HEALTH RISKS FROM PATHOGENS IN LIVESTOCK MANURES

S. Williams
Pirbright Laboratory, Institute for Animal Health, Ash Road, Pirbright GU24 0NF, U K.
stuart.Williams@bbsrc.ac.uk

INTRODUCTION

The operation of large scale farms housing high numbers of animals in one location creates problems with regard to possible spreading of various diseases, some of them transmissible to humans (zoonotic), either directly through excrement or by animal products. Such diseases include parasitic diseases (e.g. helminthoses), viral diseases (e.g. rotavirus infections) and a variety of bacterial diseases such as haemolytic uraemic syndrome from *E. coli* O157, salmonellosis, leptospirosis and tularemia. Notifiable diseases require immediate and far reaching measures but these would not be expected to fit into the normal operation of the farm. However, there are a range of health hazards relating to animal wastes on a day by day basis owing to the presence (or potential presence) of a wide range of bacteria and other organisms. Although these are not necessarily the cause of contagious illness, a series of measures that can be classified as "good housekeeping" are still justified in reducing the health risk to individuals who come in close contact with livestock wastes. Even more nebulous is the potential hazard to food crops when wastes are applied to arable land. Although there is a lack of data to support this theory, the genuine fears of food producers and the general public may require positive confirmation of a negligible health risk from livestock manure in the future.

Livestock farms operating without bedding produce liquid wastes, which may pose a particular risk. This type of system often fails to create conditions for biothermal processes in the manure (i.e. self-heating) which, in conjunction with other factors, control pathogen numbers in stored farmyard manure. One way to deal with these problems is by using aerobic or anaerobic treatment of liquid manure on livestock farms or in centralised large scale treatment plants (Strauch, 1998). The presence of oxygen in the system is believed to be a key factor because many pathogens are strict anaerobes and numbers are rapidly reduced in an oxidizing environment; similarly, aerobic organisms may be destroyed during anaerobic processes, e.g. a biogas plant. Elevated temperatures will enhance this effect. The mere presence of competing bacteria can also reduce numbers of a range of organisms that may flourish within the host but dwindle in external environments. However, for some infective organisms, the risks are so great that only a comprehensive treatment, e.g. chemical or thermal sanitation will suffice.

THE EPIDEMIOLOGY OF DISEASES ASSOCIATED WITH LIVESTOCK WASTES

The main pathogens of concern

It is not feasible to mention all pathogens found in livestock wastes because of the huge number and variety of organisms (bacterial and viral) propagating in the gut and related tissue as well as glands. This section is limited, therefore, to those which are excreted in infections of the urinary, respiratory and genital tracts.

Salmonella

The main reservoir is the intestinal tract of animals, but only a few serotypes are adapted to a specific host. Distribution is promoted by abundant reservoir hosts, efficient faecal shedding from carrier animals, persistence within the environment, and the effective use of transmission vectors (feed, vehicles, etc.) (Schwartz, 1999). Young animals are more susceptible to salmonellosis than older ones. Poor sanitation, overcrowding, inclement weather, the stress of hospitalization and surgery, parturition, parasitism, transportation, overtraining, and concurrent viral infections are all factors that predispose animals to clinical salmonellosis. Many animals, particularly swine and poultry that are fed rations containing salmonellae suffer inapparent infections during their lifetimes.

Wild birds and rodents such as rats and mice can also be a source of infection for livestock through faeces contamination of feed or buildings. Salmonellosis usually begins as an enteric infection that may subsequently generalize after entry of the organism into the bloodstream. The animal may develop septicemia, meningitis, arthritis, pneumonia, abortion, or a combination of these diseases.

Human salmonellosis finds its source mainly in foodstuffs of animal origin. However, a *Salmonella* infection is rarely fatal, although figures are conflicting. For example, the mortality rate is estimated at 0.4 cases in 100,000 people in The Netherlands (Pelt et al., 2000), however, the SCVPH (2001) estimated that within the EU, one in every 1000 cases is fatal. Individuals with a deficient immune system are most susceptible (Poppe, 1999). Nevertheless, consumers are showing growing concern about food safety, and food borne diseases have top priority with respect to integral quality control of the food chain. Good management practice (GMP) and hazard analysis of critical control points (HACCP) programs implemented over the whole production chain are aimed at achieving *Salmonella* free products (Nielsen et al., 1995).

The main problems with *Salmonella* associated with manure are:

- The intestinal tract is the main reservoir for *Salmonella*;
- *Salmonella* may survive for a long time in suitable organic substrates;
- Where application methods inject the manure in the soil, inactivation of *Salmonella* by sunlight is not possible;
- *Salmonella* can be transmitted from manure to crops through the soil;
- Farm animals may be infected by eating contaminated feed.

***Escherichia coli* O157**

Escherichia coli is part of the normal flora of the intestinal tract of most adult mammals and is generally used as a faecal indicator but some serovars may be pathogenic for humans and/or certain species of animals, for example *E. coli* O157. Toxigenic strains may cause high losses in newborn or young animals associated with diarrhoea and with fulminating septicaemias.

Because the intestinal tract of animals, and cattle in particular, is an important reservoir of the bacterium, cattle faeces and manure should be considered to be potentially contaminated with this organism, and care should be taken during the handling of manure. Apart from the risk of direct contamination of foodstuffs, the application of cow manure to land may also lead to the infection and re infection of livestock.

The faeces of cows have been reported to contain *E. coli* levels of 5×10^5 (Witzel et al, 1996) during periods of shedding; a higher value of 5×10^7 is given by Hrubant et al (1972). Survival in the environment is an important epidemiological factor if microorganisms are distributed by solid or liquid manure and the way of transmission to the susceptible host is the faecal-oral.

Thermal destruction of bacterial pathogens may well depend on factors other than temperature, e.g. moisture content, free ammonia concentration, duration of heat treatment and the presence of other microorganisms which may enhance or inhibit pathogen inactivation. For instance, in an industrial compost, *Salmonella* and *E. coli* were found to survive for 59 days at about 60ºC, although they were destroyed during the cooler, curing process (Droffner and Brinton, 1995).

The Genus Mycobacterium

The diseases produced by mycobacteria are known as tuberculosis, a term derived from the small granulomatous nodules or tubercles that are seen in advanced infections by *M. tuberculosis* or *M. bovis*.

Mycobacterium bovis: *M. bovis* is present in cattle populations throughout the world, but its prevalence in the United States and in Great Britain and other European countries where intensive eradication programs have been in place for many years is very low. The principal route of transmission is aerogenous, although a small proportion of infections are milk borne, congenital, or sexually transmitted.

M. bovis infection in humans is responsible for only 1% of new tuberculosis infections and is not currently a widespread problem due to milk pasteurisation and slaughter of infected cattle. (Yates and Grange, 1988). However, some individuals may be at higher risk, such as abattoir workers (Grange and Yates, 1994; Robinson et al., 1988) and those that have other infections, such as HIV.

Generally, around 10% of infected cattle excrete *M. bovis* in their faeces, but it can be as high as 80% (Reuss, 1955), and for this reason, all infected cattle faeces should be considered as a potential source of the organism. *M. bovis* survival in faeces on pasture is related to sunlight and the thickness of the deposit, with the organisms remaining viable for up to 6 months in winter, but generally less than 2 months in summer (Mitscherlich and Marth, 1984). However, it is unlikely that cattle are directly infected with *M. bovis* from the faeces deposited by grazing cattle because cattle avoid grazing close to faeces initially because of its smell (Marten and Donker, 1966).

Another risk from slurry is that its spreading can create aerosols, which may be carried for several hundred metres, particularly on a windy day (Hahesy et al., 1995). Rain guns which spread dirty water on pasture can also propel aerosolised infectious droplets some distance, and cattle have also become infected with *M. bovis* where grazing pasture was sprayed two days previously with contaminated dirty water (Schellner, 1956).

Mycobacterium paratuberculosis: This disease is known as Johne's disease, paratuberculosis, chronic bacterial enteritis, chronic hypertrophic enteritis, and other local names. In tissues and faeces it commonly appears in clumps, some containing a great many organisms. *M. paratuberculosis* is enzootic throughout the world, but is more common in temperate zones and in wet climates. The organism is transmitted in faeces. Young animals are most susceptible, and therefore infections that later become Johne's disease are contracted during the first year of life.

The Genus Yersinia

There are seven species of Yersinia, of which only *Y. pseudotuberculosis* is important as a cause of disease in animals. *Y. enterocolitica* is frequently present in the alimentary tract of animals and can therefore serve as a source of infection for humans.

Yersinia pseudotuberculosis: Y. pseudotuberculosis occurs worldwide in wild rodents, birds and soil. Animals become infected by ingestion of the organism, and most infections occur during cold weather.

Y. enterocolitica: Y. enterocolitica is ubiquitous and since the mid 1970s has been isolated with increasing frequency from humans and chinchillas with alimentary tract disturbances, from healthy cattle and swine, and from meat, shellfish, and ice-cream. Some human infections are derived from animal sources and from water contaminated with animal faeces. However, most human cases appear to be associated with consumption of food contaminated by human carriers.

The genus Campylobacter

Campylobacter fetus: *C. fetus* sub sp. venerealis and *C. fetus* subsp. fetus characteristically produce comma shaped or S shaped Gram negative bodies (0.2 to 0.5 µm by 1.5 to 4.0 µm). The comma forms have a single polar flagellum while the S forms may have bipolar flagella. The catalase positive species form spirals when a number of S-forms remain joined together. Cells in old cultures form coccoid or spherical bodies. *C. fetus* subsp. fetus has a glycoproteinaceous microcapsule Growth is microaerophilic to anaerobic. The organisms require an atmosphere of 10 to 20 percent CO_2, and an oxygen concentration of 5 percent or less.

Campylobacter fetus subspecies fetus: There are two serovars of *C. fetus* subspecies fetus, A 2 and B, based on heat resistant O antigens (Berg et al. 1971). *C. fetus* subsp. fetus is an occasional cause of abortion during the latter half of gestation in cattle. The pathogenesis of the infection is similar to that caused by *C. fetus* subsp. venerealis, but the organism probably enters the animal by ingestion of infectious material. Only heavily gravid animals are susceptible to bacterial invasion of the placenta with subsequent abortion. The incubation period is from 7 to 25 days.

Campylobacter jejuni: C. jejuni is a thermophilic campylobacter that has emerged in the 1970s as an important zoonotic pathogen. Milk, minced meat, dogs, cats and poultry have been shown to be important sources of infection for humans. *C. jejuni* forms tightly coiled, Gram negative cells 0.2 to 0.3 µm in diameter and 1.5 to 5.0 µm long. *C. jejuni* is widely distributed in the intestine of domestic and wild animals but is rarely isolated from the faeces of normal humans. The organism is pathogenic for humans, and as few as 500 organisms in milk have caused enteritis. It has been implicated in the etiology of winter dysentery or black scours of cattle, diarrhea in calves, dogs, cats, and foals, hepatitis in chickens, and abortion in sheep and mink.

Campylobacter coli: C. coli greatly resembles *C. jejuni* except that it is unable to hydrolyze hippurate. It is an inhabitant of the intestinal tract of swine, poultry, and humans. Although it has been implicated as a cause of enteritis in weaned piglets with clinical signs of mild swine dysentery, it does not appear to be an important pathogen of domestic animals

The Genus Pasteurella

The genus *Pasteurella* belongs to the family *Pasteurella*ceae, a complex group of highly adapted parasitic organisms that includes the genera *Actinobacillus* and *Haemophilus*. *P. multocida* is the type species and embraces a variety of biotypes and serotypes. The other species currently recognized are *P. aerogenes, P. gallinarum, P. haemolytica, P. pneumotropica*, and *P. ureae*. *P. multocida* and *P. haemolytica* are important pathogens of domestic and wild animals. *P. multocida* causes primary septicaemias in cattle and in domestic and wild birds and is also an important opportunistic invader of the respiratory tract of a variety of species.

Pasteurella multocida: P. multocida is normally maintained as a commensal of the oropharynx of mammals. Unlike mammals, though, healthy birds do not carry the organism, and its presence in birds is almost invariably associated with acute or chronic diseases. Fowl cholera, the pasteurellosis of birds, affects chickens principally, although ducks, geese, turkeys, swans, and other birds are susceptible. Wild birds frequently become infected and at times may be source of infection for domestic flocks. Many outbreaks of respiratory diseases in pigs and cattle caused by *P. multocida*, however, are apparently the result of invasions by endogenous infections of the nasopharynx. A variety of stressful situations such as shipment, viral infection, bad weather, poor nutrition, and overcrowding can impair the physical and immunological defences of animals and allow *P. multocida* to multiply on the nasopharyngeal mucosa, with subsequent penetration of the lower respiratory tract. Shipping fever characteristically begins a few days to a few weeks after transport. Affected animals cough, are febrile, and have a nasal discharge.

The Genus Brucella

Infections typically are found in the reproductive organs and in the reticulo-endothelial tissues. In cattle, sheep, and goats, lesions of the female reproductive tract and of the placenta and fetus can lead to abortion with consequent severe economic loss.

Brucella abortus: *B. abortus* is found in cattle populations throughout the world, although it is rare or absent in some countries where eradication programs have been actively pursued. *B. abortus* is a short rod or a coccobacillus measuring 0.5 to 0.7 µm. *B. abortus* is non-motile and does not have a well developed capsule. Poorly developed capsules have been demonstrated on freshly isolated strains using special stains. Growth is aerobic, but many strains require increased tensions of CO_2 for growth, especially on primary isolation. *B. abortus* is an obligate facultative intracellular parasite of cattle and some wild ruminants and is freely transmitted by ingestion of contaminated discharges or feed.

Brucella suis: B. suis occurs in most swine raising areas of the world. Survival in the environment has not been well studied but probably is similar to that of *B. abortus*. The organism survives in faeces and urine for at least 6 weeks.

Brucella melitensis: The usual natural hosts for *B. melitensis* are goats and sheep, but pigs, cattle, camels and humans are also easily infected. Infection has been found in many species of feral animals, including hares and impala.

Listeria monocytogenes

Cattle and sheep are commonly infected with serotypes 1 or 4. Meningoencephalitis is perhaps the most easily recognized form of listeriosis seen in domestic animals, and is called circling disease in sheep in New Zealand. The disease has since been observed in cattle and sheep in many parts of the world.

Erysopelothrix rhusiopatiae

E. rhusiopathiae is the cause of swine erysipelas, which has long been recognized as a destructive disease of young pigs in many parts of the world. A variety of disease forms are observed, including skin lesions (diamond skin disease), septicemia, polyarthritis, endocarditis, and abortion. In addition to swine, *E. rhusiopathiae* has been found in lambs suffering from polyarthritis, in calves suffering from a similar disease, in turkeys and ducks suffering from acute septicemic infections, in wild and laboratory mice in the tonsils and on the mucous membranes of apparently normal swine, in various decaying plant and animal tissues, on the skin of freshwater and saltwater fish, and in skin lesions in humans known as erysipeloid.

The source of the organisms that infect the first cases in outbreaks of erysipelas is uncertain. Many healthy swine carry the organism in their tonsils and reticulo-endothelial tissues, and it is probable that stresses such as excessive heat and humidity or other predisposing factors cause impairment of the antibacterial defence mechanism with subsequent multiplication of the organism in the animal's body. Many cases of wound infection of human hands have been reported. In Europe most of the human cases have been attributed of the handling of infected swine and pork, but some have occurred in fishermen and fish dealers who have had no contact with swine.

The Genus Streptococcus

Streptococci cause a variety of diseases in humans and animals and are important saprophytes in milk and milk products. They are frequently present as parasites of the mucous membranes and intestines of animals and, given the appropriate conditions, can opportunistically produce disease. Streptococcal species are important causes of mastitis in cattle, of strangles and other diseases in horses, and of meningoencephalitis, arthritis, endocarditis and cervical lymphadenitis in swine. Less frequently they have been associated with septicemia in chickens and with respiratory infections and other infections in kittens and puppies.

Streptococcus agalactiae: S. agalactiae is a common cause of bovine mastitis in most areas of the world. It also causes mastitis in sheep and goats. The organism is less frequently a cause of bovine mastitis in Great Britain than in other countries, including the United States. The reasons of this disparity are not fully understood. In Denmark an eradication program has reduced the infection rate to less than 1 percent in cows.

Enterococci: Streptococcus faecalis, S. faecium, S. avium and *S. gallinarum* comprise the group of enterococci. *S. faecalis* and *S. faecium* are residents of the intestinal tracts of humans and most animals, and the latter two species occur in poultry. *S. bovis* and *S. equines*, residents of the bovine and equine intestinal tracts and possessing the group C antigen, differ physiologically and in the properties of their fructose diphosphate aldolases. They also have a high minimum temperature for growth, 20 22ºC, and they do not survive outside the animal host.

Aerobic Sporeformers

Aerobic sporeformers are widely distributed in nature and are mainly involved in the degradation of organic material in soil. Most of them are apathogenic for warm blooded animals but some form endotoxins, e.g. *Bacillus cereus*, *Bacillus anthracis* and *Bacillus thuringiensis*.

Bacillus anthracis: B. anthracis causes the disease known as anthrax. Herbivorous animals are highly susceptible to infection by *B. anthracis*; carnivorous birds and reptiles are resistant. The disease is often fatal in humans, although they are not as susceptible as herbivores. They are also resistant to disinfectants such as 5 percent phenol or mercuric chloride. Spores have been shown to remain viable for more than 100 years. The major anthrax enzootic zones of the world are found in the tropics and subtropics India, Pakistan, Africa, and South America, for example. Natural infection of animals can occur via the skin or respiratory tract but usually occurs by ingestion of spores, which geminate and produce vegetative bacilli, either in the mucosa of the throat or in the intestinal tract.

The Genus Clostridium

The clostridia can be divided into two groups on the basis of their disease producing mechanisms. The first group consists of those species that have little or no ability to invade and multiply in living tissues. Such organisms owe their pathogenicity to the production of powerful toxins that are produced outside the body of the host or in localized areas within the body. Two organisms of this group are *C. tetani* and *C. botulinum*. The second and larger group consists of species that can invade and multiply in the tissues or the intestine of the host animal. These organisms in most cases also produce toxins, but the toxins are less potent than those of the first group.

Clostridium perfringens: The spores are oval and fairly small. *C. perfringens* is ubiquitous in nature and is part of the normal intestinal flora of healthy animals. Highly proteinaceous diets lead to great increases in populations of the organism in the gut. Type-A strains are well adapted to survival in the soil, whereas type B, C, D, and E strains appear to be more highly adapted to the intestine.

Clostridium chauvoei: C. chauvoei causes "Blackleg" in ruminants. It occurs throughout the world and is a source of considerable economic loss. The disease rarely occurs in species other than ruminants. Once pastures or grazing grounds become infected, the disease reappears in susceptible animals year after year. Spring and autumn are the seasons of greatest occurrence of blackleg in the United States. In Europe the disease frequently occurs in the summer. The infection is most common on permanent pastures and certain pastures in a locality appear to be at especially high risk. Land cultivation diminishes this risk.

Cryptosporidium

The organism: Cryptosporidium parvum is a protozoan parasite, which infects humans and animals through oocysts which are small (0.4 to 0.6mm) oval particles. These are protected by a tough membrane wall inside of which four sporozoites are stored. After ingestion of the oocysts by an animal or human, the sporozoites exit from the oocyst, attach themselves to the gut wall and rapidly multiply. This causes cryptosporidiosis, a severe diarrhoea which is self limiting, lasting for approximately 2 3 weeks in humans and 10 to 15 days in neonatal lambs and calves before spontaneous recovery. Not all Cryptosporidium parvum isolates excreted by animals are pathogenic to humans, while those adapted to humans always are (Awad - el - Kariem et al., 1998).

Transport and Survival of Oocysts: Faeces of young infected calves containing on average 3 x 10^6 oocysts per gram of faeces (MAFF, 1995) will be mainly retained in the bedding. The leachate from bedding can contain large numbers of oocysts - in the region of 8 x 10^7 per calf. In the midden, the number and viability of oocysts can rapidly decrease in the collected bedding containing faeces, mainly due to the high temperature and ammonia concentration generated by composting. Oocysts in cattle slurry or manure, however, survived storage for more than 3 months, especially at lower temperatures, i.e. 4ºC, while at 15 and 20ºC they were killed in 30 and 20 days respectively (Svoboda et al., 1997).

RISK OF INFECTION FROM LIVESTOCK MANURES

Transfer mechanisms

Recent work has highlighted the potential risks to humans from exposure to livestock pathogens via the food chain or through direct or indirect contact with the environment, animals and animal manure. Transfer within the environment (more specifically via slurry, solid manure, poultry litter, dirty water, sewage sludge and abattoir wastes) to where they are at risk of infecting humans can occur in a number of ways. Likely vectors for pathogen distribution are: physical dispersion of waste (e.g. using slurry and solid manure spreaders); transport of material; run off from housing, pastures and handling areas and inadequate storage; livestock themselves (during grazing or movement); wild animals and birds; and physical movement of people associated with livestock. These may lead, in varying degrees, to contamination of crops, soil, water and air. People may pick up infections from these sources through recreational activities such as walking through countryside, camping, festivals and water sports, as well as farming activities. Meteorological conditions will have large effect on aerosol dispersion and deposition together with the growth and death rates of pathogens in the environment.

Risks to public and livestock

Bacteria, viruses and parasites from faecal sources as slurry or solid manure are of environmental importance since the relevant materials are stored and handled on farms and applied to agricultural soils. This means that pathogenic and non pathogenic bacteria, fungi, viruses and parasites are released, because treatment of the faeces for the inactivation of pathogens is only done in rare cases and storage alone is generally not enough for this purpose (Munch et al. 1987). Bacteria, viruses, fungi and parasites from the gut of farm animals may be also spread via the air from the animal stalls to the environment, when using liquid or solid manure as a fertilizer, or from the slaughterhouse via waste water into surface water. Depending on the circumstances and type of microorganisms, this may result in an introduction of pathogenic, or genetically engineered microorganisms or antibiotic resistant bacteria, into the environment, e.g. the soil, the surface and occasionally ground water. Little attention has been paid in the past to bacterial and fungal metabolites and toxins in faecal media, which may be produced by propagation or lysis of bacterial or fungal cells during storage, aerobic or anaerobic treatment and will be released by the spreading of solid or liquid manure. Products such as endotoxins or exotoxins may also be propagated through aerosols, and this risk has to be taken into account if these materials are taken up by humans or animals and via the pollution of water, air or soil.

There are two different epidemiological risks to be regarded here, which may differ under quantitative and qualitative aspects depending on whether the target is human or animal. The relevant epidemiological pathways are summarized in Figures. 1.1 (non-zoonotic) and 1.2 (zoonotic). With respect to manure, two different levels of risk do exist for the animal population. Manure of clinically healthy herds must be regarded generally as of low risk because pathogens can be found only in a certain percentage for a limited period of time. In such cases the germ count is relatively low, i.e. around the detection level.

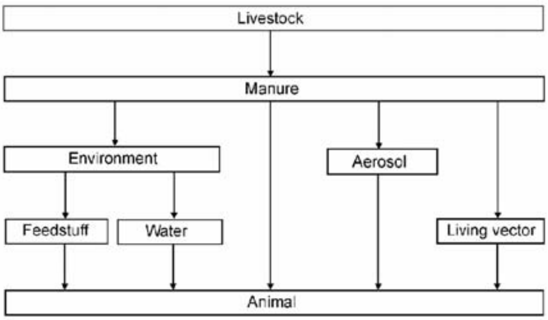

Figure 1.1 *Epidemiological pathways for non-zoonotic infectious agents (pathogens affecting animals only).*

The situation is different if the herd or parts of it are showing signs of clinical illness. A wide variety of pathogens is excreted in faeces and/or may be found in manure. *Salmonella* are among the most important bacterial pathogens in manure. From the group of viruses, the enteroviruses, rotaviruses and the influenza viruses seem to be of special importance. In the case of infected herds the number of pathogenic microorganisms that may be found in slurry is considerably higher than mentioned above, e.g. 10^4-10^5 CFU per ml for salmonella. The number of microorganisms that are found depends on the number of infected animals per herd, the volume of slurry in the storage tank before the infection, the housing system and the water consumption.

If zoonotic agents are excreted and/or zoonotic agents are the cause of illness in the herd, the human population is at risk too. The risk depends on number of excreted organisms, on tenacity of the agent in manure and the environment, the necessary infectious dose for humans, its host spectrum and thus ability to generate carriers in the environment and the ability to contaminate food or drinking water under the given conditions.

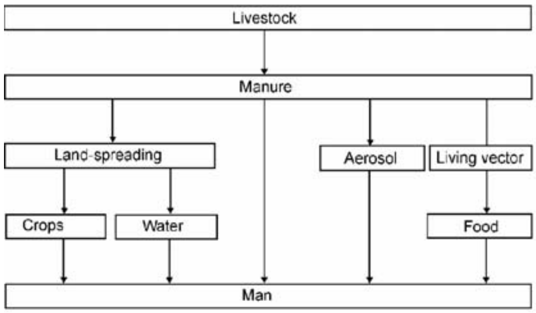

Figure 1.2 *Epidemiological pathways for zoonotic agents (animal pathogens affecting man).*

REFERENCES

Awad-El-Kariem, F.M., Robinson, H.A., Petry, F., McDonald, V., Evans, D., Casemore, D. 1998. Differentiation between human and animal isolates of cryptosporidium parvum using molecular and biological markers. Parasitol. Res., 84: 297-301.

Berg, R.I., Jutila, J.W., Firehammer, B.D. 1971 A revised classification of Vibrio fetus. Am. J. Res., 32: 11-22.

Droffner, M.L., Brinton, W.F. 1995. Survival of *E.coli* and *Salmonella* populations in aerobic thermophilic composts as measured with DNA gene probes. Zbl. Hyg., 197: 387-397.

Grange, J.M.; Yates, M.D. 1994. Zoonotic aspects of *Mycobacterium bovis* infection. Vet. Microbiol., 40: 137-151.

Hahesy, T., Scanlon, M., Carton, O.T., Quinn, P.J., Cuddihy, A. 1995. Aerosol Dispersal of Cattle Slurry on Holdings Restricted due to Bovine Tuberculosis. Tuberculosis Investigation Unit, University College, Dublin.

Hrubant, G.R., Daugherty, R.V., Rhodes, R.A. 1972. Enterobacteria in feedlot waste and runoff. Applied. Microbiol., 24: 3, 378-383.

MAFF (Kemp, J.S., Wright, J.E., Coop, R.L., Mawdsley, J.L.I., Bargett, R.D., Merry, R.J., Pain, B.F., Theodorou, M.K., Svoboda, I.F., Read, I.A., Bukhari, Z., Smith, H.V.) 1995. Protozoan, Bacterial and Viral Pathogens, Farm Animal Wastes and Water Quality. Final Report to MAFF (CSA 2064), p.155.

Marten, G.C., Donker, J.D. 1966. Animal excrement as a factor influencing acceptability of grazed forage. Tenth International Grassland Congress, Helsinki. pp. 359-363.

Mitserlich, E., Marth, E.H. 1984. Microbial Survival in the Environment. Springer-Verlag, Berlin.

Munch, B., Errebo- Larsen, H., Aalbaek, B. 1987. Experimental studies on the survival of pathogenic and indicator bacteria in aerated and nonaerated cattle and pig slurry. Biol. Wastes, 22: 49-65.

Poppe, C. 1999. In Saeed, A.M., Gast, R.K., Potter, M.E., Wall, P.G. (eds) *Salmonella* Enterica Serovar Enteritidis in Humans and Animals. Iowa State university Press, Ames, USA.

Reuss, U. 1955. Tuberkelbakterien im Kot tuberculinpositiver Rinder und ihre weidehygienische bedeutung (Occurence of tubercule bacilli in the faeces of tuberculin- positive cattle and its significance in pasture hygiene) Die Rinder- Tuberkulose, 4: 232-234.

Robinson, P., Morris, D., Antic, R. 1988 Mycobacterium bovis as an occupational hazard in abattoir workers. Aust. NZ. J. Med., 18: 701-703.

Schellner, H. 1956. Risk of infection in cattle grazing pastures contaminated with tubercle bacilli. Rindertuberkulose, 5: 179-188.

Schwartz, K.J. 1999. Salmonellosis. In Diseases of Swine, eds B.E.Shaw; D`Allaire; W.I.Mengeling; D.J.Taylor. Blackwell Science, pp 535-551.

Strauch, D. 1998. Pathogenic microorganisms in sludge. Anaerobic digestion and disinfection methods to make sludge useable as a fertiliser. Eur. Water Manag., 1: 2, 12-26.

Svobodova, I.F., Read, I.A., Kemp, J.S., Wright, S.E., Coop, R.L., Mawdsley, J.L.I., Bargett, R.D., Merry, R.J., Pain, B.F., Theodorou, M.K., Bukkhari, Z., Smith, H.V. 1997. Cryptosporidium on cattle farms. In "Cryptosporidium in Water- the Challenge to policy Makers and Water managers", Symposium by Chartered Institute of Water and Environmental Management, Glasgow, pp 3-20.

Yates, M.D., Grange, J.M. 1988. Incidence and nature of human tuberculosis due to bovine tubercle bacilli in South East England: 1977-1987. Epidemiol. Infect., 101: 225-229.