Contagious Caprine Pleuropneumonia

The rapidity with which the disease had spread among Mr Nierkert’s flock, affecting more than 700 goats within a fourteen day period, induced me to believe that its rapid spread could not be due to contagion alone... but after carefully watching the progress of the disease for three days, and noting the uniform character of the pathological appearances in the lungs, I began to realise that I had a specific disease to deal with (Duncan Hutcheon, Colonial Veterinary Surgeon, Cape of Good Hope, South Africa, 1881).

Introduction

In the autumn of 2002, a severe respiratory disease characterized by high levels of morbidity and mortality was first seen in goat herds in the Thrace region of Turkey, 250 km west of Istanbul (Ozdemir et al., 2005). The disease, which was diagnosed as contagious caprine pleuropneumonia (CCPP), spread throughout the region to an unknown number of herds, some close to the Greek and Bulgarian borders. Goats of all ages were affected and showed a reluctance to walk, a fever of over 41°C and accelerated respiration with frequent coughing. The lungs of dead and euthanized animals showed characteristic lesions with abundant pleural fluid, fibrin and unilateral hepatization; in one herd alone nearly 150 of 400 adults and over 100 of 400 kids died in a single year (Ozdemir et al., 2005). This was the first reported outbreak of CCPP on European soil and today threatens the countries of the EU.

CCPP was first described in 1873 in Algeria and known under the local name of ‘bou frida’ because, in the majority of diseased goats, only one lung was affected (McMartin et al., 1980). Its contagiousness was not initially recognized because the disease was endemic in most areas under investigation, so climatic conditions were thought to be responsible for disease outbreaks. However, a major outbreak in South Africa in 1881 following the introduction of goats from Turkey led the
colonial veterinary surgeon, Duncan Hutcheon, to conclude that CCPP was highly infectious.

Research into the control of CCPP was initially hampered by the uncertainty over the exact cause of the disease. Two mycoplasmas, *M. mycoides* subsp. *mycoides* LC and *M. mycoides* subsp. *capri*, were for some time implicated in the aetiology of the disease because they caused a pleuropneumonia in small ruminants that resembled CCPP. It was not until 1976 that a highly fastidious mycoplasma, designated F38 but later renamed *M. capricolum* subsp. *capripneumoniae*, was isolated for the first time *in vitro* by MacOwan and Minette (1976). Once these workers had developed a suitable medium for the mycoplasma and carried out experimental infections, its role as the primary cause of classical CCPP was confirmed.

However, in spite of this confirmation, respiratory diseases caused by *M. m. capri* and *M. mycoides* LC are still referred to erroneously as CCPP, particularly in the Middle East and India. A condition should only be termed as CCPP when the following criteria have been satisfied:

- *M. c. capripneumoniae* is isolated or there is strong serological evidence of the mycoplasma;
- Lesions are restricted to lung and pleura and consist of a pleuropneumonia;
- The condition is highly contagious with high levels of morbidity/mortality;
- There is no enlargement of the interlobular septa of the lung.

### Geographic Distribution

While the clinical disease has been reported in nearly 40 countries in Africa and Asia, *M. c. capripneumoniae* has only been isolated in 13 countries because few have the facilities for isolating and growing mycoplasmas (Nicholas, 2002) (Table 9.1). Serious problems exist in Oman, where nearly 500 outbreaks were reported in 2006 with a mortality rate of nearly 10% from 15,000 cases, and in Iran, where nearly 300 outbreaks have affected over 13,000 goats (OIE, 2008). The 31 reported outbreaks in Ethiopia almost certainly represents an underestimate, as this disease is having a big socio-economic impact here.

Table 9.1. Distribution of CCPP.

<table>
<thead>
<tr>
<th>Confirmed by isolation of mycoplasma</th>
<th>Clinical disease reported or suspected</th>
</tr>
</thead>
<tbody>
<tr>
<td>Africa: Chad, Eritrea, Ethiopia, Kenya, Niger, Sudan, Tunisia, Uganda</td>
<td>Algeria, Benin, Burkina Faso, Cameroon, Central African Republic, Djibouti, Egypt, Libya, Mali, Nigeria, Somalia, Zaire</td>
</tr>
<tr>
<td>Asia: Nepal, Oman, Turkey, United Arab Emirates, Yemen</td>
<td>Afghanistan, Bangladesh, India, Iran, Iraq, Israel, Jordan, Kuwait, Lebanon, Pakistan, Saudi Arabia, Syria</td>
</tr>
<tr>
<td>Europe: Thrace (Turkey)</td>
<td></td>
</tr>
</tbody>
</table>
In 2003, CCPP was diagnosed in Thrace, the region of Turkey on the European mainland bordering Greece and Bulgaria. Prior to this infection, the only previous report of CCPP in Europe dates to the 1920s, when an outbreak occurred in Greece following the seizure of goats from Turkey. Interestingly, Greece reported two outbreaks of CCPP in 2006, but it seems likely that this was caused by *M. m. capri*, which is endemic in Greece, rather than by *M. c. capripneumoniae*, as less than 1% of goats died in a herd of over 150. It is probable also that the outbreak of CCPP in the Czech Republic in 1902 was similarly misdiagnosed, although it is clearly impossible to confirm over a century later. There have been no reports of the isolation of *M. c. capripneumoniae* on the American continent, although other members of the *mycoides* cluster have been described there.

**Causative Agent**

The taxonomic status of F38 has long been unclear, and it was only in 1993 that it became a subspecies of *M. capricolum* and classified as *M. capricolum* subsp. *capripneumoniae* (Leach et al., 1993). Five distinct groups of mollicutes have been identified by phylogenetic analysis of the 16S rRNA sequences, one of which, the spiroplasma group, contains *M. c. capripneumoniae*, which has been subdivided within the *M. mycoides* cluster. This cluster contains six important ruminant mycoplasmas, including *M. m. mycoides* SC, the cause of contagious bovine pleuropneumonia, *M. m. mycoides* LC and *M. m. capri*, which share immunological and biochemical properties. Their close relationship can lead to problems for diagnosis. Tables 9.2 and 9.3 summarize the properties of *M. c. capripneumoniae* and some members of this cluster as well as other mycoplasmas capable of causing diseases in small ruminants.

**Host Susceptibility**

For a long time goats were believed to be the only susceptible host for CCPP (Litamoi et al., 1990), although it was reported that sheep could be infected and seroconvert in the face of exposure (Bolske et al., 1995). Following the introduction of CCPP into Eritrea with the livestock of returning refugees from Sudan, sheep mixing with affected goats were reported to be suffering respiratory disease (Houshaymi et al., 2000). More recently there have been confirmed reports from Qatar of CCPP in captive wild ungulates, including wild goat, Nubian ibex, Laristan mouflon and gerenuk, kept in animal breeding parks (Arif et al., 2007). Even more surprisingly, however, were the outbreaks of acute respiratory disease in a private collection of captive but free-ranging gazelles and other deer species in the United Arab Emirates, in which over 10% died. The disease was almost certainly introduced via sick goats and spread by close contact with the gazelles at feed stations (Nicholas et al., 2008); it is likely that CCPP is far more widespread in wildlife species in the Middle East as a result of infected escapees from these parks.
Table 9.2. Mycoplasmas, including *M. capricolum* subsp. *capripneumoniae*, isolated from small ruminants with respiratory disease.

<table>
<thead>
<tr>
<th>Mycoplasma</th>
<th>Host(s)</th>
<th>Primary site of isolation (other)</th>
<th>Disease*</th>
<th>Pathogenicity</th>
<th>In vitro growth</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>M. c. capripneumoniae</em></td>
<td>Goat (sheep)</td>
<td>Lungs</td>
<td>CCPP</td>
<td>High</td>
<td>Slow (5–7 days)</td>
</tr>
<tr>
<td><em>M. m. mycoides LC</em></td>
<td>Goat (sheep, cattle)</td>
<td>Resp. tract (udder, joints)</td>
<td>Plp, M, A, C</td>
<td>Moderate</td>
<td>Fast</td>
</tr>
<tr>
<td><em>M. m. capri</em></td>
<td>Goat (sheep)</td>
<td>Resp. tract (joints)</td>
<td>Plp, A, C</td>
<td>Moderate</td>
<td>Fast</td>
</tr>
<tr>
<td><em>M. c. capricolum</em></td>
<td>Goat, sheep</td>
<td>Joints/resp. tract (udder)</td>
<td>Plp, M, A</td>
<td>High</td>
<td>Fast</td>
</tr>
<tr>
<td><em>M. ovipneumoniae</em></td>
<td>Sheep, goat</td>
<td>Resp. tract</td>
<td>P</td>
<td>Low</td>
<td>Moderate</td>
</tr>
<tr>
<td><em>M. conjunctivae</em></td>
<td>Sheep, goat</td>
<td>Eyes</td>
<td>KC</td>
<td>Moderate</td>
<td>Moderate</td>
</tr>
<tr>
<td><em>M. agalactiae</em></td>
<td>Sheep, goat</td>
<td>Udder (joints, eyes)</td>
<td>M, A, KC, P</td>
<td>High</td>
<td>Fast</td>
</tr>
<tr>
<td><em>M. putrefaciens</em></td>
<td>Goat (sheep)</td>
<td>Udder (joints)</td>
<td>M, A</td>
<td>Moderate</td>
<td>Fast</td>
</tr>
<tr>
<td><em>M. arginini</em></td>
<td>Ubiquitous</td>
<td>Resp. tract</td>
<td>None</td>
<td>Low/non-pathogenic</td>
<td>Fast</td>
</tr>
</tbody>
</table>

Table 9.3. Major biochemical differences between mycoplasmas of small ruminants.

<table>
<thead>
<tr>
<th>Mycoplasma</th>
<th>Glucose fermentation</th>
<th>Arginine hydrolysis</th>
<th>Phosphatase activity</th>
<th>Film and spots</th>
<th>Casein digestion</th>
<th>Tetrazolium aerobic</th>
<th>Tetrazolium anaerobic</th>
<th>Reduction</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>M. c. capripneumoniae</em></td>
<td>+/−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>+</td>
<td>varies</td>
<td>weak/+</td>
<td></td>
</tr>
<tr>
<td><em>M. m. mycoides LC</em></td>
<td>+</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td><em>M. m. capri</em></td>
<td>+</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td><em>M. c. capricolum</em></td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>−</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td><em>M. ovipneumoniae</em></td>
<td>+</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>varies</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td><em>M. conjunctivae</em></td>
<td>+</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>+</td>
</tr>
<tr>
<td><em>M. agalactiae</em></td>
<td>−</td>
<td>−</td>
<td>+</td>
<td>+</td>
<td>−</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td><em>M. putrefaciens</em></td>
<td>+</td>
<td>−</td>
<td>+</td>
<td>+</td>
<td>−</td>
<td>varies</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td><em>M. arginini</em></td>
<td>−</td>
<td>+</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>+</td>
</tr>
</tbody>
</table>
Clinical Signs and Pathology

Goats of all ages and sex can be affected (Thiaucourt and Bölke, 1996). The acute disease is more noticeable in naive populations in newly affected areas, with high mortality and morbidity rates. The incubation period generally lasts on average 10 days but may vary between 2 and 28 days. The first signs are a reluctance to walk and the onset of fever, typically 41°C and occasionally 42°C, although animals continue to feed and ruminate. Respiration accelerates and becomes painful, with violent coughing episodes. Animals stand with limbs abducted and neck extended (Fig. 9.1). There is continuous salivation and noses become blocked with a mucopurulent discharge. In the terminal stages the goats are unable to move and death follows quickly. In subacute or chronic forms, signs are milder, with coughing usually noticeable only following exercise. High mortality can be seen in kids, where death is usually the result of septicaemia (Fig. 9.2).

The best description of the pathology of the disease was made by the colonial surgeon Hutcheon (1889) writing over a century ago: ‘There is no thickening of the interlobular tissue in the diseased lung of the goat, which forms such a striking feature in bovine pleuropneumonia; the section of the diseased lung in the goat has the appearance of a somewhat granular-looking liver’. These features also clearly differentiate disease caused by *M. m. capri* and *M. m. mycoides* LC. In short, the pathological lesions are localized exclusively in the lung and pleura and consist of a pleuropneumonia, unilateral hepatization, adhesions, pleuritis and an accumulation of pleural fluid (Fig. 9.3). The pleural exudate can

Fig. 9.1. Goat with acute CCPP showing head lowered and limbs abducted.
solidify to form a gelatinous covering on the lung (Fig. 9.4). Hepatization of the lung as Hutcheon describes is a key feature for differential diagnosis (Fig. 9.5).

A study to correlate clinical signs and early lesions showed that affected goats killed up to a week after contact with affected animals were free of lung lesions or
clinical signs; between 2 and 3 weeks after contact, lung lesions were generally small and superficial, characterized by hyperaemia and oedema with clinical signs being restricted to an infrequent cough; fever was first seen after nearly 4 weeks, which correlated with lung consolidation, the area of which increased as the fever progressed (Wesonga et al., 1993).

Fig. 9.4. Lung of goat with CCPP showing fibrin deposition.

Fig. 9.5. Lung of goat with CCPP showing granular hepatization of the cut section.
In an experiment by Ozdemir et al. (2006), it was possible to follow the course of the disease, with and without antibiotic intervention. Healthy Angora goats, confirmed free of CCPP, were exposed to clinically affected animals from a natural outbreak in Thrace, Turkey. After 14 days' exposure, the majority of contacts showed pyrexia ($\geq 41^\circ C$). Shortly after, the Angora goats were randomly divided into two groups. Half of these were injected twice over 2 days with danofloxacin; the remaining animals received saline. Goats which survived were euthanized at day 42. All danofloxacin-treated goats showed resolution of clinical disease by the end of the trial. Two saline-treated goats were euthanized after 4 weeks due to severe disease. Danofloxacin-treated goats showed fewer lung lesions, which were generally sequestrated (Fig. 9.6), and had significantly lower combined clinical scores than saline controls, which showed severe and acute lesions.

Fig. 9.6. Lung of goat with CCPP, treated with antibiotics, showing sequestrum.
The outbreaks of CCPP in gazelles in the Middle East had a very similar clinical (Fig. 9.7) and pathological (Fig. 9.8) appearance to that seen in goats.

Histological examination of the lung tissues often reveals an acute serofibrinous to chronic fibrino-necrotic pleuropneumonia, with infiltrates of serofibrinous fluid and inflammatory cells, mainly neutrophils, in the alveoli, bronchioles, interstitial septae and subpleural connective tissue. Intralobular oedema is more prominent, but interlobular oedema has also occasionally been reported. Peribronchial and peribronchial lymphoid hyperplasia with mononuclear cell infiltration is also present (MacOwan and Minette, 1976; Kibor, 1990; Wesonga et al., 1998).

Ultrastructural examination of the lungs of goats naturally affected with CCPP confirms histopathological findings of congested septal capillaries, with inflammatory cells invading thickened septal walls (Johnson et al., 2002). The alveolar lumen contains serous fluid mixed with neutrophils and lymphocytes, some of which are necrotic. The most significant findings are a widespread hyperplasia of type II pneumocytes that have lost most of their characteristic lamellar ultrastructure and large numbers of mycoplasma-like structures lying close to the microvilli and membranes of these cells. It was proposed that the loss of these lamellae may reduce surfactant production, as well as synthesis of key enzymes, leading to increased surface tension within the alveoli and contributing to the atelectasis often seen at post-mortem examination of CCPP cases.

Conditions which may exacerbate CCPP include concurrent viral infections, in particular orf and pestes petits de ruminant (PPR), and possibly other mycoplasma
infections such as *M. ovipneumoniae*, a cause of disease in its own right; adverse weather conditions and stress caused by transhumance may also compound or accelerate ongoing disease.

**Immunology**

Little is known of the immunology of CCPP despite a number of reported experimental infections (Muthomi and Rurangirwa, 1983; Perreau et al., 1984). More recently March et al. (2000) monitored the humoral response of goats infected with a multipassaged *M. c. capripneumoniae* strain 19/2 with several serological tests and PCR. While there was little evidence after infection of the infectious agent or clinical or pathological disease, apart from elevated temperatures and a transient cough in one goat, serological responses were detected by latex agglutination test and competitive ELISA. IgG immunodominant bands of 23, 40 and 44 kDa were seen by immunoblotting in all experimentally infected animals, as well as in some sera from a natural outbreak of CCPP in Eritrea, which additionally showed bands of 62, 70 and 108 kDa.

**Transmission**

Outbreaks follow the introduction of an infected animal into a susceptible herd. The mycoplasma is transmitted over short distances through the expulsion of
infected droplets during coughing. The disease is very readily contagious and only brief periods of contact are necessary for successful transmission (Thiaucourt and Bölske, 1996). A single surviving seropositive goat from a natural outbreak in Thrace was able to infect all ten disease-free contacts within 2 weeks (Ozdemir et al., 2006). However, the infectious period is quite short as a second contact infection from the same region the following year failed to transmit disease to healthy contacts despite the seropositivity of the naturally infected goats.

No evidence of indirect transmission has been shown as the mycoplasma is highly fragile in the environment. As with many mycoplasma diseases, in particular contagious bovine pleuropneumonia, the disease is introduced into a region by clinically healthy carrier animals.

**Molecular Epidemiology**

Unlike other members of the *M. mycoides* cluster, *M. c. capripneumoniae* shows a surprising degree of heterogeneity, particularly in the sequence of its two rRNA operons, which both contain the genes for 5S, 16S and 23S rRNA (Pettersson et al., 1996). These polymorphisms, representing point mutations in either gene, can be used to subtype strains, some of which may have epidemiological and, possibly, evolutionary significance. Sequencing the 16SrRNA genes of African strains identified two distinct lines, I and II, both of which were represented in Central, North and East Africa; isolates from the Middle East were of the type II only, although they could be further divided (Pettersson et al., 1998; Heldtander et al., 2001). Strains isolated from Thrace in Turkey are placed in the groups II and IIb, which suggests that they were introduced illegally across the Dardanelles from Asian Turkey, while those from gazelles are very similar to goat isolates from neighbouring Oman (Nicholas et al., 2008).

Sequencing the amplified products of a different gene(s), the H2 locus, Lorenzon et al. (2002) divided strains into four major groups, in which lineage A occurred exclusively in East Africa, B mostly in North Africa and the Middle East, C in Central Africa, and D, represented by only a single strain, from the United Arab Emirates. Subtyping with amplified fragment length polymorphism (AFLP) strongly supported the 16S rRNA sequence analysis by identifying two main lineages (Kokotovic et al., 2000).

On a more local level, ten of 11 strains of *M. c. capripneumoniae* isolated from four different regions of Tanzania had very similar profiles with AFLP. These profiles were also indistinguishable from two Kenyan and one Ugandan strain, indicating the close association between small ruminants in these three neighbouring countries (Kusiluka et al., 2000). Using the PCR/DGGE, it was possible to differentiate strains from Eritrea from all others by their distinctive pattern (McAuliffe et al., 2005).

Isolates from gazelles with CCPP in the UAE (Nicholas et al., 2008) were similar to those from Thrace, Turkey but dissimilar to the type strain F38 and those from Eritrea and mainland Turkey, suggesting disease may have come from contact with affected goats; however, further work is needed to compare with strains from other parts of the UAE.
Substantial diversity was reported in the metabolism of strains of *M. c. capripneumoniae* (Abu-Groun *et al.*, 1994). In an extension of this work, Houshaymi (1999) divided a range of strains into two major groups, which was also confirmed by DNA–DNA hybridization patterns. Some strains, including the type strain F38, only oxidized organic acids and glycerol but not glucose, while others, including strains from Eritrea, metabolized glucose. The patterns of substrate utilization shown by the non-glucose-oxidizing strains were similar and had a high affinity for 2-oxybutyrate; those for the glucose-metabolizing strains were also similar but failed to oxidize fructose and had a low affinity for 2-oxybutyrate.

Such biochemical diversity within a *Mycoplasma* species is unique and may present diagnostic problems as glucose fermentation is often a key characteristic in their preliminary identification. However, even with glucose-metabolizing strains, the addition of pyruvate to the medium leads to significantly higher yields *in vitro* (Houshaymi *et al.*, 2002). Thus it may be that organic acids are also important energy sources for glucose-oxidizing strains. Table 9.4 illustrates the diversity of strains of *M. c. capripneumoniae* in their substrate requirements.

### Table 9.4. Substrate utilization by strains of *M. c. capripneumoniae*.

<table>
<thead>
<tr>
<th>Substrate</th>
<th>19/2 Oman</th>
<th>T3 Eritrea</th>
<th>G183 Kenya</th>
<th>F38 Kenya</th>
<th>7/1a Turkey</th>
<th>G1943 Kenya</th>
<th>G94/83 Kenya</th>
<th>4/2 LC Oman</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lactate</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
</tr>
<tr>
<td>Pyruvate</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>Glucose</td>
<td>+++</td>
<td>++</td>
<td>+++</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>+++</td>
</tr>
<tr>
<td>Glycerol</td>
<td>–</td>
<td>+++</td>
<td>+++</td>
<td>–</td>
<td>+++</td>
<td>–</td>
<td>–</td>
<td>+++</td>
</tr>
<tr>
<td>2-oxybutyrate</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>–</td>
</tr>
</tbody>
</table>

### Biochemistry

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### Diagnosis

In spite of greatly improved media formulations, the isolation of *M. c. capripneumoniae* remains one of the more difficult tasks for the mycoplasma diagnostic laboratory. The samples of choice are the pleural fluid, which contains high numbers of mycoplasmas, and sections of hepatized lung, preferably at the interface of normal and diseased tissue; however, attempts to isolate mycoplasmas from pleural fluid from goats dying in Thrace were unsuccessful for reasons which remain unclear but may be the result of excessive use of antibiotics. Samples must be sent quickly in a cool condition but will become of little value if journey time is longer than 2 days. Sending samples frozen is recommended but not always practical. During the recent investigation of CCPP in Eritrea, excellent isolation rates of *M. c. capripneumoniae* were achieved from lyophilized lung samples even though isolation was not carried out for several weeks after arrival (Houshaymi *et al.*, 2000). Choice of medium is critical and best results were obtained during the
same investigation with a commercial medium (Mycoplasma Experience, Reigate, UK) (Houshaymi et al., 2002). A diagnostic medium for CCPP has also been developed by this company in which *M. c. capripneumoniae* develops coloured colonies in semi-solid medium (Fig. 9.9). Other media which have been shown to support the growth of most strains of *M. c. capripneumoniae* include H25P (Nicholas and Baker, 1998) or FP medium supplemented with 2 g/l of sodium pyruvate (Bölske et al., 1996). Overgrowth of this fastidious mycoplasma by other mycoplasmas is another major problem with isolation. In particular, the ‘centreless’ colonies of *M. ovipneumoniae* will grow at a much faster rate but can be separated from *M. c. capripneumoniae* by early cloning.

The development of PCR has greatly improved CCPP diagnosis as it is now possible to detect the mycoplasma quickly, even in mixed cultures, directly from clinical material such as pleural fluid and lung, and from this material dried on filter paper (Lorenzon et al., 2002). PCRs based on the 16S RNA genes have been reported that enable the detection of all members of the *M. mycoides* cluster followed by specific identification of *M. c. capripneumoniae* by restriction enzyme digestion (Bölske et al., 1996). A specific PCR which does not require a restriction enzyme step was described by Woubit et al. (2004). The PCR/denaturing gradient gel electrophoresis offers great advantages in detection because it is sensitive, rapid and specific: a single pair of mollicute-specific primers can amplify DNA from all species, which can then be identified by their migration pattern following DGGE (McAuliffe et al., 2005). The test also uniquely detects more than one

**Fig. 9.9.** Colonies of *Mycoplasma c. capripneumoniae* stained with specific stain (Mycoplasma Experience, Reigate, UK).
Mycoplasma species in a sample. However, because of the close similarity of the sequence of the 16S rDNA gene, the target of the PCR/DGGE, in the M. mycoides cluster members, a confirmatory PCR such as that described by Woubit et al. (2004) should be performed.

Serodiagnosis of CCPP, on the other hand, is a relatively easy task, thanks to a rapid, specific and relatively sensitive test developed initially in Kenya. The latex agglutination test (LAT) uses a carbohydrate extracted from M. c. capripneumoniae linked to latex particles which agglutinate in the presence of specific antibodies in the blood of affected goats (Rurangirwa et al., 1987). The test, which takes minutes to complete, is more sensitive than the complement fixation test and easier to perform than the CFT (OIE, 2004) or indirect or competitive ELISA, which should be used for confirmation (Houshaymi et al., 2002). An LAT has also been described for circulating antigen and could provide earlier detection in affected animals before antibodies have appeared (March et al., 2000). The test also provides a convenient means of monitoring growth of the mycoplasma in vitro, which is often quite difficult to detect because of the small pH changes produced by some strains; the test requires just a few drops of culture fluid.

Disease Prevention and Control

Protection against CCPP was shown to be possible more than a century ago when Hutcheon subcutaneously inoculated goats with lung extract from affected animals (McMartin et al., 1980). Later, goats vaccinated with an attenuated broth culture of F38 did not succumb to contact infection (MacOwan and Minette, 1978). This clearly demonstrated control was possible. Since then a number of different preparations have been produced which are reported to produce strong immunity even after 1 year. These include a vaccine composed of sonicated antigens emulsified with incomplete Freund’s adjuvant (Rurangirwa et al., 1984) and another in which lyophilized F38 was inactivated with saponin immediately before immunization (Rurangirwa et al., 1987). The latter vaccine has been in use in Kenya for the last few years but this somewhat dangerous practice was modified so that the mycoplasma was inactivated with saponin for at least 12 h at 4°C, enabling successful inactivation to be checked (OIE, 2004). Kids older than 10 weeks of age are vaccinated to avoid maternal antibody, although there is little evidence to show such interference.

In countries where vaccination is not practised, other control measures are used. Antibiotics such as the tetracyclines, fluoroquinolones and the macrolide family are generally effective clinically if used early enough (Onovarian, 1974; Hassan et al., 1984; Ozdemir et al., 2006). However, the complete elimination of the mycoplasma is rarely achieved and treated animals should always be considered as potential carriers.

Movement restrictions and slaughtering of infected and contact animals are recommended for countries or regions that are newly infected.

The risk of introducing CCPP to the USA is probably very small as it does not import small ruminants from Asia or Africa. There is, however, a risk of introducing CCPP to those EU states bordering countries like Turkey, where disease is
endemic, or from Eastern Europe, where surveillance for CCPP rarely occurs. Italy may also be threatened from North Africa, particularly Tunisia and Libya, which are only a short boat ride away. Once introduced into the EU, the disease could theoretically spread throughout the member states, including the UK, via animal trade. The widespread use of antibiotics would suppress the overt clinical signs, reducing the direct economic consequences, with the disease remaining undetectable for several months or years, as was the case in Italy following the introduction of contagious bovine pleuropneumonia in 1990. However, the costs of eradication would be significant. The key to control of this disease, therefore, given an outbreak in Europe, would be the rapid identification of the disease, enabling the destruction of affected and contact animals.

References


Contagious Caprine Pleuropneumonia


