $df = 9$, **), and varied from 0 to 56.0%; the highest was in Nurin and Tengiz, both in Karaganda *oblast*. Among small ruminants, the difference between *raions* in seroprevalence was highly significant ($\chi^2 = 70.2$, $df = 9$, ***), varying from 0 – 100 % between *raions*. Seroprevalence was highest in Otrar and Suzak *raions*.

The effect of village, farm and owner

Seroprevalence among livestock overall varied from 0 to 61% between villages. The variation between different villages was greater than that between different *raions*, but this was expected as the values for the *raions* were the means of the values for the villages. In several villages, livestock belonging to only one owner were sampled; thus it was difficult to assess whether the observed variation between villages was caused by differences in the villages, or differences between owners. Table A5.9 (Appendix 5) shows the seroprevalence of livestock belonging to different owners.

9.2.4 Comparison with official data

At the time of writing, bluetongue had not been reported in Kazakhstan, there were thus no data available on seroprevalence. In areas where bluetongue is known to be present, seroprevalence of 46 – 52% in sheep, 44% in goats, 33 – 95% in cattle, 56% in wild African herbivores, 20% in water buffalo and 13% in bighorn sheep has been reported (Clark *et al.*, 1993; Formenty *et al.*, 1994; Sreenivasulu & Rao, 1999).

None of the midges (*Culicoides* spp.) identified as vectors of bluetongue are known to exist in Kazakhstan, however, other *Culicoides* spp. do exist there. It is possible that these species are responsible for transmitting bluetongue virus, otherwise it may be that *C. imicola* has extended its range from the Middle East into central Asia. Bluetongue has been reported in some parts of Russia (Vishnyakov *et al.*, 1994), thus Soviet scientists have been aware of the existence of the disease. However, if bluetongue is endemic, it may cause only sporadic deaths, which could easily be attributed to other

causes, e.g. pneumonia. State veterinary surgeons seemed to believe the disease did not exist in Kazakhstan. It may simply be unidentified, never having caused a major disease outbreak.

9.2.5 Comparison with saigas

Seroprevalence of bluetongue was 23.2% in domestic livestock, and zero in saigas. This is very interesting, as many species of wild ungulates have been reported seropositive to bluetongue virus (discussed in chapter 3). It is possible that the ELISA test developed to detect antibodies in domestic livestock did not manage to detect antibodies in saigas, however this seems strange as other authors have used the same test in wild ungulates and reported positive results. It was expected that saigas would have a similar seroprevalence as domestic livestock, having been exposed to the vector to a similar extent. There was no statistically significant difference between seroprevalence among livestock sampled within and outside the saiga range area. Many of the livestock were grazing in areas where saigas were known to graze, and one would assume that the midges would bite saigas as well as domestic livestock when these are at nearby pastures. One explanation is host preference by the insects; it is possible that they are not so attracted to saigas. In South Africa, farmers usually include cattle in their herds of sheep and goats to "distract" the vectors of bluetongue virus from the small ruminants (J.Anderson, pers.comm). It is also possible that in the few cases where they are infected, they suffer high mortality, such that there are very few alive with antibodies. A sample size of 297 would be required to be 95% confident that disease was not present at or below a prevalence of 1% in a population of 300,000 animals (see chapter 5). This study sampled 531 saigas in Betpak-dala and 620 saigas in Ustiurt; thus we can say with <95% confidence that if bluetongue had been present in the Betpak-dala or Ustiurt saiga population, we would have found it.

9.2.6 Hierarchical model

Model development

The model for bluetongue was developed in collaboration with Dr Chris O'Callaghan in the same way as the hierarchical model for FMD (described in chapter 8), thus the same caveats apply with respect to the distribution of the data. As for the FMD model, every level above farm in the hierarchy was tested and all were found not to be significant in a variance components only model. The model was therefore reduced to a two-level model

(farm, animal). In this model, the farm-level variance estimate was significant ($\chi^2=9.13$, $df=1$, $p=0.003$), showing significant clustering of responses by farm.

Additional fixed effects (as for the FMD model) were added to this model (species, origin, sex, age and age²). Parameter estimates are shown in Table 9.5. There were no significant differences between species, gender, or whether the animal was bought or born on the farm. Only age and age² parameters contributed significantly to model fit. These results match those found in the univariate analyses earlier in the chapter.

Table 9.5 Parameter estimates for the most parsimonious hierarchical model, with two levels (farm, animal). Among the fixed effects, the intercept represents the mean prevalence of bluetongue among cattle, ovine represents the difference in mean prevalence between cattle and sheep, and caprine represents the difference in mean prevalence between cattle and goats. Origin represents the difference in mean prevalence between animals bought in and those born on the farm. The age and age² parameters cannot be interpreted independently, as age gives the linear relationship between prevalence and age (in years) and the quadratic parameter the quadratic relationship between prevalence and age

<i>Parameter</i>	<i>Parameter estimate</i>	<i>SE</i>
<i>Random effects</i>		
Farm level variance	0.804	0.262
Animal level variance	0.958	0.045
<i>Fixed effects</i>		
Intercept	-2.777	0.77
Ovine	-0.163	0.218
Caprine	0.203	0.285
Origin	0.795	0.741
Age	0.348	0.084
age ²	-0.016	0.007

Model results

There was a significant linear and quadratic relationship between the probability of a positive test and age ($\chi^2 = 30.4$, $df = 2$, $p < 0.001$) and there was a significant farm-level clustering ($\chi^2 = 9.3$, $df = 1$, $p = 0.002$) independent of the effect of age.

Testing the adequacy of the distributional assumptions in the model

The normal-probability plot of standardised *versus* normalised residuals at the farm-level yielded the graph in Figure 9.10, which approximated a straight line, indicating that the distributional assumptions seemed to be met at the farm-level. The farm animal level standardised residuals revealed several high positive residuals (i.e. greater than 3, Figure 9.11). However, from examination of leverage it is clear that there were no inordinately large residual values exhibiting undue influence, hence the significant fixed effects observed were probably valid.

Fig_chap9_5

Fig 9.10

Fig 9.11

9.3 Epizootic haemorrhagic disease

9.3.1 Prevalence of antibodies to EHD among domestic livestock

In domestic livestock the overall seroprevalence to EHD virus was 1.0% (10/958). Seroprevalence in cattle was 2.9% (8/279), sheep 0.4% (2/542), and goats 0 (N=137). The difference in prevalence between species was statistically significant (Fisher exact **).

9.3.2 Factors affecting the prevalence of antibodies to EHD virus

Table 9.6 shows the seroprevalence among different breeds of cattle and sheep. Seroprevalence to EHD was significantly higher among the 'Local mixed breed' than among the 'Bely galov' breed of cattle (Fisher exact **). There was no significant difference in seroprevalence among different breeds of sheep. EHD is not a disease that is well researched, but breed-related effects on infection rates have not been reported (J.Anderson, pers.comm.).

5.5% (N=545) of livestock sampled in 1998 were seropositive, compared to zero (N=403) of those sampled in 1997, a statistically significant difference (Fisher exact **). This may be because different farms were sampled. On the only farm to be sampled both years ('Zhenis'), there was no significant difference. Seroprevalence among both cattle and sheep was higher in 1998 than in 1997, but only among cattle was it significantly higher (Fisher exact **, Table 9.7). On 'Zhenis' farm, seroprevalence was also higher in 1998, but the difference was not statistically significant, due to the low sample size. The χ^2 test for condition could not be performed due to the small sample size.

Table 9.6 Seroprevalence to EHD by breed. The difference in seroprevalence between breeds of cattle was significant (Fisher exact **)

	<i>Sample size</i>	<i>Number positive</i>	<i>% positive</i>
<i>Cattle</i>			
Kazakh	20	2	10
Bely galov	143	0	0
Local mixed breed	116	6	5.2
<i>Sheep</i>			
Edilbayev	47	0	0
Karakul	60	0	0
Local mixed breed	433	2	0.5

Table 9.7 The effects of year of sampling on the prevalence of antibodies to EHD virus in cattle and sheep. The difference in seroprevalence between years for all farms is significant in cattle (Fisher exact **), but not in sheep

<i>Year of sampling</i>	<i>Cattle</i>		<i>Sheep</i>	
	<i>% positive</i>	<i>Sample size</i>	<i>% positive</i>	<i>Sample size</i>
<i>All farms</i>				
1997	0	128	0	206
1998	5.3	151	0.6	336
<i>Zhenis</i>				
1997	0	9	0	17
1998	4.5	22	0	56

9.3.3 The effect of age on the seroprevalence

Seroprevalence among older animals was higher than in younger animals, but the difference was not statistically significant, due to the low number of seropositives (Table 9.8).

Table 9.8 Prevalence of antibodies to EHD virus in livestock, by age. Mean age is given in years with the 95% confidence interval. The differences in age are not statistically significant

	<i>Sample size</i>	<i>Mean age</i>	<i>Range</i>	<i>Std. Dev.</i>
<i>Cattle</i>				
Seronegative	271	3.9 ± 0.4	0.08 – 18.0	3.2
Seropositive	8	4.4 ± 2.6	1.0 – 9.0	3.5
<i>Sheep</i>				
Seronegative	540	3.2 ± 0.2	0.04 – 15.0	2.4
Seropositive	2	4.5 ± 1.0	4.0 – 5.0	0.7
<i>Total</i>				
Seronegative	948	3.3 ± 0.2	0.04 – 18.0	2.6
Seropositive	10	4.4 ± 1.9	1.0 – 9.0	3.1

9.3.4 The effect of location

All livestock with antibodies to EHD were located in Dzhezkazgan *oblast*, where seroprevalence was 5.3% (N=151) among cattle and 0.5% (N=374) among small ruminants, however, the difference in seroprevalence between *oblasts* could not be assessed for significance due to the lower sample sizes in the other *oblasts*. The proportion of cattle with antibodies to EHD virus varied from 0 to 8.2% between villages (Table 9.9), whereas the proportion of sheep with antibodies to EHD virus varied from 0 to 2.9%.

Table 9.9 Spatial distribution of EHD virus in Dzhezkazgan *oblast*; seroprevalence of cattle and sheep by villages. The difference in seroprevalence between *raions* and between villages was not statistically significant

Raion	Cattle		Sheep	
	% positive	Sample size	% positive	Sample size
Village				
Dzhezdin	3.4	59	1.3	150
Moibulak	0	6	2.9	70
Saryuskii	3.8	53	0	133
Zhana-arkin	6.5	92	0	155
Druzhba	8.2	61	0	91
Zhenis	3.2	31	0	80

9.3.5 The effect of the owner

The proportion of cattle with antibodies to EHD virus varied from 0 to 22.2% between different owners, whereas the proportion of sheep with antibodies to EHD virus varied from 0 to 8.3% (Table A5.10, Appendix 5).

9.3.6 Summary

- Seroprevalence was significantly higher among cattle than among sheep.
- The ‘local mixed breed’ of cattle had significantly higher seroprevalence than the ‘Bely galov’ breed.
- Seroprevalence among cattle sampled in 1998 was significantly higher than in 1997, possibly related to a yearly variation in the abundance of the vector.
- All seropositive livestock were located in Dzhezkazgan *oblast*, however, the sample size in this *oblast* was much greater than in the others. Within the *oblast*, there was no significant difference between *raions*.

9.4 Peste-des-petites-ruminants

9.4.1 Prevalence of antibodies to PPR among domestic livestock

In domestic livestock the overall seroprevalence to PPR virus was 1.0% (10/958). Seroprevalence in cattle was 2.2% (6/279), sheep 0.6% (3/542), and goats 0.7% (1/137).

The difference between cattle and sheep, but not between cattle and goats, was significant (Fisher exact *).

9.4.2 Factors affecting the prevalence of antibodies to PPR virus

Among cattle, the χ^2 analyses for significance with respect to PPR seroprevalence and breed type were not valid, however, when the cattle breed type 'Kazakh' (of which the sample size was very small, N=20) was removed from the analysis, there was a significant difference between the two remaining breeds 'Bely galov' and 'Local mixed breed' (Fisher exact *). Seroprevalence among the Kazakh breed was 5.0% (N=20), among Bely galov zero (N=143) and among local mixed breed 4.3% (N=116). Among sheep, seropositives were only found among the local mixed breed (0.7%, N=433), but the statistical analysis was invalid because an expected cell value was <5. Among goats, seropositives were also only found among the local mixed breed (1.1%, N=90). However, the difference between this and the Angora breed was not statistically significant, possibly due to the small sample size.

In 1997 seroprevalence was zero (N=403), in 1998 it was 5.5% (N=545). This difference was statistically significant (Fisher exact **), possibly because different farms were sampled. In cattle (Table 9.10) the seroprevalence in 1998 was also significantly higher than in 1997 (Fisher exact *). On 'Zhenis' farm, seroprevalence was higher in 1998 (3.7%, N=81), but the difference was not statistically significant, due to the small sample size.

Table 9.10 The association of year of sampling with the seroprevalence of antibodies to PPR virus in cattle and small ruminants. The difference in seroprevalence between years for all farms was significant in cattle (Fisher exact *), but not in small ruminants

Year of sampling	Cattle		Small ruminants	
	% positive	Sample size	% positive	Sample size
<i>All farms</i>				
1997	0	128	0	275
1998	4.0	151	1.0	404
<i>Zhenis</i>				
1997	0	9	0	20

1998	4.5	22	3.3	60
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9.4.3 The effect of age on the seroprevalence

The proportion of livestock with antibodies to PPR virus was higher in older animals (Table 9.11), but the low number of seropositives means that the difference was not statistically significant.

Table 9.11 Prevalence of antibodies to PPR virus in livestock, by age. Mean age is given in years with the 95% confidence interval. The differences in age are not significant

	<i>Sample size</i>	<i>Mean age</i>	<i>Range</i>	<i>Std. Dev.</i>
<i>Cattle</i>				
Seronegative	273	3.9 ± 0.4	0.08 – 18.0	3.3
Seropositive	6	3.6 ± 1.5	0.5 – 6.0	1.9
<i>Sheep</i>				
Seronegative	675	3.1 ± 0.2	0.04 – 15.0	2.3
Seropositive	4	2.5 ± 2.4	0.33 – 5.0	2.4
<i>Total</i>				
Seronegative	948	3.4 ± 0.2	0.04 – 18.0	2.6
Seropositive	10	3.1 ± 1.3	0.33 – 6.0	2.1

9.4.4 The effect of location

All livestock with antibodies to PPR were sampled in Dzhezkazgan *oblast*, where seroprevalence among cattle was 4.0% and among small ruminants was 1.1%. However, the statistical analysis was invalid due to the lower sample sizes in the other *oblasts*. The proportion of cattle with antibodies to PPR virus varied from 0 to 4.9% between villages (Table 9.12), whereas in sheep it varied from 0 to 2.5%.

Table 9.12. Spatial distribution of PPR virus in Dzhezkazgan *oblast*. Seroprevalence of cattle and small ruminants by villages. The difference in seroprevalence was not statistically significant

<i>Raion</i>	<i>Cattle</i>		<i>Small ruminants</i>	
	<i>% positive</i>	<i>Sample size</i>	<i>% positive</i>	<i>Sample size</i>
<i>Village</i>				
<i>Dzhezdin</i>	3.4	59	0.5	203
<i>Saryuskii</i>	3.8	53	0.8	133
<i>Zhana-arkin</i>	4.3	92	1.8	171
<i>Druzhba</i>	4.9	61	1.1	91
<i>Zhenis</i>	3.2	31	2.5	80

9.4.5 *The effect of owner*

The proportion of cattle with antibodies to PPR virus varied from 0 to 25.0% between different owners (Table A5.11, Appendix 5), whereas the proportion of sheep with antibodies to EHD virus varied from 0 to 3.6%.

9.4.6 *Summary*

- Seroprevalence was significantly higher among cattle than among small ruminants.
- The ‘local mixed breed’ of cattle had significantly higher seroprevalence than the ‘Bely galov’ breed.
- Seroprevalence among cattle sampled in 1998 was significantly higher than among cattle sampled in 1997, possibly because different farms were sampled.
- All seropositive livestock were located in Dzhezkazgan *oblast*, however, the sample size in this *oblast* was much greater than in the others. Within the *oblast*, there was no significant difference between *raions*.

9.5 *Rinderpest*

All samples from domestic livestock were negative for antibodies to rinderpest. This was expected, as rinderpest has not been reported in Kazakhstan.

9.6 *Summary of chapter*

In this chapter, I analysed data on the seroprevalence of several diseases of ungulates that are known to be of importance among livestock. These diseases have not been looked for or reported in Kazakhstan previously, thus the results of this study could be important in highlighting potentially significant diseases for Kazakhstan’s livestock industry. I also analysed the seroprevalence of these diseases among saiga antelopes; all of the diseases tested have previously been found among wild ungulates. The results were:

- Although there was a significant difference in seroprevalence to bluetongue between *oblasts*, seropositives were found among livestock in every *oblast*, indicating that bluetongue is widespread throughout Kazakhstan.
- Among cattle, seroprevalence to bluetongue was highest in Karaganda *oblast*, furthest north of the *oblasts* surveyed. Among small ruminants, seroprevalence to bluetongue was highest in South Kazakhstan *oblast*; the most southern of the *oblasts* surveyed.

Seroprevalence was expected to be higher in the more southern *oblasts*, as *Culicoides* spp. prefer warmer and more humid climates (Sellers, 1991).

- The proportion of livestock with antibodies to bluetongue virus was significantly higher in older animals, probably because these had been exposed to the infective agent for a longer period of time.
- There were significant linear and quadratic relationships between the probability of a positive bluetongue test and the age of the animal, and there was a significant farm-level clustering independent of the effect of age.
- Seroprevalence of bluetongue was 23.2% in domestic livestock, and zero in saigas. This was a highly unexpected result, as it was expected that saigas would have a similar seroprevalence as domestic livestock, having been exposed to the vector to a similar extent.
- Seroprevalence to EHD and PPR was low in livestock overall, and zero in saigas.
- Seroprevalence to EHD and PPR was significantly higher among cattle than among small ruminants, however there was no significant difference between species-type with regard to seroprevalence to bluetongue.
- Among both cattle and sheep there was a breed-related difference in seroprevalence to all viruses; for bluetongue it was significantly lower among the 'local mixed breed', whereas for EHD and PPR it was significantly higher among the 'local mixed breed'.
- Small ruminants sampled in 1997 had significantly higher seroprevalence to bluetongue than those sampled in 1998, however seroprevalence to EHD among cattle sampled in 1998 was significantly higher than among cattle sampled in 1997. Thus the difference in seroprevalence between the years 1997 and 1998 is more likely to reflect the location where the animals were sampled than a yearly difference in the abundance of the vector.
- All livestock seropositive to EHD or PPR were located in Dzhezkazgan *oblast*, however this may have been a chance result as the sample size in this *oblast* was much greater than in the others. Within the *oblast*, there was no significant difference between *raions*.
- None of the animals seropositive to PPR had antibodies to EHD virus, and only one of the bluetongue reactors also had antibodies to PPR virus, thus it is unlikely that the tests were picking up non-specific antibodies.

- All samples from domestic livestock and saiga antelopes were negative for antibodies to rinderpest. This was expected, as rinderpest has not been reported in Kazakhstan.

Chapter 10

A preliminary framework for modelling foot-and-mouth disease transmission among saiga antelopes in Betpak-dala

10.1 Introduction

This chapter discusses the development of mathematical models for the study of FMD virus transmission in the Betpak-dala saiga population. The basic model is expanded to incorporate seasonally varied transmission rates, and the two models are presented and discussed. Parameterisation is discussed, and recommendations are made for the use of the model and for future development.

10.2 Literature review of mathematical models for use in the study of infectious disease

10.2.1 Background

The application of mathematics to the study of infectious disease appears to have been initiated by Daniel Bernoulli in 1760 (Anderson & May, 1991). Over a century later, Hamer (1906) postulated that the course of an epidemic depends on the rate of contact between susceptible and infectious individuals. Developments in the field of epidemiology are reviewed in Anderson & May (1991) and Grenfell & Dobson (1995), the latter being specific to wild animal populations.

10.2.2 Basic concepts

A key measure of the transmissibility of an infectious agent is provided by a parameter termed the basic reproductive rate and denoted by the symbol R_0 . In the simplest models, R_0 is the number of successful offspring or secondary cases that a parasite is maximally capable of producing in a completely susceptible population (May & Anderson, 1979). In mathematical terms, $R_0 = pcD$ where p is the probability of infection, c the contact rate and D the length of time the primary case is infectious to others (Anderson & May, 1991). The basic reproductive rate is of major epidemiological significance since the condition $R_0 = 1$ defines a transmission threshold below which the generation of secondary cases is insufficient to maintain the infection within the population (May & Anderson, 1979).

10.2.3 Compartmental models

For the purpose of modelling infection by microparasites, it makes sense to divide the host population into mutually exclusive classes or compartments of individuals, for example: susceptible, infected, recovered-and-immune (Anderson & May, 1991).

Most models employ the ‘mass action’ principle, which implicitly assumes random homogenous mixing amongst the population or within groups. Despite the simplicity of this notion, the predictions of simple compartmental models based on this assumption often mirror observed epidemiological patterns surprisingly well (Anderson & May, 1982). However, the less able an infection is to spread through a particular population (lower R_0) the more important are slight deviations from homogenous mixing.

10.2.4 Wildlife disease models

Wildlife disease models are developed with a variety of objectives. Most commonly they aim to understand the dynamics of the disease, so as to assess the effects of control measures (e.g. culling or vaccination) or to predict the temporal and/or spatial spread of disease. Accurate quantitative predictions from models are practically impossible, especially for dynamic wildlife host populations (Heesterbeek & Roberts, 1995). However, qualitative predictions are possible, and these are often the most useful, and may be achieved using relatively simple models.

A particular feature of wildlife diseases, compared to diseases of managed populations such as livestock, is that host population size is a dynamic variable. This requires significant modifications of the classical models that are based on constant host population sizes. In many models of wildlife disease each class of animal is, therefore, expressed as a density, rather than in absolute numbers (Anderson *et al.*, 1981; Anderson & Trewhella, 1985; Coyne *et al.*, 1989; Heesterbeek & Roberts, 1995; Smith, 1995; Smith & Harris, 1991). The main reason for using densities is the immediate association with the ‘mass-action’ principle, and that natural mortality is usually assumed to be density-dependent, linked to the carrying capacity of the habitat. In particular, densities must be used in situations when the population cannot expand the area it occupies as its numbers increase.

Anderson *et al.* (1981) used a deterministic compartmental model to study the population dynamics of fox rabies, in which the host population was divided into three classes, susceptibles, latently infecteds (not yet infectious) and infectious. The model uses densities

rather than individual numbers of foxes, and thus the population is affected both by the disease and the density dependent constraints imposed by resource limitation. Coyne (1989) developed a model based on the Anderson *et al.* model, using it to investigate the conditions for endemic maintenance of rabies in a population of racoons. The principal modification to the model was the inclusion of a class of hosts with naturally acquired immunity. As in the Anderson *et al.* model, mortality was considered density dependent and seasonal aspects of reproduction were ignored. Culling and vaccination were compared as control measures, and it was estimated that at least 99% of the population would have to be vaccinated every year, or 32% of the population culled every year to eliminate disease.

Most wildlife models treat both birth and mortality as continuous functions in time, and many fail to take account of the age structure of the population. Starting with a simple model of tuberculosis (TB) in badgers, Anderson & Trewhella (1985) showed how these biological complexities can be added to make the model more realistic. The demographics of the badger population, with its well-defined age structure and seasonal breeding habits, were described in terms of a modified Leslie-matrix model. Age specific mortality rates and fecundity were thus incorporated as a function of density, age and time of year. They also incorporated stochasticity in to the model (important for determining events within small populations of animals and in small areas of badger habitat) by using Monte Carlo methods to examine the impact of demographic stochasticity on temporal changes in badger abundance and disease prevalence. The model predicted levels of disease that broadly matched those observed, and suggested that bovine tuberculosis acts to significantly depress badger density below disease-free levels.

10.2.5 Control measures

The effect of control measures, and comparison between different measures has been modelled by several authors (Barlow, 1996; Coyne *et al.*, 1989; Heesterbeek & Roberts, 1995; Smith, 1995; Smith & Harris, 1991). Rabies in particular, has been very intensely studied, probably because it is zoonotic.

Heesterbeek & Roberts (1995) showed how a deterministic, compartmental model for any wildlife disease could be expanded to incorporate control strategies such as vaccination or chemotherapy, and that comparisons of the efficacy of these control strategies could be

made quite simply by comparing the ratio of the proportion of hosts that must be vaccinated annually, to that which must be treated by chemotherapy. They concluded that it is more difficult to compare the effects of culling and vaccination, as culling establishes a new population size, which may reduce both the host mortality rate (if it is density-dependent) and the contact rate.

Barlow (1996) took a standard deterministic model for an endemic wildlife disease with no recovery, and expressed the sustained rate of control required to eliminate disease in terms of the basic reproductive rate of the disease (R_0). Using the formulae for *per capita* culling rates, sterilisation rates and successful immunisation rates taken from Coyne *et al.* (1989), he compared the effects of vaccination, sterilisation and culling in terms of the sustained rates of control (e.g. percentage per year). He used parameter values for six specific wildlife disease models (Anderson *et al.*, 1981; Anderson & Trehwella, 1985; Pech & Hone, 1988; Pech & McIlroy, 1990; Barlow, 1991) and showed that in all these models culling is theoretically the most effective control strategy (given the author's assumption in the original models).

10.2.6 Mathematical models of brucellosis in wildlife and livestock

The presence of brucellosis infected bison herds in North America is thought to constitute a serious threat to the brucellosis control programs, and for this reason brucellosis has been modelled in wildlife species. Almeida & Louza (1988) developed an age stratified compartmental model using linear equations to explore the spread of brucellosis through a herd of cattle. They investigated alternative control measures such as calf vaccination, herd vaccination, isolation of aborting and calving cows, serological testing and removal of reactors, serological testing and retention of reactors (if herd is fully vaccinated). Parameter rates were adjusted to fit with those observed, and the transmission rate was then adjusted to produce a prevalence rate similar to that observed in reality. Testing herds at four-monthly intervals combined with immediate removal of all reactors was shown to eradicate the disease from the herd after 10 years.

Gonzalez-Guzman & Naulin (1994) constructed a compartmental model of bovine brucellosis spread within a herd of cattle, and analysed it in order to decide the amount of vaccination and the elimination rate of seropositives necessary in order to avoid an abortion storm. Their recommended strategy was elimination of all seropositives.

Peterson *et al.*, (1991) developed an age structured model for a brucellosis-free herd of bison, which accurately simulated historical changes in herd size, annual recruitment and population age structure. The model also accurately simulated historical changes in herd size in a bison herd that contracted brucellosis during a 20-year period. The model was then used to evaluate a proposed bison brucellosis management plan that involves annual vaccination of female calves. Simulations suggested that after 20 years the proportion of the herd infected with brucellosis might be reduced from 69% to between 50% and 20%, depending on the vaccine efficacy. The model thus shows that the suggested vaccination plan is likely to give poor results, as a 20-50% infection rate is still high enough to pose unacceptable risks to neighbouring livestock.

A basic SIR model (Anderson & May, 1979) was used by Dobson & Meagher (1996) to study the dynamics of brucellosis within a single host population of bison. The model assumed vertical transmission in a proportion of calves and included loss of fecundity of infected animals. The dynamics of the model are very stable for the broad range of parameter values that correspond to brucellosis in wild and domestic ungulates. It was most sensitive to the population density at which the herd would equilibrate, the transmission rate and the impact of the pathogen on host fecundity and mortality. The model was extended to include a second species (red deer), with separate transmission rates for intra-species and inter-species transmission. The dynamics of this two-species model was also very stable. The models gave similar levels of prevalence of infected animals as those observed in bison populations. The threshold population size that allowed persistence of brucellosis in a bison population was so low that it would not be ecologically possible to cull the population down to this level. The authors suggest that the best approach to brucellosis control would be to create a buffer zone around the parks with vaccinated livestock.

10.2.7 FMD models

As a highly infectious and economically significant disease, FMD has received a lot of attention from modellers. Several authors have modelled FMD, both in domestic livestock and in wildlife populations. In general there have been three main focuses of FMD modelling:

- economic analyses
- forecasting airborne spread
- predicting the spread of disease between and/or within livestock herds or wildlife populations.

Economic studies

By far the greatest use of modelling in FMD has been for economic studies (Miller, 1979; Carpenter & Thieme, 1980; Thieme, 1983; Berentsen *et al.*, 1992; Dijkhuizen, 1988; Lembit & Fisher, 1993; Jalving *et al.*, 1997; Singh *et al.*, 1997). This is probably because of the fact that FMD is considered by most health authorities to be the most economically significant disease of domestic livestock. Enormous losses have been recorded, like those estimated at £250,000,000 in Britain in 1967 when a major FMD epidemic disrupted the whole economy and required the slaughter of thousands of highly productive animals (Wilson, 1987).

Various approaches have been used to model an epidemic of FMD. Thieme (1983) developed a deterministic simulation model, which he then used to calculate the net value of FMD control programs. Miller (1979) used a state-transition approach based on a Markov chain model to simulate the size of an outbreak of FMD in the United States. Miller's model was used to generate outbreak scenarios for use in economic studies, and was based on herd contact rates modified from those observed in the very extensive and well documented 1967-68 epidemic in the United Kingdom (Wilson, 1987). Dijkhuizen (1988) used Miller's model for an economic evaluation of the two different control strategies of vaccination and stamping out (slaughter of all infected herds) in the Netherlands. Even when comparing the direct costs in the most favourable situation (0 outbreaks in 10 years) for a vaccinated cattle population with the most unfavourable situation (4 outbreaks in 10 years) for a non-vaccinated population, routine vaccination is still the less profitable choice.

Singh *et al.* (1997) used a stochastic model to show that in FMD endemic areas in India it was profitable for individual farmers to vaccinate regularly. He included only direct economic losses (reduced milk yield, decreased carcass value, medicines, nursing care), and the model was run varying the parameters for morbidity and mortality as caused by the disease. Even in the event of no mortality in the herd, the cost of vaccinating the herd was

considered lower than the overall expected losses caused by an outbreak. The most probable reason for the difference in outcome between this and the economic evaluation by Dijkhuizen (1988) was that Dijkhuizen evaluated the costs to the state, whereas this model evaluated the costs and benefits for an individual farmer.

A great deal of FMD modelling has been done in Australia and New Zealand, although neither of these has ever experienced an outbreak of FMD. If they ever were to have an epidemic, it could prove catastrophic for their agricultural (livestock) based economy. The Australian Bureau of Agricultural and Resource Economics has funded several studies into control strategies to contain FMD. Jalvingh *et al.* (1997) used a spatial and stochastic model that simulates the day to day spread of FMD between farms. It forms a part of the Epiman decision support system, originally developed in New Zealand, and is used for economic comparison of control strategies.

Airborne spread

Airborne spread of FMD has been modelled extensively after it was believed to have played a major role in the 1967-68 FMD epidemic in the UK, which involved more than 2000 farms (Hugh-Jones & Wright, 1970). Mathematical models have been developed to analyse and predict the role of airborne spread in outbreaks of FMD (Gloster *et al.*, 1981; Gloster *et al.*, 1982; Donaldson *et al.*, 1982; Donaldson *et al.*, 1987; Donaldson, 1988; Moutou & Durand, 1994). Donaldson *et al.* (1982) used a numerical model to predict the risk of FMD spread from an outbreak in Brittany, France. The model predicted outbreaks of FMD on the Channel Islands, which subsequently did occur, in 1981.

Moutou & Durand (1994) used epidemiological data linked to viral particle excretion and added meteorological data to develop a predictive model. The model computes the quantity of viral particles that a susceptible animal within a 10km radius of the outbreak could have breathed, its aim being to define a risk area for use by decision-makers in the event of an outbreak of FMD. Gerbier *et al.* (1997) modelled the effect of herd size on diffusion of FMD disease. The model assumed that animals were independent within a herd, and that the probability of infection of an animal was a logistic function of the quantity of airborne disease received. The risk for the herd was mainly linked to the total quantity of airborne virus received. This may be realistic for a small herd, however, larger herds are commonly kept in separate, smaller groups, whereby the risk of infection for each individual depends on which group they are in.

Many of the economic models simulate the spread of FMD (Miller, 1979; Thieme, 1983; Jalving *et al.*, 1997; Singh *et al.*, 1997), however most authors have modelled inter-herd transmission rather than intra-herd transmission. This is because FMDV is such a highly contagious virus that once an individual animal within a herd is diagnosed with FMD, a large proportion of the herd is likely to already be infected, and the entire herd is at great risk of infection. The economically most important aspect is, therefore, to prevent infection of other herds. In countries where 'stamping out' is practised, the entire herd will be slaughtered immediately if one animal is diagnosed with FMD. Even where 'stamping out' is not practised, the entire herd will almost certainly be quarantined.

Intra-herd modelling

Hutber & Kitching (1996) modelled the intra-herd transmission of FMD using vector-transition rather than state-transition. The authors decided on this method because results of serological investigations in Saudi Arabia showed a variation in antibody levels between segregated age groups on managed farms. Antibody titres are likely to account for a high proportion of incidence variance in FMD epidemics, and incorporating this would have complicated an intra-herd model to the extent that a state-transition technique would be unmanageable. The model was based on a stochastic model of rinderpest by James & Rossiter (1989). The model assumes spread of disease through contact. Vector-transition combines the daily change in both viral output of infected animals and the antibody titres of susceptibles with the transition of herd animals between disease states. Separate vectors were used for recovered and vaccinally immune animals, since these groups exhibit different rates of waning antibody. Segregated age groups could be modelled as discrete entities with differential input parameters, thus effectively becoming submodels within the model. Parameter values were estimated using data from Saudi Arabia detailing daily outbreak incidence within large dairy herds.

Cleland *et al.* (1994) developed a state transition model of the temporal development of herd immunity to FMD in response to vaccination. In the model, animals occupied one of three states: susceptible, immune or removed. The herd was divided into age classes of six month intervals from birth to >8 years. There was assumed no natural challenge, immunity developed only by vaccination. An animal was considered immune if its serum neutralisation titre was 1 in 32 or greater. A level of 80% prevalence of immune animals

was assumed to be necessary to prevent spread of FMD virus (herd immunity effect). Data for the model parameters were taken from a vaccination response study in Thai villages, in addition data were collected on the population dynamics of village cattle and buffaloes. Modelling was at village level, one herd defined as the aggregate of all cattle and buffalo in one village (set at 250 animals). The model indicated that even with 90% vaccination coverage there were periods immediately prior to revaccination where herd immunity dropped below an acceptable level. To ensure herd immunity at all times, vaccination coverage would need to approach 100%, an unrealistic objective in a village.

Models of FMD in free-living populations

There are surprisingly few models of FMD in wildlife populations. Although generally few authors have modelled wildlife diseases, FMD is a disease of great economic significance world-wide, and it is known to have wildlife reservoirs, for example amongst wild buffalo in southern Africa (Bengis *et al.*, 1987).

Pech & Hone (1988) used a mathematical model of the outbreak dynamics of FMD in feral pigs to study the factors necessary for persistence of virus in the population and the culling rates necessary for effective control. They concluded that very high culling rates would be necessary to eradicate FMD from feral pig populations should it become established. This is likely to be unrealistic in a wild population. Hone & Pech (1990) used this model to assess the probability of detecting FMD infected feral pigs using two different surveillance schemes (opportunistic and structured). With the current opportunistic surveillance, the model estimates that the probability of detecting an infected pig is less than 0.0015. Using a structured surveillance scheme, the model predicts that between 28 and 3077 FMD cases could occur before the disease outbreak is detected (the range depends on the level of certainty required for detection). The authors highlighted the need for an epidemiological model that included more than one host species. Pech & McIlroy (1990) used a diffusion model to calculate the minimum velocity of advance of an outbreak of FMD in feral pigs in Australia. The model is based on the Pech and Hone (1988) model and used the same parameter values. The diffusivity and contact rate were calculated from data collected during radio tracking of feral pigs. In contrast to fox/rabies models, the movements of animals in all classes were included, as it was assumed that FMD would be transmitted in the course of normal daily behaviour. They assumed that intra-group contact is so frequent that FMD virus would spread rapidly to all group members. This model took no account of

climatic variability or seasonality of births, but it is mentioned that seasonal patterns could be modelled with a drift term (forcing function). The model predicted that an outbreak of FMD in an isolated population of 24 feral pigs would die out naturally. However, it must be taken into account that feral pigs have a more or less continuous distribution over large areas of the tablelands of south-eastern Australia, such that the opportunity for rapid spread of FMD is considerable (Pech & McIlroy, 1990).

10.3 Modelling FMD in a population of saiga antelopes

10.3.1 Key factors in modelling FMD in saigas

The population dynamics of saiga antelopes have been modelled (Rakhimberdiev *et al.*, 1974; Milner-Gulland, 1994), however, modelling of FMD in a saiga population has never been attempted.

Models of FMD spread in domestic livestock have concentrated mainly on inter-herd spread, thus considering each herd a unit, rather than the individual animal. It could be possible to model saigas in such a way, as they congregate in herds, however, the size of the herds varies constantly and dramatically throughout the year, from less than 50 animals in December to tens of thousands in May. This makes such an approach very difficult. Also, it is common to use population density in models where the population cannot expand the area it occupies as its numbers increase. However, saigas, being nomadic animals, can to quite an extent expand their range area as their numbers increase, and are, therefore, more easily modelled as numbers. Density dependent transmission was not employed in the model, as the current saiga population is well below carrying capacity and density dependence was thus thought not to be an issue. Furthermore, the fact that saiga can expand their range area means that frequency dependent transmission was likely to more accurately reflect transmission of FMD among saiga antelopes.

Saiga population size varies greatly during the year due to high mortality rates and a seasonal birth period of very short duration. Annual population size also varies greatly, for instance the Betpak-dala population has varied between an estimated 120,000 and 995,000 animals during the past 30 years (Bekenov *et al.*, 1998). The large fluctuations in population size and density, and thus in contact rates, by year and season mean that infectious disease may spread more rapidly at certain times. The migratory pattern of saigas adds a further seasonal aspect to their risk of acquiring infection from domestic

livestock. It is crucial to include these aspects of saiga demography in any model of disease transmission. Such a model cannot be based on other wildlife models, such as Pech & Hone (1988), where seasonal aspects of reproduction and climatic variability have been conveniently ignored. Many wildlife species have seasonal reproduction, but most do not have quite such a short birthing period or as dense a population at this time of year as do saiga antelopes.

Ideally, the stochastic effects of climatic variability also need to be taken into account, because they cause such a high variability in mortality rates, and also affect fecundity rates. Additionally, climatic factors may influence virus transmission rates.

Pech & Hone (1988) collected data on contact rates between herds from radio tracking, and they assumed that any contact would cause infection. This is a reasonable assumption for FMD. Unfortunately, there have been no studies of saiga contact rates. It may be possible to estimate contact rates roughly from herd sizes, however, saiga herds are constantly regrouping and mixing, and this does of course increase the contacts each individual animal has. Most models assume homogenous mixing. This may be an adequate approximation of saiga behaviour in the spring, but certainly not at the times of year when they are in smaller herds, such as during the rut or on the summer pasture. At low R_0 , deviations from homogenous mixing become more important, this may cause a problem for modelling. An approximation of R_0 can be found, using available data on disease outbreaks, to check whether there is a problem in the case of saigas.

10.3.2 Aims of model

The aims of a model of FMD in saigas would be to identify the dominant processes of FMD transmission, and to give qualitative predictions, such as the risk of disease spread through saiga and livestock populations, and the velocity of disease spread incorporating the seasonal aspects of migration routes. A further aim would be to demonstrate the importance (if any) of herd size for spread, and the effects of seasonality. A spatial model is not necessary for predicting the velocity or direction of spread in the case of the saiga; a temporal model will have the same function as saigas migrate.

10.3.3 Model Description

The basic model

The model is deterministic, with density dependence on mortality. Transmission of disease is frequency dependent. The basic framework used the SIR model, and was created using ModelMaker software (SB ModelMaker Version 2.0c, 1993). Continuous weekly time steps were used for output. The duration of infectiousness for FMD in saiga is approximately one week, integration time steps are much less than this. The model ran over periods of 52 weeks, week 0 corresponding to the first week of January. This allowed seasonal aspects to be added, for example summer occurred during weeks 13 - 39. The model had two age classes, calves and adults. All calves were transferred simultaneously to the adult compartments; this was related to the natural phenomenon of mass calving which meant that all calves were more or less of the same age every year. The transferral occurred every year in week 45, which was 26 weeks after the birthing period starts, i.e. when the calves were 6 months of age. Females can reproduce from the age of 7-8 months, so 6 months was seen as an appropriate time for calves to become adults. For both age classes the population was divided into susceptible, latent (infected, but not yet infectious), infectious and recovered (immune) classes. In addition, the calf population had a maternal immunity class.

In the absence of FMD infection, all animals are born into the susceptible calf compartment (S_c). Seasonality of births is introduced into the model by defining a short period of 1.5 weeks, (week 18.5 to 20, equivalent to mid-May), during which all births occur. This emulates the natural occurrence of mass calving amongst the saiga, and this is made to occur every 52 weeks in the model. The seasonal birth-rate (B) is related to the adult population size (P_a), giving the equation:

$$1) \quad B = \frac{\mu_b p_f P_a}{D_b}$$

where

B = birth rate (number of calves born per week)

μ_b = average number of calves born per female

p_f = proportion of females in population (all females are reproductively active)

P_a = adult population size

D_b = duration of birth season in weeks

In the presence of FMD infection, births enter into either the susceptible class S_c or maternal immunity class (M_c) at a rate of $B(1-R_a/P_a)$ or $B(R_a/P_a)$ respectively, where R_a is the recovered (immune) adult class. The model assumes homogenous mixing, and uses the mass-action principle for the net transmission of infection, βSI , where β is the transmission coefficient that defines the probability of contact and infection between a susceptible and infectious individual. Assuming that any contact is sufficient to permit transfer of FMD between saigas, β represents the effective contact rate (number of contacts per week) of saigas. A system of differential equations describes the rates of change of the populations with respect to time:

Equations for the adult population:

$$3) \quad \frac{dS_a}{dt} = \frac{R_a}{\omega_a} - \beta S_a (I_a + I_c) - \mu_a S_a$$

$$4) \quad \frac{dL_a}{dt} = \beta S_a (I_a + I_c) - \frac{L_a}{\sigma} - \mu_a L_a$$

$$5) \quad \frac{dI_a}{dt} = \frac{L_a}{\sigma} - \frac{I_a}{\nu} - \mu_a I_a - \alpha_a I_a$$

$$6) \quad \frac{dR_a}{dt} = \frac{I_a}{\nu} - \mu R_a - \frac{R_a}{\omega_a}$$

where $P_a = S_a + L_a + I_a + R_a$

Equations for the calf population:

$$7) \quad \frac{dM_c}{dt} = B(1 - \frac{R_a}{P_a}) - \frac{R_c}{\omega_m} - \mu_c M_c$$

$$8) \quad \frac{dS_c}{dt} = B(\frac{R_a}{P_a}) + \frac{M_c}{\omega_m} - \beta S_c (I_a + I_c) - \mu_c S_c$$

$$9) \quad \frac{dL_c}{dt} = \beta S_c (I_a + I_c) + \frac{L_c}{\sigma} - \mu_c L_c$$

$$10) \quad \frac{dI_c}{dt} = \frac{L_c}{\sigma} - \frac{I_c}{\nu} - \mu_c I_c - \alpha_c I_c$$

$$11) \quad \frac{dR_c}{dt} = \frac{I_c}{\nu} - \mu_c R_c$$

with $P_c = M_c + S_c + L_c + I_c + R_c$; $N = P_a + P_c$

where

P_c = calf population

N = total population

It is here assumed that animals acquire latent infection at a rate of $\beta S(I_a + I_c)$, become infectious at a rate of $1/\sigma$ (average latent period in weeks, σ), and do not 'recover' to a latent inactive state. Infectious animals are assumed to have an increased mortality rate, α_c , over that suffered by susceptible, latent or recovered saigas. All infectious animals that recover are assumed to develop immunity at a rate of $1/\nu$ (average duration of infectiousness in weeks, ν). This acquired immunity is lost at a rate of $1/\omega$ (duration of immunity, ω), and recovered animals re-enter the susceptible class. There is no loss of acquired immunity amongst calves, because the duration of acquired immunity is 3 years, and calves become adults at 6 months of age.

Infection is introduced into the population via x latently infected adult saigas. The time at which this occurs is specified in the model, and can be varied such that infection can be introduced at any time of the year. The disease is considered eliminated when the total number of infected animals is less than y ($L_a + L_c + I_a + I_c < y$); at this stage all latent and infectious animals are moved to the recovered class.

Natural mortality was made dependent on age, season and population density. The natural adult mortality rate for the winter season (week 13 to 39) was defined as:

$$13) \quad \mu_a = \mu_w + \frac{\mu_{dd} N}{K}$$

where μ_w = adult winter mortality coefficient

μ_{dd} = adult density dependent mortality coefficient

N = total population

K = carrying capacity

for the summer season :

$$14) \quad \mu_a = \mu_s + \frac{\mu_{dd} N}{K}$$

where μ_s = adult summer mortality coefficient

μ_{dd} = adult density dependent mortality coefficient

N = total population

K = carrying capacity

The natural calf mortality for the first 4 weeks of life (weeks 19 to 23) is defined as:

$$15) \quad \mu_c = \mu_e + \frac{\mu_{dc} N}{K}$$

where μ_e = calf (early) mortality coefficient

μ_{dc} = calf density dependent mortality coefficient

The natural calf mortality for the age of 1 to 6 months :

$$16) \quad \mu_c = \mu_l + \frac{\mu_{dc} N}{K}$$

where μ_l = calf (late) mortality coefficient

μ_{dc} = calf density dependent mortality coefficient

These equations make up the basic model, in the latter sections modifications are made to this framework to take account of additional biological processes and assumptions. The model variables have been summarised in Table 10.1. The possibility of a carrier state has been ignored, owing to the large amount of literature stating that experiments have failed to prove that carrier animals transmit infection to susceptible animals (Sellers, 1971; Thomson, 1996). Flows between compartments are indicated in Figure 10.1.

Figure 10.1

Fig chap10_1

Table 10.1 Model variables	
<i>Parameter</i>	<i>Symbol</i>
Birth rate (births per weeks)	B
Adult population size	P_a
Susceptible adults	S_a
Susceptible calves	S_c
Immune calves (maternal immunity)	MI_c
Latently infected adults	L_a

Latently infected calves	L_c
Infectious adults	I_a
Infectious calves	I_c
Recovered adults	R_a
Recovered calves	R_c
Total population	N
Mortality coefficient	μ
FMD-induced mortality	?
Latent period (in weeks)	?
Infectious period (in weeks)	?
Duration of immunity (in weeks)	?

Incorporating seasonally varied transmission rate

This model is identical to the basic model, with the exception that the effective β (β_e) is not constant. Instead there is a saiga component (β_s) relating to the transmission of FMDV within the saiga population, and a livestock component (β_l) which relates to FMDV transmission between the saiga and domestic livestock populations. Together, these two components influence the value of β_e by weighting of this parameter. Transmission is still frequency dependent, as in the basic model. The livestock component can be taken out, allowing the model to look at transmission of FMD exclusively within the saiga population, without regard to the adjacent domestic livestock.

$$17) \quad \beta_e = \beta (a\beta_s + b\beta_l)$$

where

a = weighting coefficient for saiga to saiga transmission

b = weighting coefficient for livestock to saiga transmission

10.3.4 Parameterisation

Background

When building a model it is desirable to have estimates available for each of the parameters used in the model. In a population of wild animals there is usually no accurate census, and although the numbers of births and deaths may have been estimated by observations in the field these are rarely accurate, especially when involving species that roam over large areas and shy away from human contact. In saiga populations mortality rates vary enormously according to the climatic conditions, additionally there is illegal hunting all year round by poachers that is difficult to quantify.

Parameter Estimates

The following parameter estimates are based on ecological, biological and experimental data collected in Kazakhstan and Russia over a 40-year period, including data from this study. Published data for each parameter are quoted where they are considered reliable. Where data from the saiga were unavailable an estimate was made using data from other species. Mean or best-estimate values were selected for numerical simulation, and are shown in table 10.2 at the end of this section (page 225).

Carrying capacity (K)

The saiga population has been hunted for commercial purposes since 1954, with between 12% and 39% of the population killed every year (Bekenov *et al.*, 1998). The population has thus been kept at a level well below carrying capacity. Population size greatly varies during the year due to high mortality rates and a seasonal birth period of very short duration (Bekenov *et al.*, 1998). The maximum population size in Betpak-dala that was estimated by aerial counts was 995,000 in 1974 (Fadeev & Sludskii, 1982). All aerial counts have taken place in April, just before the calving season. In the absence of reliable data, a value of 2,000,000 was chosen as best-estimate of the current theoretical carrying capacity of the Betpak-dala population in April, the time of year when the population size is at its lowest.

Proportion of females (p_f)

According to published data on the population structure of the saiga, the proportion of adult males varies between 0.03 – 0.25, adult females 0.37 – 0.66 and juveniles 0.23 – 0.47 (Table A6.1, Appendix 6). These values are all for a selectively hunted population, so do not represent the “natural” age structure. The sex ratio of 6-month-old calves is close to 1:1 (Bekenov *et al.*, 1998). For the model, the assumed population structure took the form of 25% adult males, 50% adult females, 12.5% juvenile males and 12.5% juvenile females, with the total proportion of females (all are reproductive) in the population being 0.625. This structure is taken from the stable age distribution of an age-structured model (Milner-Gulland, 1994b), and is thus representative of the underlying dynamics of the population. As the population dynamics are highly distorted by hunting, and this may have an effect on FMD transmission, this needs to be addressed in future research.

Average number calves per female (μ_b)

In the Betpak-dala population, the mean number of embryos per adult female has been reported as 1.7 and per juvenile female 0.9 (Bekenov *et al.*, 1998). These figures were used to calculate a value of 1.5 for the entire population, taking into consideration its structure (Table A6.2, Appendix 6). A value of 1.54 was calculated using the hypothetical population structure in the model. It was decided to use the mean value of 1.5 calculated from actual data from the Betpak-dala population. Density dependent effects were put on mortality rather than fecundity (discussed in section on density dependent mortality).

Calculation of mortality rates

There are no good data available for mortality rates, unlike for fecundity rates. For this reason, mortality rates were estimated crudely from data giving mortality as percentages of the total population or of an age or sex category over a specified period of time. A stochastic model would be necessary to include the yearly climatic conditions that cause dramatic differences in mortality from one year to the next. In this deterministic model, climatic effects were incorporated into an average mortality by weighting according to the time of year and the age of the animal.

Example 1.

Average winter mortality of the adult population was calculated from data giving sex based mortality in normal and bad years (Milner-Gulland, 1994b), using the population structure defined above. In a normal winter, 10% adult females and 25% adult males died. In a bad summer, 20% of adult females and 50% of adult males died. Assuming a population structure of 50% adult females and 25% adult males, there is an adult sex ratio of 2/3 females and 1/3 males. The total adult mortality was calculated from these figures. In a normal winter, total mortality was $(0.1 * 2/3) + (0.25 * 1/3) = 0.15$, i.e. 15%. In a bad winter (*dzhuts*) total mortality was $(0.2 * 2/3) + (0.5 * 1/3) = 0.3$, i.e. 30%. The mortalities were weighted according to likelihood of occurrence. Using the assumption that *dzhuts* occurs once every 10 years, a normal winter was weighted 0.9, a bad winter weighted 0.1. Average winter mortality was thus $(0.15 * 0.9) + (0.3 * 0.1) = 0.165$, i.e. 16.5% mortality. Conversion from absolute mortality to continuous mortality rates per week, as needed in the model, were calculated using the following formula:

$$18) \quad m_r = - \ln (1-m) / t$$

where m_r = mortality rate per week

m = mortality

t = time period (in weeks) over which mortality occurs

Using the example of adult winter mortality in the calculation:

$$m_r = -(\ln(1-0.165)) / 26 = 0.0069$$

Example 2

Assuming 33.3% mortality in the first month of life and 52.7% mortality in the first 6 months of life (see section: *Natural Calf Mortality rates* (μ_c), page 400), mortality from weeks 4 to 26 was calculated as follows: At 4 weeks 66.7% are alive, at 26 weeks 47.3% are alive. I.e. from 4 to 26 weeks an additional 19.4% of calves died. Given that 66.7% of the original number were alive at 4 weeks, the proportion of those that died during the period was $0.194/0.667 = 0.291$, i.e. 29.1% died.

$$m_r = -(\ln(1-0.291)) / 22 = 0.0156$$

Natural Adult Mortality rates (μ_a)

According to the literature, adult mortality rates can vary between 0.1 and 0.5 per annum, depending heavily on climatic conditions, with seasonal variation and gender playing an important role (Bannikov *et al.*, 1961; Rashek, 1963; Fadeev & Sludskii, 1982; Bekenov *et al.*, 1998). Milner-Gulland (1994) included climatic variability (the occurrence of *dzhuts* and summer droughts) in her stochastic population model that had specific age and sex categories. This model assumes the same basic parameter values (Table A6.3, Appendix 6), but the values were recalculated using Milner-Gulland's assumption of *dzhuts* occurring every 10 years and summer drought every 3 years. This gave an average value of 16.1% (0.0068 per week) for summer mortality and an average value of 16.5% (0.0069 per week) for winter mortality averaged over sex and climate, using the method explained above.

Natural Calf Mortality rates (μ_c)

Calf mortality rates varying from 14 to 76% have been reported (Bannikov *et al.*, 1961; Fadeev & Sludskii, 1982; Milner-Gulland, 1994b; Bekenov *et al.*, 1998). In general, higher mortality rates are seen the first few weeks of life. Bannikov *et al.* (1961) reported

mortality in the first month of life of 20% in normal years and 60% in drought years. As described above, the data were recalculated using the assumption of a summer drought occurring every 3 years, giving an average mortality rate of 33.3% (0.1012 per week) for calves 0 – 4 weeks of age (Table A6.4, Appendix 6). Calf mortality from spring to autumn in the Betpak-dala population was recorded to vary between 29.2% and 72.2% (mean 52.7%) in the years 1990 – 1993 (Bekenov *et al.*, 1998). Using the mean value of 52.7% and assuming that 33.3% of the calves died in the first four weeks of life, the mortality of calves 4 – 26 weeks of age was calculated to be 29.1% (0.0156 per week) (see example above).

Adult Mortality due to FMD (%)

Epidemics of FMD may result in the death of 3 – 10% of adults (Sokolov & Zhirnov, 1998). Mortality is high amongst calves, but is also high among females in the late stages of pregnancy (Table A6.5, Appendix 6). In the model a value of 10% (0.0976 per week) is used for the 4 weeks preceding the birthing period. For the remainder of the year a value of 3% (0.0282 per week) is used. Table A6.6 in Appendix 6 describes the calculations.

Calf Mortality due to FMD (%)

In the 1957 outbreak in Kalmykia, Bannikov *et al.* (1961) reported 66.7% mortality among newborn calves in one area. In another area a mortality of 78.8% was seen in calves during the first month of life (Bannikov *et al.*, 1961). In the model FMD induced mortality is additional to natural calf mortality, so a value of 45.5% (0.5620 per week) was chosen for calves 0-4 weeks of age. For calves 4 – 26 weeks of age the model uses the same mortality as for adults, 3% (0.0282 per week). Table A6.6 in Appendix 6 describes the calculations of weekly mortality rates. The time from infection to death was assumed to be the duration of the infectious period (1.089 weeks, 7.6 days).

Density dependent mortality (μ_{dd} , μ_{ddc})

Density dependence is accepted to occur in the saiga; Coulson *et al.* (2000) recently demonstrated density dependence in the fecundity of the saiga. However, there are no reliable data available on density dependent mortality in the saiga. As mentioned earlier, hunting has prevented the saiga populations in Kazakhstan from reaching carrying capacity in recent times. Because density dependence has been demonstrated to affect fecundity, this is approximated in the model by putting density dependent mortality mainly on the

juvenile animals (0.00598, equivalent to 14.4% mortality at carrying capacity) additional to natural mortality. It is possible that adult saigas could be constrained by resources, in particular in winter, so density dependence, albeit at a low value (0.00039, equivalent to 1% mortality both for winter and summer at carrying capacity) was included also for the adults. These values were chosen because they gave a population size at equilibrium of roughly 2,000,000, the assumed carrying capacity.

Latent period (?) and Infectious period (?)

Experimental data are available from Nagumanov (1972), including data from animals inoculated with FMD virus, and from animals exposed to contact infection (Table A2.1, Appendix 2). Contact infection, whereby the animal is exposed to infectious material, is more similar to natural infection than is infection by inoculation, where viral particles are inoculated directly into the tongue. In the model the values found for contact infection are used, as it was thought these would be more realistic. The experimental data showed that the latent period of saigas that experienced contact infection was 24 – 48 hours, and the duration of viral excretion was 144 – 240 hours. The mean values of 36 hours (0.2143 week) for the latent period (?) and 183 hours (1.0893 week) for the infectious period (?) were chosen for the model.

Duration of immunity (?_a)

An experiment by Bukhtiyarov (1967) showed that saigas retain antibodies for at least 200 days post infection. Fadeev & Sludskii (1982) noted that in the field, saigas seemed to retain protective immunity against FMD for at least two years. It is known that immunity in cattle (to a homologous virus) lasts at least three years following infection (R.P. Kitching, pers. comm.) In the absence of reliable data it was decided to use a value of 3 years (156 weeks).

Duration of maternal immunity (?_m)

There are no data for the duration of conferred maternal immunity in the saiga; it was, therefore, necessary to extrapolate from other species. Maternal antibodies specific to FMDV have been found to persist for up to five months in domestic calves and 3 – 7 months in buffalo calves (Ahl & Wittman, 1987; Condy & Hedger, 1974). For modelling, a value of 5 months (21 weeks) was used.

Basic reproductive rate (R_0)

The basic reproductive rate (R_0) can be calculated from previous outbreaks by the following formula (Dietz, 1993):

$$19) R_0 = \frac{\ln(s_b) - \ln(s_a)}{s_b - s_a}$$

where s_b = proportion of susceptible animals before the outbreak

s_a = proportion of susceptible animals after the outbreak

In the autumn cull after FMD outbreaks of the 1950s and 1960s, 17 – 20% of males were found with stunted horns, indicating they had been infected (Fadeev & Sludskii, 1982; Sokolov & Zhirnov, 1998). Assuming the proportion susceptible before the outbreak was 100%, and afterwards 80%, R_0 is calculated to be 1.116 when these figures are put into the equation above. However, mortality among saigas of 3.0 – 78.8% has been observed after outbreaks of FMD (Sokolov & Zhirnov, 1998). R_0 was calculated using these figures, giving a range of 1.054 – 1.968.

Transmission coefficient (β_t)

β_t is the transmission coefficient that defines the probability of contact and infection between a susceptible and infectious individual. Assuming that any contact is sufficient to permit transfer of FMDV, β_t represents the contact rate. This assumption is valid, considering that FMDV is probably the most contagious virus known. There is unfortunately no reliable data on contact rates of saigas. The only information available is the population size and range area, and average herd sizes during the year. β_t can be calculated from R_0 using the following formula

$$20) R_0 = \beta_t D_i N$$

where D_i = duration of infectiousness (in weeks)

N = population size

The full range of values for R_0 calculated above, combined with a population size ranging from 200,000 (population size of Betpak-dala in the 1990s) to 2,000,000 (assumed

carrying capacity), was used to calculate a range of $4.7E^{-07} - 9.1E^{-06}$ for β_t (table A6.7, Appendix 6). Within this a more realistic range of β_t $5.07E^{-07} - 5.17E^{-06}$ was chosen by cutting out the extreme values.

In model 2 (which incorporates a seasonal transmission coefficient), there is a saiga component (β_s) relating to the transmission of FMDV within the saiga population, and a livestock component (β_l) which relates to FMDV transmission between the saiga and domestic livestock populations. Together, these two components are used to weight β_t , giving the effective value of β_e . β_s was calculated using the monthly variation in saiga herd size (Table A6.8, Appendix 6). Weighting was achieved by dividing average monthly herd size by the annual average herd size and was thus a direct approximation of the contact rate of saiga (Table A6.9, Appendix 6).

Saiga migration is included indirectly in the model by weighting the transmission from domestic livestock, using a higher transmission coefficient at times of year when saigas are more likely to contract FMD from livestock, β_l . For calculating β_e it is assumed that the risk of saigas acquiring infection from other saigas is 10 times greater than the risk of saigas acquiring infection from domestic livestock, hence $a = 0.9$ and $b = 0.1$.

β_l was calculated using two factors: the risk of saigas acquiring infection through contact with domestic livestock (calculated using the mean density of ruminant livestock at pasture, Table A6.10, Appendix 6), and the monthly variation in relative humidity (RH) as an approximation of the likelihood of saigas acquiring wind-borne infection from domestic livestock. Details of the calculation are described in Table A6.11, Appendix 6. Close contact between saigas and domestic livestock is a rare event because saigas fear humans, however, transmission between saigas and livestock may occur through shared pastures and water points, or from airborne spread. Our survey suggests that at present (1997 – 1999) the saiga summer and migration ranges are almost devoid of livestock, whilst the winter range contains greatly reduced numbers of livestock. Livestock density at pasture was used to calculate the relative increased risk of infection experienced by saigas in their winter range, using data on livestock densities from the Kazakhstan State Committee on Statistics and Analysis (Kulekeev, 1998). The most important factor for airborne virus

survival is relative humidity; this was, therefore, used to estimate the likelihood of airborne infection. Figure 10.2 shows how β_i , β_s and β_e vary throughout the year.

Figure 10.2

Fig chap_10_2

Table 10.2 Parameters, with best estimate values used in the model			
<i>Parameter</i>		<i>Symbol</i>	<i>Value</i>
Average number calves produced per female per season		μ_b	1.5
Proportion of females in population		P_f	0.625
Duration of birth season in weeks		D_b	1.5
Adult winter mortality coefficient		μ_w	0.0069
Adult density dependent mortality coefficient		μ_{dd}	0.00039
Carrying capacity		K	2,000,000
Adult summer mortality coefficient		μ_s	0.0068
Calf (early, 0-4 weeks) mortality coefficient		μ_e	0.1012
Calf density dependent mortality coefficient		μ_{dc}	0.00598
Calf (late, 1-6 months) mortality coefficient		μ_l	0.0156
Transmission coefficient		β_t	$5 \times 10^{-7} - 5 \times 10^{-6}$
Effective transmission coefficient		β_e	See equation 17
Weighting of saiga component		β_s	See table 10.8
Weighting of livestock component		β_l	See table 10.9
FMD-induced mortality in adults	Wks 14.5 – 18.5	γ_a	0.0976
	All other wks		0.0282
FMD-induced mortality in calves	Wks 19 – 23	γ_c	0.5620
	All other wks		0.0282
Latent period in weeks		?	0.2143
Infectious period in weeks		?	1.0893
Duration of immunity in weeks		γ_a	156
Duration of maternal immunity in weeks		γ_m	21

10.3.5 Results

The demography of the saiga population in the absence of FMD infection is shown in Figure 10.3. The initial population is at equilibrium ($N = 2,298,044$) when the model is run. During weeks 0 – 18 (winter) there is a steady decline in the adult population size due to natural mortality, then in week 45 (autumn) there is a dramatic increase, as the calves become adults. The calf population only exists between weeks 18.5 and 45 every year (from birth in spring until adulthood in autumn). During this period they experience a dramatic decline during the first 4 weeks, due to high mortality among young calves, then a more gradual decline caused by natural mortality of the older calves.

Figure 10.3

Figure 10.4

Fig chap10_3

FMD infection is introduced into the saiga population by the addition of two latently infected animals. The calving season is a key component of the model, determining the dynamics of any FMD epidemic by producing a large pool of susceptible individuals. Consequently, the date at which infection is introduced into the population is a key determinant of an epidemic's progress (Figure 10.4). If infection is introduced in autumn, it will persist in the population, leading to cyclical epidemics regardless of the value of β . If infection is introduced in winter or spring, it will persist in the population if β is high or low, but for a medium value of β there will be only one outbreak. The reason is that for a medium value of β , there are too few susceptible animals left after the initial outbreak to sustain transmission. With a high value of β , infection will circulate even though there are few susceptible animals, whereas with a low value of β the initial epidemic is on a much lower scale, leaving enough susceptible animals for infection to persist. In summer a high value of β allows persistence of the virus, but for medium and low values a single outbreak occurs after which it 'burns out'.

Figure 10.4 The effect of varying the time of year that infection is introduced. Initial population size in week 0 is at equilibrium ($N= 2,298,044$). Dark shading indicates cyclical epidemics, light shading one epidemic

Figures 10.5 a

Fig chap10_4

10.3.6 Investigating the model

The time from the introduction of the infection to the peak of the first epidemic, the size of the peak of the epidemic and the duration of the first epidemic varies according to the value of β (Figures 10.5 a – c). With a higher β value the time taken to reach the peak of the epidemic is shorter, a higher peak is reached and the duration of the epidemic is shorter. For some values of β at the lower end of the scale, the results deviate from the overall trend, for example the size of the epidemic peak for $\beta = 8 \times 10^{-7}$ is higher than for $\beta = 9 \times 10^{-7}$, even though the latter value of β is higher. This is because the value of β is low enough for the epidemic to continue from the point of introduction in late autumn into the calving period, when a new pool of susceptible animals is introduced. The epidemic thus reaches a higher level than expected from its β value.

Figures 10.5 b

Figures 10.5 c

Figure 10.6

Fig chap10_5

For a high value of β (5×10^{-6}) there are two epidemics per year, a large epidemic which coincides with calving in spring, and a smaller one in the autumn. The initial epidemic is on a much larger scale than the others; however, this is not seen on the graph as the y-axis has been truncated for clarity. For a medium β value (9.3×10^{-7}), the second epidemic follows two years after the initial outbreak, and then there is a yearly cycle thereafter. For a low value of β , (5×10^{-7}) the epidemic cycles every two years.

10.3.7 Sensitivity Analysis

A sensitivity analysis was performed, varying the parameter values by $\pm 20\%$. Parameters for which there is reliable, accurate data and which are not subject to natural variation, such as the duration of the birthing season and the calving ratio, were not included in the sensitivity analysis. There was no variation in the qualitative output of the model when the following parameter values were varied by $\pm 20\%$; natural mortality (calf and adult), density dependent mortality (calf and adult), duration of immunity (β), duration of latent period (β) and duration of infectiousness (β), thus these results are not discussed. The parameters that did have major effects were initial population size (N) and the transmission coefficient (β).

N and β were varied together in the sensitivity analysis, as single parameter variation is less informative than multi-parameter variation (Kremer, 1983). When the model is run with a high β value (Figure 10.7a), cyclical epidemics are seen regardless of which time of year infection is introduced as long as the population size in week 0 (January) is large (2,000,000 – 2,500,000 individuals). In a medium (1,000,000 – 1,500,000) or small (250,000 – 750,000) sized population, cyclical epidemics are observed only if infection is introduced in autumn or winter. If infection is introduced in spring or summer, a single epidemic is seen because the epidemic ‘burns out’ by depleting the pool of susceptible animals, after which it cannot manage to keep circulating until the introduction of new susceptible animals into the population in the spring.

Figure 10.7 The effect of initial population size on the outcome when FMDV infection is introduced into the population at different times of the year. Small = 250,000 – 750,000; Medium = 1,000,000 – 1,500,000; Large = 2,000,000 – 2,500,000. β is constant all year. Dark shading indicates cyclical epidemics, lighter shading one epidemic, very light no epidemic

For a medium β value (Fig. 10.7b), there is no epidemic at all if the population is small. This is related to the ‘threshold theory’, whereby the number of susceptibles must be above a certain critical value for an epidemic to occur. For a low β value (Fig. 10.7c), outbreaks of disease are only seen when the population is large, otherwise there are not enough susceptible animals available to allow continuous transmission of virus.

Seasonally varying β ?

More realism can be introduced by allowing the effective β (β_e) to vary seasonally, taking into account the saiga's intra-specific contact rate and their risk of acquiring FMD from domestic livestock (Figure 10.8).

Figure 10.8 The effect of varying β according to the season. Initial population size in week 0 is at equilibrium ($N= 2,298,044$). All parameters are as in table 10.9. Dark shading indicates cyclical epidemics, lighter shading one epidemic, very light no epidemic

With a variable β value, regardless of the value of β , the model predicts that only one epidemic will occur if infection is introduced in spring (Figure 10.8b). This is because such a high proportion (92%) of the population is infected that there are insufficient susceptible animals left for the virus to persist through the rest of the year when β is much lower. When β is set at a high value and infection is introduced at other times of year, less than 70% of the population is infected, leaving enough susceptible animals for virus to persist even when β is at a lower value.

When β is set at a medium value, β is not high enough in summer or autumn to perpetuate the epidemic. If infection is introduced in winter, no epidemic occurs, as this is the time of year with the lowest population size. A single epidemic occurs when infection is introduced at any other time of year. For a low value of β , the model predicts that an epidemic will only occur if infection is introduced in spring. During the rest of the year β is too low to cause an epidemic.

The effect of population size on the output of the model when β changes seasonally was investigated (Figure 10.9). In both a medium (1,000,000) and small (500,000) sized population outbreaks of disease are only seen when β is high. The exception is when infection is introduced into a medium sized population in spring, when a single outbreak occurs.

Figure 10.9 The effect of varying population size. β varies by season. Dark shading indicates cyclical epidemics, lighter shading one epidemic, very light no epidemic

10.4 Discussion

In this study, a model framework has been developed for the study of FMD dynamics in saiga antelopes. The model has been parameterised using the best available data, however, the data are generally not of the greatest reliability. The preliminary results show some interesting interactions between seasonality, demographics, periodicity and infection dynamics. This type of dynamic interaction, caused by strong seasonal forcing in the saiga's population dynamics combined with their risk of acquiring FMD from domestic livestock, has not been fully explored in the epidemiological modelling literature.

Both this study and the literature have suggested that domestic livestock are the source of FMD infection for saigas, so there is a crucial need for livestock to be included in any model. When including a seasonally dependent β value, the model produces a pattern that is qualitatively similar to that seen in Kazakhstan in the 1950s and 1960s, when there were large epidemics in spring which died out in the summer or autumn. This model includes a livestock component, as livestock movement patterns are important for transmission of FMD virus to saigas. However, livestock movement patterns are rather complicated. Previously, they mirrored saiga movement somewhat, as livestock also moved seasonally to the best pastures. However, they have dramatically changed in recent years. There are now very few people who migrate with their livestock. Yet the data presented in Chapter 6 show that people in villages in saiga range areas are still observing saigas close to villages and drinking from places where livestock drink. Thus contact between saigas and livestock still occurs, although probably at a much lower level than previously. Assuming that an outbreak of FMD among livestock could cause an epidemic among saigas, the epidemic is probably unlikely to persist. Furthermore, the saiga population is diminishing in size, reducing the likelihood of an epidemic of FMD actually occurring.

10.5 Future work

The current model represents an initial attempt to understand the interaction between time-varying components of the system, involving saigas, livestock and FMDV. A fuller analysis is required to develop a quantitative, predictive model, which could be used as part of a risk analytic or strategic framework. Knowledge of saiga migration routes together with meteorological data (wind direction and relative humidity) would enable estimation of the direction of spread, and the likelihood of spread into populations of domestic livestock. Ideally, migration patterns of livestock should also be incorporated into the model. Before using the model to advise on control policy it needs to be validated.

However, this will be very difficult, due to the lack of reliable epidemic data. There are some data available on FMD outbreaks among saigas (before 1975), but these do not include accurate numbers of infected animals or information on the duration of epidemics.

The exercise of developing and parameterisation of the model has been extremely valuable in highlighting the key factors that must be taken into account in considering the epidemiology of FMD in saiga antelopes. These include:

- the interactions with livestock; a single-species model is not appropriate.
- the seasonality of saiga population dynamics and migration patterns.

The model-building exercise has also highlighted the complexity of the system. A model of FMD in saigas needs to be significantly more detailed than those developed thus far in the literature on wildlife disease dynamics. The model described here is deserving of more attention. Future work will include exploring the predictions of the model, improving the parameterisation and development of a stochastic model to allow incorporation of environmental variability. The large seasonal and annual variation in fecundity and mortality rates may have a big effect on whether an epidemic takes off. Key factors for which better data are required include saiga mortality rates and saiga-livestock contact rates. These could be addressed using saiga tagging experiments and radio-tracking.

Chapter 11

Discussion

11.1 Infectious diseases of livestock

The diseases that were studied had slightly different profiles in terms of the factors predisposing an animal to producing a seropositive result (Table 11.1).

Table 11.1 Factors significant in predicting seroprevalence for FMD, brucellosis, bluetongue, EHD and PPR. Crosses indicate significance, zeros lack of significance, and – that the test could not be carried out.

<i>Factors</i>	<i>FMD</i>	<i>Brucellosis</i>	<i>Bluetongue</i>	<i>EHD</i>	<i>PPR</i>
<i>Species</i>	X	X	0	X	X
<i>Breed</i>	0	0	X	X	X
<i>Age</i>	X	X	X	-	-
<i>Year of sampling</i>	X	0	X	X	X
<i>Herd size</i>	X	X	0	0	0
<i>Origin</i>	X	X	0	0	0
<i>Condition</i>	0	-	X	-	-
<i>Farm</i>	X	-	X	-	-
<i>Owner</i>	X	-	X	-	-

Of the livestock sampled, 0.8% had serological evidence of infection with FMDV. Vaccine-induced immunity to FMDV was highest among cattle (29.0%), then sheep (13.8%) and goats (5.8%), reflecting species-dependent vaccination. Our results fit well with the information provided by the official data for two *oblasts*: Dzhambul and South Kazakhstan, which may indicate that both the results achieved in this study and the official statistics on FMD vaccination are reliable. However, our sampling technique was flawed, additionally the official statistics are not entirely reliable, thus the similarity of the data may be a coincidence. Seroprevalence to brucellosis among sampled livestock was 5.4% among cattle, 1.3% among sheep, and 0.7% among goats. In most *raions* our results were similar to the official data. Considering the likelihood of serious bias in the official data, this may be a coincidence, but could equally suggest that brucellosis is a serious but unacknowledged problem in some areas and is certainly worthy of further study.

Serological evidence of infection with bluetongue virus was found among 25.4% of cattle, 21.4% of sheep and 25.5% of goats. Seropositive animals were found in all six oblasts sampled, indicating that bluetongue is endemic throughout central and southern

Kazakhstan. Bluetongue infection can have significant effects on reproductive performance in sheep, and has been associated with still births and congenital defects (Parsonson, 1991). It may thus be decreasing the productivity of livestock in Kazakhstan. Serological evidence that both EHD and PPR viruses exist among domestic livestock in Kazakhstan was found, but no evidence for infection with rinderpest virus. None of these diseases has been reported in Kazakhstan. The findings, although not conclusive, do justify further investigation.

11.2 Infectious diseases of the saiga antelope

Seroprevalence to *Brucella* spp. in saiga antelopes was found to be within the range previously reported, indicating that brucellosis may be endemic in the Betpak-dala saiga population. It was not possible to identify the species of *Brucella* responsible for infection. There has been no discernible increase in the prevalence of brucellosis among saigas after the independence of Kazakhstan, in contrast to the situation among domestic livestock. This may be related to the dramatic drop in livestock numbers, and the likely consequent decrease in contact between saigas and domestic livestock. This is backed up by the result that no brucellosis was found in the Ustiurt saiga population, which has much less contact with livestock than the Betpak-dala population.

This study found no evidence of infection with FMDV in saiga antelopes, suggesting that the entire saiga population may be susceptible. With only 15.6% of livestock within the saiga range area immune to FMDV, there is a high risk of FMDV transmitting to saiga antelopes in the event of an epidemic near saiga range areas. As this study found evidence of FMDV infection among livestock, and there have been recent reports of FMDV spreading through the south-eastern parts of Kazakhstan, this is of serious concern.

The zero seroprevalence to bluetongue in saigas was highly unexpected, considering the high seroprevalence found by this study in domestic livestock, and the fact that many species of wild ungulates in other countries have been reported seropositive to bluetongue virus. Further work is clearly required on this issue.

11.3 Overall status of the saiga antelope

The saiga population, certainly in Betpak-dala, seems at greater risk than it has been since the early 1920s when the saiga was driven close to extinction by overhunting. Heavy

poaching is a serious and continuing problem. The ban on saiga hunting in 1999 – 2000 will not be effective without funding for law enforcement to prevent poaching. In these circumstances with saiga numbers continuing to dwindle, an outbreak of any major disease could prove disastrous for the population. The Betpak-dala population seems at greatest risk, because it seems to have greater contact with domestic livestock than the Ustiurt population, and because there have been a number of outbreaks of FMD among livestock situated in areas close to saiga range area. Our survey showed that nearly 88% of small ruminants, the livestock type with which saigas are most likely to have contact, are susceptible to infection with FMDV. Nevertheless, our model predicts that an epidemic of FMD is unlikely to persist.

11.4 Policy recommendations

11.4.1 Foot-and-mouth disease

A wide-spread outbreak of FMD would be catastrophic for the livestock industry in Kazakhstan, both internally, and in relation to trade in animal products with other countries. Additionally, an outbreak of FMD among saigas would be devastating if it caused mass mortality as it has done in the past.

Recommendations for prevention of FMD outbreaks among livestock include:

- Vaccination of all cattle, sheep, goats and pigs in *oblasts* with an international border.
- Compulsory testing and certification of all livestock entering Kazakhstan that they are free of FMD. In the future it may be possible to use the recently developed ELISA test that specifically detects antibodies to infection.
- Stringent border controls on movement of animals.
- Implementation of a stamping-out policy, with state compensation to farmers.

Recommendations for prevention of FMD outbreaks among saigas include:

- Vaccination of livestock in areas in and near the saiga ranges.
- Temporary nature reserves in calving areas would prevent contact with livestock at this key time.

11.4.2 Brucellosis

Brucellosis is a disease of serious public health concern, justifying control of the disease among both the domestic and wild host populations. Previous control in livestock was

based on routine surveillance and slaughter of affected animals, but this has broken down recently. There has never been a policy on brucellosis control in saigas.

Recommendations for control of brucellosis among livestock include:

- Serological surveillance, with immediate slaughter of all reactors.
- Reinstatement of full vaccination.
- Keeping livestock in pens when they are due to give birth, combined with disinfection of each pen after use, and careful disposal of the afterbirth to prevent contamination of the environment. This has not been routinely done in the past.

Recommendations for control of brucellosis among saigas include:

- Temporary nature reserves in saiga calving areas to prevent livestock accessing areas where saigas are about to calve.
- Serological testing of saigas could be an option for monitoring purposes in the future, if a countrywide brucellosis eradication campaign was implemented.

11.5 Future work

Although this study had flawed sampling techniques, it has uncovered a number interesting results which merit further research. These include:

- Saiga radio tracking to estimate contact rates, livestock contact, and mortality rates.
- Further development of ageing techniques for saigas, using animals of known age.
- Investigation of the effect of brucellosis on saiga reproduction.
- The role (if any) of saigas in bovine, ovine and human brucellosis.
- Cost benefit analysis of disease eradication programs (FMD and brucellosis).
- Isolation and serotyping of bluetongue, EHD and PPR viruses.
- Identification of the vector of bluetongue, its ecological requirements and distribution in Kazakhstan.
- Investigation into the effect of bluetongue on livestock production.
- Investigation of why saigas are not infected with bluetongue; is there perhaps a particular mechanism that causes the vectors to avoid them? If so, can this be employed to prevent bluetongue infection in sheep?

- Further development of the epidemiological model for FMD transmission among saigas, with extensive use of sensitivity analyses to capture the range of possible parameter values, together with work to improve the parameterisation of the model.
- Linking the epidemiological model with spatial data using a geographical information system.