CONTAGIOUS BOVINE PLEUROPNEUMONIA (CBPP) IN TANZANIA

[1]Minga, U.M., [2]Kazwala, R.R., [3]Matovelo, J.A., [1]Gwakisa, P.S., [4]Melewas, J.N.
[1]Department of Veterinary Microbiology and Parasitology, Faculty of Veterinary Medicine, SUA, Box 3019, Morogoro;
[2]Department of Veterinary Medicine and Public Health, Faculty of Veterinary Medicine, SUA, P.O. Box 3021, Morogoro;
[3]Department of Veterinary Pathology, Faculty of Veterinary Medicine, SUA, P.O. Box 3018, Morogoro;
[4]Ministry of Agriculture and Cooperatives, Kilimo House, P.O. Box 9152, Dar es Salaam

ABSTRACT

Tanzania was free from contagious bovine pleuropneumonia (CBPP) form 1965 up till 1990. In May 1990 a serious CBPP outbreak appeared in Loliondo, Arusha, and had crossed from Southern Kenya. Another CBPP outbreak, thought to have crossed from Southern Uganda, occurred in 1992 in Kagera region. To date the disease has spread to 49 districts in 17 regions in Tanzania mainland. Tanzania has lost over 200,000 cattle due to CBPP, and has incurred a loss of over 16 billion shillings or US dollars 20 million. Tanzania did not have the capacity to confirm CBPP by culture methods and factors responsible for the re-surgence and spread of CBPP are not known. There was need therefore to establish the capacity to culture and identify Mycoplasmas and to determine factors associated with the re-surgence and spread of CBPP.

An epidemiological study was conducted between 1997 and 1999 in order to establish the capacity to culture and identify Mycoplasma isolates from CBPP suspect cases and determine factors responsible for the re-surgence and spread of CBPP in Tanzania. The capacity to culture Mycoplasma was established and over 13 CBPP cases were confirmed by culture of Mycoplasma. It was found out that although Government planned to control CBPP through imposition of quarantines and vaccination, the quarantines were found to be ineffective, cattle movements were virtually uncontrolled, the system of vaccination through cost recovery undermined vaccination coverage and there was uncontrolled use of antibiotics for the treatment of CBPP. Although serious disease outbreaks have stopped, the disease was now endemic in Tanzania.

It is suggested that efforts must be sustained in surveillance and monitoring of CBPP both serologically and abattoir surveillance. That such continuous surveillance would ensure the early detection of new CBPP outbreaks and occurrence of new cases, and would guarantee early and timely control of the disease. That the Government ought to increase funding to the livestock division if veterinary extension services are to operate efficiently and effectively and diseases controlled. It will be difficult for Tanzania to eradicate CBPP at this stage, the alternative would be vaccination free of charge like it is with rinderpest.

INTRODUCTION

Contagious bovine pleuro-pneumonia (CBPP) is a highly contagious pneumonia which can be acute, subacute or chronic and is characterized by dyspnoea and coughing and loss of weight (Schneider *et al*., 1994; Blood and Radostits, 1989). Morbidity varies from as low as 10% to as high as 100%. The mortality is around 50% and about 25% of the animals which recover remain carriers and may transmit the disease to susceptible animals (Blood and

Radostits, 1989; Fraser *et al* 1991). The disease is characterized pathologically by serofibrinous pleuritis and pericarditis and by interlobular interstitial thickening of the lung tissue giving a characteristic marbling appearance (Schneider, 1994).

CBPP is caused by *Mycoplasma mycoides* subsp. *mycoides* small colony type (sc). The disease occurs in India, China, Italy, Portugal and many parts of Africa including Tanzania, Uganda, Kenya, Angola, Ethiopia, Cameroon, Namibia, Nigeria, Somalia and Sudan (Schneider et al., 1994; Nwanta and Umoh, 1992; Trichard *et al*., 1989; Guadagnini *et al*., 1991; Hehnen,1991). The disease is reported to be endemic in neighbouring Kenya and Uganda.

From 1965, Tanzania was free from CBPP (Hammond and Branagan, 1965). The disease however re-appeared in Arusha, Tanzania in May, 1990 and is claimed to have crossed from Kenya, and in Kagera CBPP re-appeared in 1992 and had originated from Southern Uganda (Msami, 1990, Minga, et al., 1993, Boelske et al., 1995). Due to uncontrolled cattle movement, the disease had since December 1995 until May, 1999 been recorded in and spread to seventeen regions viz. Arusha, Kagera, Shinyanga, Singida, Morogoro, Tabora, Rukwa, Kigoma, Iringa, Mbeya, Ruvuma, Dodoma, Lindi, Coast, Tanga, Mara and Dar es Salaam. Over 200,000 cattle have died so far and over 7 million cattle are at risk (Epidemiology Unit, Ministry of Agriculture and Cooperatives). CBPP has cost the nation, through deaths alone, over T.Shs. 16 billion which is equivalent to Us dollars 20 million.

It was the intention of the Government to control the disease by vaccination. It was claimed that the disease had been successfully controlled in Kagera by vaccination. However there were reports of resurgence. This discrepancy could be attributed to vaccination failure, unfortunately however, the efficacy and sero-conversion were not monitored (Epidemiology Unit, Ministry of Agriculture and Cooperatives).

Tanzania lacks an established capacity to confirm CBPP by culture methods. It also lacks the capacity to monitor vaccination success and to seroscreen. Confirmation of the disease has so far been done in Kenya. In Tanzania, diagnosis of the disease has relied mainly upon clinical signs and pathological lesions. However, similar clinical signs may be manifested by pneumonia due to *Pasteurella hemolytica* infection and haemorrhagic septicaemia due to *Pasteurella multocida* infection.

The commonest and standard sero-diagnostic method employs the complement fixation test (CFT) and is the reference method recognized by the Office International des Epizooties (0IE). The test is negative two months after vaccination and hence for the vaccinated herd, it can be safely used as a serodiagnostic test two months post-vaccination (Schneider *et al.*, 1994). The newer diagnostic techniques such as the ELISA and Polymerase Chain Reaction (PCR) have added advantage of high sensitivity and specificity and are now gaining popularity.

Diagnostic techniques and vaccine development for CBPP was neglected by Tanzania all along mainly because, during the East African Community days research on CBPP was done at the then EAVRO as a common service for East Africa and also because CBPP was controlled for over a quarter century from 1965. The outbreak of CBPP in 1990 therefore caught Tanzania unprepared.

There was need therefore to establish sensitive and specific diagnostic methods in Tanzania. Such methods would allow for diagnosis, seroscreening and confirmation of CBPP using culture, serological and DNA based methods. The same methods would be adapted for the diagnosis of other important animal mycoplasma diseases viz. Contagious caprine pleuropneumonia and Chronic respiratory disease of fowl. The study would greatly complement control efforts by Government.

The main objective of the research was to study the epidemiology and control of Contagious Bovine Pleuropneumonia (CBPP) in selected regions of Tanzania and recommend appropriate and effective control methods. Specifically, the purposes were to establish the capacity to culture and identify Mycoplasma from CBPP suspect cases, to determine factors which had led to the spread of CBPP outbreaks in Tanzania and to determine the prevalence of Contagious Pleuropneumonia in Morogoro Mbeya and Iringa regions.

Mycolasma would be cultured using specific Mycoplasma media and identified using the growth inhibition method. Epidemiological factors associated with the spread of CBPP would be determined by the Partipatory Rural Appraisal (PRA) methods. CFT would be used both for the seroscreening of infected cattle and seromonitoring the vaccinated herds. The ELISA and DNA based methods would be conducted to complement the CFT and compare relative specificity and sensitivity (Taylor *et al*., 1992, Bashirudin *et al*., 1994; Ball *et al.*, 1994; Dedieu et al 1994; Brocchi *et al.*, 1993).

Finally it shall be possible to formulate appropriate and effective control methods for CBPP and recommend the same to farmers and Government and it shall be possible to develop a sensitive and specific diagnostic method for CBPP and other Mycoplasma diseases and to evaluate the efficacy of CBPP vaccine (s) currently in use.

MATERIALS AND METHODS

Study Area

The research was conducted from February, 1998 until July, 1999 in four regions which had experienced outbreaks of CBPP and which were easily accessible to the Project (Morogoro, Iringa, Mbeya and Dodoma), Dodoma was initially not included in the plan as an infected area but rather as a CBPP free area until when CBPP and CCPP cases were reported in late 1998.

Epidemiological factors which might have led to the occurrence of CBPP outbreak and persistence were studied, thus: Cattle movement, management practices, veterinary intervention measures, traditional disease management by the pastoralist related to CBPP, treatment, and observance of quarantines. The information was obtained by Participatory Rural Appraisal (PRA) method and actual on site observation.

Field Visits

Field visits were made to locations where there were reports of CBPP and CCPP outbreaks and cases in Morogoro, Iringa, Mbeya and Dodoma regions in following locations: Morogoro: Morogoro abattoir; Wami Mkongeni, Mikese, Melela, Kimamba, Kilosa town; Malolo, Gairo, Kilosa; Ifakara abattoir, Kilombero and Mtimbira; Dodoma: Mpwapwa and

Kongwa districts; Mbeya: Mbarali district; Mbeya town and Chunya; Iringa: Idodi and Iringa VIC.

Epidemiological data, CBPP cases, Specimen collection and Transportation of specimen:

Information on the history of disease outbreaks and occurrence of cases in the location visited was obtained through interview of the local veterinary extension workers and the cattle owners using the PRA method.

A follow up was made on any new CBPP cases which were reported from the four regions. Later CCPP suspect cases were also followed up. CBPP suspect cases were examined clinically for respiratory and other signs indicative of CBPP. Whenever possible and with the permission of the owner, cases were sacrificed and postmortem examination conducted. Lung and lymph nodes and other tissues showing CBPP like lesions were taken for Mycoplasma culture and for detection of Mycoplasma using DNA based methods. Tissues were kept in a cold box with ice cubes. If the location was far from SUA, the specimens were first frozen in a freezer and transported frozen on the day on travel. At the SUA laboratory the samples were kept frozen in a freezer until cultured for Mycoplasmas. Abattoir specimen and sequestra were obtained during routine meat inspection. Blood was taken from cases as well as from in-contact cattle and serum harvested the following day. The serum was for testing for anti-*Mycoplasma mycoides* subsp. *mycoides* antibodies using the CFT and ELISA.

In early 1999, there were reports of outbreaks of contagious caprine pleuropneumonia (CCPP) and lung, lymph node specimens and blood were taken from goats suspected to have suffered from CCPP.

Altogether, 38 samples were collected for Mycoplasma isolation as follows: Twenty three samples from CBPP suspect cases were collected for Mycoplasma isolation, from Morogoro region in Kilosa district (2 samples), Morogoro town abattoir (1 sample) and Ifakara town abattoir (4 samples), from Mbeya region at Mbarali (7 samples) and Chunya (1 sample) and from Iringa region at the VIC (8 samples). Thirteen samples were collected from CCPP suspect cases from Morogoro region at SUA (1 sample) and at Gairo (4 samples), Iringa region at the VIC (4 samples) and from Dodoma region in Mpwapwa and Kongwa districts (4 samples). Two samples of chronic respiratory disease (CRD) from a chicken was collected from SUA and Msolwa in Morogoro region.

The isolated Mycoplasmas were stocked on Mycoplasma agar and kept frozen ready for serotyping and for typing using DNA based method namely the Random Amplified Polymerase chain reaction (RAPD-PCR) (Gwakisa *et al.*, 1994), and then characterised for unique DNA profiles.

Mycoplasma Culture, Isolation and Identification

Tissue specimens were either ground in a sterile mortar using a sterile pestle and sand or cut into small pieces using a sterile pair of scissors and then cultured directly on selective Mycoplasma Tryptose agar medium containing penicillin and thallium acetate. In case of sequestra, specimens were first enriched in Mycoplasma Tryptose broth and then subcultured on solid Mycoplasma Tryptose agar. The plates were incubated at 37°C in a humid chamber

for at least three days before checking for colonies under low power microscope. Plates were discarded if by two weeks no growth was detectable.

The growth inhibition test was used for the identification of the *Mycoplasma* isolates using species specific typing anti-sera.

RESULTS

Field visits and Epidemiological data

Data was collected from the villages which were visited. This included the history of CBPP cases and local control measures, that is, quarantines, vaccinations and treatments.

Disease outbreaks in new areas

CBPP outbreaks in formerly disease free areas were due to cattle being moved from one area to another mostly illegally, either through cattle rustling, movement without a permit or movement with permit issued by non-professional people. Reasons for moving cattle from one area to another varied, the majority of pastoralists moved cattle for search of better pasture or water, to pay bride prize or for other rituals and ceremonies. It was found out that in other cases it was due to fear of witchcraft, the owner thinking that the massive cattle deaths was because the herd was bewitched by neighbours and hence to avoid further deaths, it was logical to move as far away from the suspected witch as possible. There was a case where one moved with his herd from Sumbawanga to Chunya and still deaths continued to occur.

Introduction of CBPP in a susceptible herd

Almost all cases CBPP occurred among pastoralist herds. In all incidences, new cases were connected with the introduction of cattle from other herds, such as the owner bringing in animals from his own boma located elsewhere, buying in animals, keeping animals for other people who are on transit and acquiring new animals as bride price or dowry. In Morogoro region and may be elsewhere, pastoralist move there herds almost annually from low grounds to high grounds and then back again. Some pastoralists spread the risk of losing their animals by maintaining two or more herds in two or more far off locations.

Disease control measures: Quarantines

CBPP is a notifiable disease and hence quarantines were imposed whenever an outbreak occurred. The quarantines in many cases were ineffective because of a number of reasons. In some cases quarantines were interfered with by local authorities on account of loss of revenue for the local government if cattle markets were to be closed. One such case occurred in Mbarali. In another case, a businessman appealed to higher authorities to allow him to transport cattle from an upcountry district which was under quarantine to Dar es Salaam. The biggest problem however, was the pastoralists themselves. There is considerable nomadic pastoralism in Tanzania, whereby cattle are moved across districts, regions and countries for a number of reasons as stated above. It was difficult for the veterinary extension staff to control such animal movement because in many cases they do not use the official routes or

corridors. In many cases the owners move their animals at night which is difficult to monitor and control.

Disease control measures: Treatment

According to regulations cattle suffering from CBPP should not be treated with anti-microbial agents. Although tylosin is said to be effective, it does not guarantee clearance of the organism and the recovered animal may develop a sequestrum in which viable Mycoplasmas may be found. In this study it was found out that sick animals were treated using Oxytetracycline (OTC) 20% by owners or in some cases by extension workers. In one case a paraveterinarian was routinely treating such animals. Some such cases recovered clinically but on ascultation, parts of the lung lobes were found to have no respiratory sounds thus indicating consolidation. It was also found out that treatment by some owners was unconventional in that some injected the OTC directly in the thoracic cavity. This was observed in Wami Mkongeni, Kilosa district. In Mbeya among the Barbaig, sick animals were treated using a concoction of gin ('Power' brand) and OTC. In Chunya a Sukuma cattle owner solicited for help from a witch doctor from Sumbawanga with doubtful results.

Disease control measures: Vaccination

Government instituted a vaccination programme to control CBPP. There was supposed to be a series of four vaccinations on day zero and then after three, six and twelve months. The first vaccination was free of charge. The response was very good and vaccination coverage was high from 80% up to 100%. The subsequent vaccinations were on cost recovery basis and owners were charged from 100/= to 150/= per dose depending on the decision of the local extension workers. The coverage for the second and third vaccinations declined to 25% and 33% respectively in Ulanga and 47% and 56% in Kilombero. Some owners did not want their cattle vaccinated for fear of side effects such swelling and sloughing at the site of injection or loss of a tail switch. One such case was found in Mbarali where the owner refused even free vaccination. When the vaccine was first imported into the country two problems arose. Proper instructions of dosage and route of inoculation was inadequately given and the first batch of the vaccine appears to have had poor efficacy and Government was compelled to import another type.

Clinical and Postmortem Examination of CBPP Cases in the Field

Clinically, CBPP cases were emaciated (except for the peracute cases), had dyspnoea and laboured breathing with stretched necks, dilated nares and elbows turned out. When moved they coughed and were easily exhausted. Usually only one lung was affected. When ascultated, the thoracic side where the lung was affected, revealed varied sounds, from no sounds at all, rales to crackling sounds. Two treated CBPP cases and one CCPP case which were ascultated revealed that the affected sides were similar to the untreated with sounds varying from no sounds to crackling sounds, confirming that the animals did not recover after OTC treatment. Four CBPP cases were offered for postmortem examination in Mbarali, Chunya and Morogoro. They all showed typical consolidation and marbling of the lung lobes, straw coloured pleural fluid and fibrinous coat and adhesions over the affected lung, pleura and pericardium. A case in Chunya which had been treated with OTC for a prolonged period and did not recover, had a tennis ball sized sequestrum.

Abattoir surveillance

The meat inspector at the Ifakara town abattoir was requested to carry out surveillance on CBPP from September 1998 to April 1999. Specimens were submitted to the SUA laboratory thus:

Two sequestra in September 1998 and they originated from Mtimbira, Ulanga, four in October 1998, one each from Ifakara and Mtimbira and two from Malinyi. There were none from December 1998 to February 1999. Two sequestra were submitted in March 1998 and one whole lung with CBPP lesions was submitted in April.

Mycoplasma isolation and identification

Altogether 38 specimens were cultured for Mycoplasmas. Out of the 23 specimens collected from CBPP suspect cases, 14 of them yielded *Mycoplasma sp.* Of the thirteen specimens from CCPP suspect cases, only one yielded *Mycoplasma* sp. All the two specimens from the two CRD suspect cases yielded *Mycoplasma* upon culture. Altogether, 17 mycoplasma strains were isolated and of these, 14 were confirmed to be *Mycoplasma mycoides* subsp. *mycoides* small colony type (MMMsc). One isolate from a goat was *Mycoplasma mycoides* subsp. *mycoides* large colony type (MMMlc) while the two isolates from the CRD suspect cases were shown to be a Mycoplasma but were not typed further.

DISCUSSION

Epidemiological data have shown that unauthorised animal movement was the most important factor in the spread of CBPP in the areas studied. The unimproved pastoralist animal husbandry practices account for the transhumance, lack of permanent settlements, pastures and water and hence nomadism and illegal animal movements. Cattle rustling are an old tradition among patoralists which dates back from the time when there was no central authority in the country. Such lawlessness has continued to date. This has contributed to the spread of the diseases. In the present study however, no such cases were reported. Low or lack of formal education was partly responsible for ignorance by pastoralists about CBPP, its control and the belief in witchcraft. The rudimentary knowledge the pastoralists have concerning use of antibiotics for the treatment of animal diseases was clearly revealed when CBPP cases were treated using OTC instead of tylosin and in some cases using the wrong route of inoculation. All those factors were in one way or another responsible for the spread of the disease and the endemicity. The problem is compounded by the fact that CBPP is a difficult disease to eradicate leave alone control because of its insidious nature, the long incubation period and the carrier state. In addition, the fact that some pastoralists can and do camp far from passable roads makes it difficult to effect disease control measures.

Many times quarantines were flouted by owners because of the reasons stated above. At times, local veterinarians lacked cooperation from local authorities who put their priority on revenue instead of disease control. Veterinarians sometimes yielded and temporarily lifted the quarantines because the owners needed money to pay for the vaccine or to buy food when there was food shortage.

Owners treated CBPP cases with OTC with doubtful success as was discovered in Chunya where a CBPP case was treated without recovery and was left to die. The ease with each

veterinary drugs are sold without prescription make them readily available to non-veterinarians and animal owners and that explains the widespread use of OTC for the treatment of CBPP. There is no doubt therefore that Tanzania will have a large population of CBPP carriers and that is supported by the frequency with which sequestra are encountered at the abattoirs and the lack of complete healing of the affected lungs as shown under Results (Masiga and Domenech (1995).

As shown above, the CBPP control in Tanzania and probably in the other East African countries has been mishandled right from the time the first outbreak occurred in Liliondo in 1990 and in Kagera in 1992. Botswana managed to eradicate CBPP within two years of the outbreak from February, 1995 to January, 1997. They slaughtered over 300,000 cattle (Raborokgwe, 1997, Amanfu *et al*, 1998). In Tanzania, failure to observe quarantines, failure to implement regular vaccination campaigns against CBPP, lack of an established disease surveillance programme, lack of a CBPP control or eradication programme with compensation, uncontrolled livestock movement, the pastoralist tradition of nomadism and free availability of veterinary drugs have all contributed to present endemicity of CBPP in the country. The low funding of the Ministry of Agriculture and the livestock Department in particular has contributed a lot to the CBPP situation in the country. The option Tanzania has at present is concerted and sustained efforts in regular sero- and abattoir surveillance and follow up on place of origin of the positive herds. That individual cattle of the positive herds be tested and those found positive be treated with tylosin and then sold for slaughter. After all, it has been shown that animals which have recovered from CBPP have zero or low productivity (Masiga and Domenech , 1995). Such surveillance will ensure early detection and timely control of the disease. Free vaccination against CBPP is strongly recommended for the animals at risk and those in border districts. The Government ought to make sure that the imported vaccine meets internationally acceptable quality standards and that proper and adequate instruction on the storage, dose and route of inoculation are given by the manufacturer and appropriately trained manpower. That calls for tangibly increased funding for the Livestock Department by Government. That way the disease will be controlled. With increased funding it shall be possible to station livestock extension workers in every village and thus improving coverage of the country with regard to disease surveillance, reporting and control.

The capacity to isolate and identify *Mycoplasma mycoides* subsp. *mycoides* sc and other Mycoplasmas has been established in Tanzania, at SUA and at ADRI. That capacity ought to be sustained. With the appropriate media it was possible to isolate Mycoplasma and type it to species level thus confirming CBPP, this was despite the fact that it took time from collection to isolation. It is therefore advised that the distance and time from the point of outbreak or abattoir should hinder the submission of samples for the confirmation of CBPP so long as the samples are kept frozen in a cold box with ice blocks while being shipped to the laboratory. The fact that Mycoplasma was isolted from only one out 13 CCPP suspect cases could mean that the media and culture conditions were not favourably for the culture of the more fastidious causative agent of CCPP, namely *Mycoplasma capricolum subsp. Capripneumoniae*. Attempts to isolate *Pasteurella* sp from the CCPP suspect cases failed when blood agar media were used.

CONCLUSION

There is need to establish a quick clinical diagnostic method such as spot ELISA for field diagnosis of CBPP and to adopt a diagnostic PCR for the abattoir samples to detect Mycoplasma organism which might be in such small numbers to escape detection by the conventional isolation methods (Brocchi *et al.*, 1993, Bashiruddin *et al.*, 1994, Kawa and Hubschle, 1996, le Goff *et al.*, 1998). There is also need to adopt available molecular epidemiological methods in order to differentiate between different MMMsc pathogenic and vaccinal strains (Poumarat and Solsona, 1995, Cheng et al, 1995).

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