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Review article

Chemical contaminants in feedlot wastes: Concentrations, effects and attenuation

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Abstract

Commercial feedlots for beef cattle finishing are potential sources of a range of trace chemicals which have human health or environmental significance. To ensure adequate protection of human and environmental health from exposure to these chemicals, the application of effective manure and effluent management practices is warranted. The Australian meat and livestock industry has adopted a proactive approach to the identification of best management practices. Accordingly, this review was undertaken to identify key chemical species that may require consideration in the development of guidelines for feedlot manure and effluent management practices in Australia. Important classes of trace chemicals identified include steroidal hormones, antibiotics, ectoparasiticides, mycotoxins, heavy metals and dioxins. These are described in terms of their likely sources, expected concentrations and public health or environmental significance based on international data and research. Androgenic hormones such as testosterone and trenbolone are significantly active in feedlot wastes, but they are poorly understood in terms of fate and environmental implications. The careful management of residues of antibiotics including virginiamycin, tylosin and oxytetracycline appears prudent in terms of minimising the risk of potential public health impacts from resistant strains of bacteria. Good management of ectoparasiticides including synthetic pyrethroids, macrocyclic lactones, fluazuron, and amitraz is important for the prevention of potential ecological implications, particularly towards dung beetles. Very few of these individual chemical contaminants have been thoroughly investigated in terms of concentrations, effects and attenuation in Australian feedlot wastes.

Keywords: Feedlots; Hormones; Antibiotics; Ectoparasiticides; Waste management

Contents

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1. Introduction

The Australian meat and livestock industry has adopted a proactive stance towards the implementation of best practices for the management of chemical contaminants that may be present in manure and effluent from commercial feedlot operations. While numerous activities will be necessary to achieve this, the first requirement is the identification of chemicals likely to warrant closest scrutiny. This review of primarily international data and research was undertaken to identify key chemicals for which local analytical efforts appear most justified.

Commercial feedlots are a major method of finishing beef cattle in preparation for slaughter in Australia. Cattle entering feedlots are typically 12–24 months of age. Depending on the intended market for the cattle, they may be fed for 60 days to up to 400 days while gaining about 100–350 kg in weight.

The main by-products from cattle feedlots are the manure harvested from the surface of the pens and liquid effluent collected during rainfall runoff events. A typical animal entering a feedlot (e.g. 340 kg for heavier markets) produces approximately 20 kg of manure per day, increasing to up to 36 kg manure per day for a heavy finished animal (600 kg). Fresh manure, which comprises of faeces and urine, is normally composed of around 90% water and 10% solids.

Good feedlot pad management requires a balance between environmental and animal health considerations and the economic cost of pen cleaning. However, the period of time between pen cleanings generally means that there is some decomposition of pad manure before it is removed from the pen.

Depending on variations in management and weather, manure harvesting rates have been reported to vary between 0.41–1.05 t dry weight per head per year (Lott et al., 1994).

While manure harvesting from pens occurs at regular intervals, manure spreading or dispatch offsite depends upon management methods, weather conditions and cropping cycles which influence when manure can be spread or sold. Accordingly, manure is often stockpiled at feedlots for periods of months or occasionally years. During stockpiling, manure undergoes partial decomposition and drying which can lead to significant mass decrease. If stockpiles are carefully managed ensuring aeration as well as optimal water content and carbon to nitrogen ratios, manure may be composted in the process. Much of the organic matter can be mineralised or volatilised during

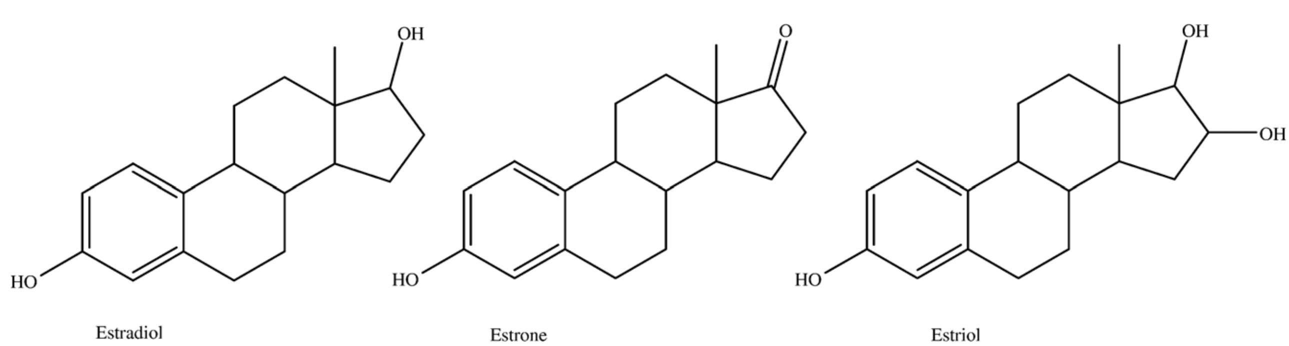

Fig. 1. Molecular structures of endogenous estrogenic steroidal hormones.

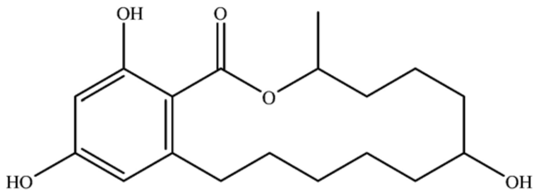

Fig. 2. Molecular structure of zeranol (α-zearalanol).

composting leading to mass reductions of a further 50% of the original bulk.

From the point of manure deposition on the pad, through stockpiling, to eventual removal from the feedlot site, opportunities exist for the transport or transformation of chemical components or contaminants. The major transport routes may include runoff from impermeable surfaces and leaching through soil to groundwater. Additional routes may involve human intervention such as manure stockpiling or application to agricultural fields. Likely chemical transformations include oxidative or reductive degradations, which may be photochemically or biochemically mediated. The degree of such transformations and/or transport is important when assessing the probability of human exposure. This in turn is necessary for the task of identifying best practices for the management of feedlot manure and effluent to ensure the full protection of human health.

Chemicals of concern in cattle feedlot manure and effluent may include endogenous chemicals such as hormones, as well as non-endogenous natural and synthetic chemicals used to maintain the health and optimum growing conditions for the animals. Furthermore, there is potential for animals (and hence manure and effluent) to be unintentionally exposed to chemicals in the environment or via contaminated feed products. To reduce the likelihood of this occurring, all feedstuff purchases in Australia require a Commodity Vendor Declaration stating the fodder type and any chemicals such as pesticides applied during production.

Veterinary chemicals may be administered to animals while at the feedlot or, alternatively residues of such chemicals applied at a previous location may be (wittingly or otherwise) transported to the feedlot with incoming animals. Again, management practices to minimise this in Australia include the requirement that all feedlot animals are subject to a National Vendor Declaration, which serves as a travel document and describes all chemicals used in the production of that animal.

After consideration of the international scientific literature on this subject, steroidal hormones, antibiotics, ectoparasiticides, mycotoxins, heavy metals and dioxins were identified as being most worthwhile for further analysis in the Australian context. Each of these categories of chemicals is addressed in the following sections.

2. Steroidal hormones

Steroidal hormones potentially present in feedlot manure and effluent include endogenous (naturally occurring) hormones and some synthetic hormones applied in agriculture. Endogenous hormones are commonly identified in animal excretions including manure and urine (Hoffmann et al., 1997). The levels of these residues vary considerably with sex, age, breed, castration, and pregnancy.

Both natural and synthetic steroidal hormones are used in many countries as hormonal growth promotants (HGPs) in cattle (Mader and Lechtenberg, 2000; Preston, 1999; Song and Choi, 2001). They are used to improve feed efficiency, rates of weight gain and relative proportions of muscle and fat (Lefebvre et al., 2006). Such HGPs have been widely used in some sectors of the beef cattle industry in mainland Australia since 1979 (Sawyer and Barker, 1988) and are registered by the Australian Pesticides and Veterinary Medicines Authority (APVMA, 2006). HGPs are also used throughout the USA and Canada. However their use is currently prohibited within member countries of the European Commission (Galbraith, 2002).

The use of HGPs may increase both the range and concentration of steroids present in livestock wastes (Preston, 1999). HGPs are normally administered via a subcutaneous implant in the animal's ear. All such compounds are known to be present in animal tissue as well as in urinary and faecal excretions. At slaughter, the ear, along with any residual drug in the implant is discarded (Galbraith, 2002).

Commercially available hormonal products are also used to improve the reproductive performance of dairy cattle (Refsdal, 2000). Natural and synthetic hormonal steroids, including progestins and estrogens, are used in Australia for a range of purposes such as the synchronisation of heifer ovulation cycles and improvement of fertility (Dairy Research and Development Corporation, 1997). However, unlike many other countries, it is not common practice to lot feed dairy cows in Australia and thus products applied for these uses are unlikely to be significant contaminants in Australian feedlots.

Subcutaneous implant devices containing active steroidal hormones are registered under a number of trade names in Australia including Elanco Compudose, Revalor, Coopers Ralgro, Crestar, Synovex and Progro (APVMA, 2006). The active hormones in these products include 17β-estradiol, estradiol valerate, estradiol benzoate, trenbolone acetate, zeranol, progesterone, and testosterone propionate.

2.1. Estrogens

17β-Estradiol is the most active of the natural estrogens. During metabolism, 17β-estradiol is primarily converted to estrone and further to estriol (Fig. 1). A variety of sulphate and glucuronide conjugates are also excreted (Hoffmann et al., 1997; Williams and Stancel, 1996). The relative potencies of the steroidal estrogens appear to be somewhat dependant on the assay used and the endpoint targeted. However it seems clear that 17α-estradiol, estriol and estrone are generally less potent than 17β-estradiol (Soto et al., 1995; Sumpter et al., 2006). Total estrogen production can vary significantly for dairy cows depending on pregnancy stage (Hoffmann et al., 1997). While estradiol is an endogenous hormone, animals may be treated with further supplements to promote growth. Commercial implants in Australia contain 12–400 mg estradiol (APVMA, 2006).

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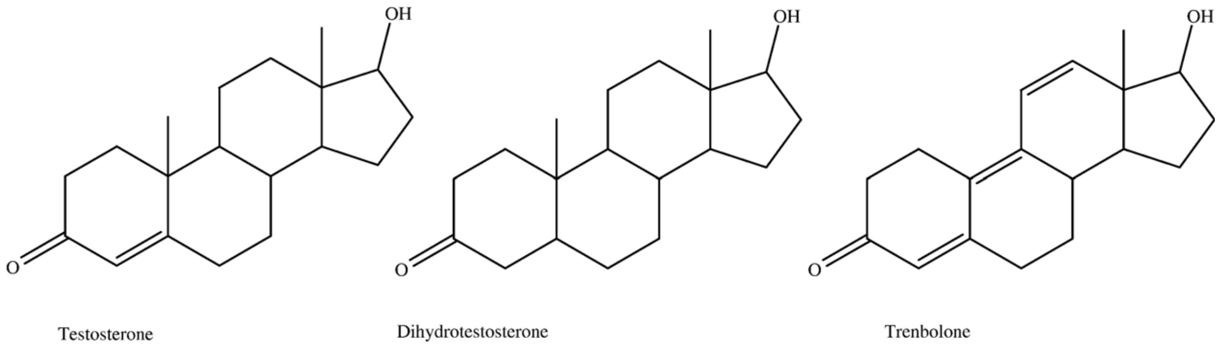

Fig. 3. Molecular structures of androgenic steroids.

Zeranol (α-zearalanol) (Fig. 2) is a synthetic beta resorcylic acid lactone administered as an anabolic growth promoter in beef cattle (Song and Choi, 2001). While zeranol is not truly a steroidal compound, it is used as a hormonal growth promotant in parenteral implant devices due to its estrogenic nature. It is structurally and metabolically related to the myco-estrogen zearalanone (see Section 5). Zeranol has been demonstrated to be estrogenic, with potencies similar to 17β-oestradiol in a range of bioassays (Le Guevel and Pakdel, 2001; Leffers et al., 2001). Once administered, zeranol is predominantly metabolised to its diastereoisomer β-zearalanol (taleranol) and to a lesser extent, further to zearalanone (Kleinova et al., 2002). Commercial implants in Australia contain 36 mg zeranol (APVMA, 2006).

2.2. Androgens

Testosterone secreted by the testes is the main androgen in males, along with its similarly active metabolite dihydrotestosterone (see Fig. 3). These natural androgens are metabolised and excreted in urine as both free steroids and water-soluble conjugates (Wilson, 1996). The major urinary metabolites are etiocholanolone and androsterone, both of which are physiologically weak or inactive (Wilson, 1996). In addition to endogenous testosterone, further testosterone or testosterone propionate may be used as a HGP in bovines. Commercial implants in Australia contain 200 mg testosterone propionate (APVMA, 2006).

Trenbolone (Fig. 3) is a synthetic androgenic steroid. It is used to promote growth and enhance the efficiency of feed utilisation in beef cattle. Trenbolone is administered to feedlot cattle as trenbolone acetate, which is hydrolysed to form the potent androgen receptor agonist 17β-trenbolone. The major metabolic route of 17β-trenbolone is oxidation to trendione, followed by reduction to the less potent epimer, 17α-trenbolone (Schiffer et al., 2001). 17β-trenbolone has an anabolic activity several times above that of testosterone. Commercial implants in Australia contain 60–200 mg trenbolone acetate (APVMA, 2006).

2.3. Progestins

Secretion of progesterone (Fig. 4) is most prevalent during the second half of the female estrous cycle. Progesterone is metabolised to hydroxylated metabolites and their sulphate and glucuronide conjugates which are eliminated in the urine (Williams and Stancel, 1996). Progesterone is further used, in combination with estrogenic hormones, as a HGP, as well as to suppress estrus in feedlot heifers. Commercial implants in Australia contain 100–200 mg progesterone (APVMA, 2006).

Melengestrol acetate (Fig. 4) is an orally applied synthetic steroidal progestin widely used as a HGP as well as for estrus synchronisation and estrus suppression in feedlot heifers in the USA. However, it is not currently approved for use in Australia.

2.4. Environmental and public health concerns

During the last few decades, reports of hormonally related abnormalities in a wide range of species have accumulated

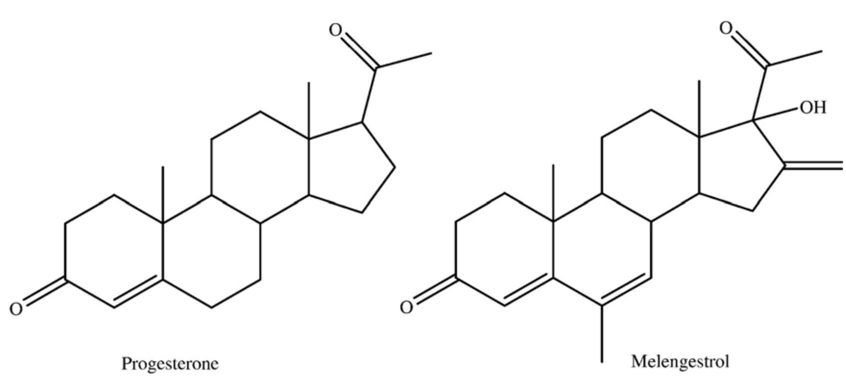

Fig. 4. Molecular structures of progestinal steroids.

(Matthiessen, 2003). Chemical contaminants are believed to be responsible for many of these abnormalities, acting via mechanisms leading to alteration in endocrine function. This phenomenon, known generally as 'endocrine disruption', has been identified by the World Health Organization as an issue of global concern (Damstra et al., 2002). The chemicals implicated have been collectively termed 'endocrine disrupting chemicals' (EDCs), or simply 'endocrine disruptors'.

A particular form of this endocrine disruption has been the inducement of biochemical hormonal responses in freshwater fish (Jobling and Tyler, 2003). A growing number of natural and synthetic environmental chemicals have been implicated as causative agents of these observed disruptions. However, in terms of potency, the most significant have been natural and synthetic steroidal hormones (Körner et al., 2001; Metcalfe et al., 2001). Some steroidal hormones have been observed to cause disruption of the endocrine system of fish at ambient concentrations less than 1 ng L^{-1} (Purdom et al., 1994).

Environmental exposure to estrogenic hormones has been shown to cause feminisation of male fish (Jobling and Tyler, 2003; Rempel et al., 2006). More recently, exposure to androgens has been implicated in the masculinisation of fish (Ankley et al., 2003; Gray et al., 2006; Jensen et al., 2006; Sone et al., 2005). Furthermore, scientists suspect that anthropogenic estrogens, androgens and progestins may act as reproductive pheromones in fish, thus adversely affecting reproduction (Kolodziej et al., 2003, 2004).

Much attention has focused on the discharge of hormonal steroids from municipal sewage treatment plants (Braga et al., 2005; de Mes et al., 2005). However, livestock wastes have been identified as a further potentially important but poorly understood source of these compounds in the environment (Hanselman et al., 2003; Johnson et al., 2006; Kolodziej et al., 2004; Shore and Shemesh, 2003; Ying and Kookana, 2002).

Municipal sewage effluents have been generally characterised as being 'estrogenic' in nature, due largely to trace concentrations of estrogenic steroidal hormones as well as some other natural and synthetic chemicals. Similarly, dairy wastes appear to be strongly estrogenic due to the presence of a number of the same estrogenic steroidal hormones (Johnson et al., 2006; Raman et al., 2004; Sarmah et al., 2006).

Conversely, discharges from a beef cattle feedlot in the USA have been shown to be significantly androgenic in nature (Durhan et al., 2006). Consistent with this, studies also undertaken in the USA have demonstrated adverse effects to the endocrine and reproductive systems of exposed wild fish populations (Gray et al., 2006; Orlando et al., 2004).

The androgenic nature of feedlot discharges may be in part due to numerous steroidal hormones such testosterone. However, in recent years, particular attention has been focused on trenbolone. Beef cattle feedlot effluents have been shown to contain detectable concentrations of both 17α-trenbolone to 17β-trenobalone (Durhan et al., 2006). 17β-trenbolone is a potent androgenic agent to fish (Ankley et al., 2003; Sone et al., 2005), as well as to some exposed mammals (Wilson et al., 2002). More surprisingly, 17α-trenbolone also appears to have a high potency to some fish, which may arise from substantial conversion of 17α-trenbolone to 17β-trenbolone by the fish (Jensen et al., 2006). This observation demonstrates the importance of considering both epimers when assessing potential ecological risk of androgens associated with beef feedlot effluents.

2.5. Steroid hormones in animal excretions

Steroidal hormones are excreted by mammals in free form, or as sulphate or glucuronide conjugates which may be readily hydrolysed to the corresponding free steroid under appropriate conditions (Layton et al., 2000). Cattle excretions contain considerable quantities of steroidal hormones, depending largely on age, sex and stage of the female oestrus cycle (Hanselman et al., 2003; Shore and Shemesh, 2003).

The banning of hormonal growth promotants for use in cattle production in the European Union has led to the rapid development of sensitive analytical methods to determine illegal use targeting bovine excretions (Aman et al., 2006; Buiarelli et al., 2003; Dickson et al., 2003; Launay et al., 2004; van Bennekom et al., 2002). In some cases, this has facilitated improved understanding of expected excretion patterns of some compounds under specific conditions.

17α-estradiol, 17β-estradiol and estrone account for more than 90% of the estrogens excreted by cattle as free and conjugated steroids (Hanselman et al., 2003). However, endogenous loads of estrogens excreted in dairy cow faeces and urine is highly variable depending on pregnancy stage (Hanselman et al., 2003; Hoffmann et al., 1997; Shore and Shemesh, 2003). Furthermore, the urine of heifers implanted with 2×25 mg zeranol pellets has been reported to contain 2–5 μg L^{-1} zeranol and equivalent concentrations of its diastereoisomer taleranol (Kleinova et al., 2002).

It has been estimated that calves excrete approximately 16 mg of natural estrogenic hormones per year and that male calves excrete around 120 mg of natural androgens per year (Lange et al., 2002). Similarly, bulls were estimated to excrete 200 mg of estrogens and 390 mg of androgens per year.

Both trenbolone and melengestrol acetate have been identified in effluent and solid manure from treated cattle in Germany (Schiffer et al., 2001). Initial concentrations of trenbolone were reported at 1700 ng/kg in effluent and 5–75 μg/kg in solid manure. After 4.5–5.5 months storage, residues of 1100 ng/kg in effluent and up to 10 μg/kg in solid manure were measured. Melengestrol acetate was reported in the solid manure at 0.3–8 μg/kg and remaining at up to 6 μg/kg after 4.5 months storage.

17α-trenbolone predominates over 17β-trenbolone in manure excreted by treated livestock by a ratio of about 10 to 1. Both metabolites are relatively stable in effluent, with half-lives of about 260 days having been previously reported (Schiffer et al., 2001).

2.6. Environmental fate of steroidal hormones

The fate of any excretion-borne hormones depends upon a number of possible transport pathways and transformation mechanisms. Hormones may be retained in soil or transported by surface run-off or groundwater to surface waters. Manure containing hormones may be stabilised through storage or composting before being spread on paddocks. The available information suggests that steroid hormones slowly degrade in manure, soil and water, but the exact mechanisms or factors controlling the rates are not yet fully understood.

Unconjugated steroidal hormones are chemically very stable, non-volatile, have low water solubility and are moderately hydrophobic (Hanselman et al., 2003; Layton et al., 2000). Their environmental fate from livestock manure depends upon both storage conditions and management (Lange et al., 2002). The degradation of estrogens in separated dairy manure waste solids (press cake) has reportedly been characterised by first-order decay kinetics (Raman et al., 2001). The reported decay constant (K) ranged from 0.029 day^{-1} to 0.12 day^{-1}, increasing with incubation temperature.

Manure and effluent application rates to soils are typically macronutrient-based. The hormone-to-macronutrient ratios effectively determine the rates at which hormones will be applied to soils from manure and effluent. One approach that has been used to predict hormone loading rates in fields receiving dairy wastes has been to determine the mass ratios of specific hormones to the macronutrients nitrogen, phosphorus, and potassium in dairy wastes applied (Raman et al., 2004).

Once released to soils, the environmental fate of steroid hormones depends upon the nature of the soil (Lange et al., 2002). In particular, particle size and organic components strongly affect adsorption and migration in soils. 17β-estradiol and estrone have high sorption affinities to soils (Casey et al., 2003; Casey et al., 2005; Colucci and Topp 2002). The sorption affinity has been well correlated with mineral particle size and organic matter content. Furthermore, there is an apparent relationship with surface area and/or cation-exchange capacity of the soil (Casey et al., 2003). Testosterone appears to behave differently, with lower soil sorption affinity only weakly correlating with soil particle size, organic matter and specific surface area (Casey et al., 2004).

Degradation/transformation of 17β-estradiol occurs in the sorbed phase in soil and is rapid (Casey et al., 2003). Although it was found that testosterone degraded more readily than 17β-estradiol, it appeared to have a greater potential to migrate in the soil because it was not as strongly sorbed (Casey et al., 2004). These results are consistent with field observations where testosterone was shown to reach groundwater, while estrogen remained bound to the upper crust of the soil (Shore and Shemesh, 2003). Both testosterone and estradiol have been measured in surface run-off from soils amended with animal manure (Finlay-Moore et al., 2000).

Synthetic hormones, trenbolone and melengestrol acetate appear to behave similarly to testosterone, having a significant affinity to the organic fraction of soils, leading to a high retardation, but remaining nonetheless mobile in agricultural soils (Lange et al., 2002). In a study undertaken in Germany, traces of trenbolone remained measurable in soil up to eight weeks after application of pre-stored manure and effluent of treated cattle, while melengestrol remained detectable even after cultivation of a maize crop (Schiffer et al., 2001).

Estrogenic, androgenic and progestinal steroidal hormones have been shown to leach into groundwater from dairy waste lagoons (Kolodziej et al., 2004). However, they appear to be strongly absorbed or degraded over distances of 10–100 m in subsurface waters (Kolodziej et al., 2004).

The use of non-chemical-specific screening assays has revealed that feedlot retention ponds and some feedlot effluents in the USA also contain significant concentrations of both estrogenic and androgenic substances and that these may contaminate local watercourses (Soto et al., 2004). Indeed, one study of ponds receiving runoff from US beef cattle farms identified elevated 17β-estradiol concentrations similar to what has been reported in streams receiving sewage treatment plant effluents. The concentrations measured by a radioimmunoassay (ELISA) were 0.05 to 1.80 ng L^{-1} (Irwin et al., 2001).

It has been proposed that overland flow of steroidal hormones during rain events may be a much more significant means of transport to surface waters compared to seepage through soils (Kolodziej et al., 2004). In a few cases, steroidal hormone have been identified in USA regional groundwaters (Peterson et al., 2000) and external surface waters (Kolodziej et al., 2004) where livestock farming has been suspected as the predominant source. However, all Australian cattle feedlots with a capacity of 1000 head or more (and many of the smaller ones) are required to catch and store runoff in effluent holding ponds, preventing direct transport of contaminants in runoff to watercourses.

Few studies have been undertaken to trace the fate of zeranol in the environment. However, interestingly, this compound has been observed at low ng L^{-1} concentrations in municipal sewage effluents and in a river receiving sewage discharges (Laganá et al., 2004, 2001). Nonetheless, given that these studies were undertaken in Europe, transformation of the related myco-estrogen zearalenone would seem a more likely source than residues from growth promotant use.

While data is lacking, it has been suggested that steroidal hormones are biodegraded in the environment by many types of organisms (Hanselman et al., 2003). In soils, estradiol is converted, biotically or abiotically, to estrone, which is slowly further degraded or mineralised (Colucci et al., 2001; Shore and Shemesh, 2003).

3. Antibiotics

Antibiotics include naturally-occurring, semi-synthetic and synthetic chemical compounds with antimicrobial activity. They are used in veterinary medicine to treat and prevent disease, and for other purposes including growth promotion in food animals (Prescott et al., 2000). Depending on their chemical nature, they can be administered orally, parenterally or topically. Antibiotics are commonly administered to livestock in the USA and Australia via feed or water or via a slow-release implant (Page, 2003; Shuford and Patel, 2005).

Antibiotics used to treat and prevent disease may be undertaken as therapy (for animals exhibiting clinical disease), control (administration to a herd to control the spread of disease) or prevention (administration to healthy animals to

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prevent onset of disease) (National Committee for Clinical Laboratory Standards, 2002).

Antibiotic use for growth promotion involves the administration of an antibiotic agent, usually as a feed additive over a period of time, which results in improved physiological performance. Many antibiotics are known to improve average daily weight gain and feed efficiency in livestock in a variety of applications (Gaskins et al., 2002; Lefebvre et al., 2006; Nagaraja and Chengappa, 1998). Some growth-promoting effects involve the alteration of the normal intestinal microbiota, resulting in more efficient feed digestion and metabolism (Dennis et al., 1981; Nagaraja et al., 1987). Others are the result of pathogen and disease suppression and immune system release.

Feedlot cattle are highly susceptible to rumenitis and hepatic abscessation as a result of high carbohydrate diets (Montgomery et al., 1982; Nagaraja and Chengappa, 1998; Page, 2003). Continuous inclusion of antibiotics in the diet can significantly reduce these afflictions (Page, 2003).

A wide range of antibiotic agents are registered for therapeutic use on cattle in Australia. Some of the most important include benzathine penicillin, procaine penicillin, ampicillin, amoxycillin, cloxacillin, cefuroxime, cephalonium dihydrate, cefuroxime, ceftiofur, erythromycin, oxytetracycline, sulfadiazine, sulfadimidine, sulfadoxine, dihydrostreptomycin, novobiocin, trimethoprim, florfenicol, neomycin, tylosin and flunixin (APVMA, 2006). By virtue of their use, all of these compounds have some potential to be present in livestock wastes.

However, the agents that are used 'enterically' as feed additives are likely to be significantly more important feedlot contaminants than the agents used only intermittently to treat illness. There are more than 30 antibiotic products registered for such 'enteric' uses in Australia (APVMA, 2006). Of these, monensin is the most common active ingredient, but others include salinomycin, tylosin, lasalocid, oxytetracycline, narasin, and neomycin. Virginiamycin is also registered, but growth promotant claims have been voluntarily withdrawn by the manufacturer, limiting its use to therapeutic applications.

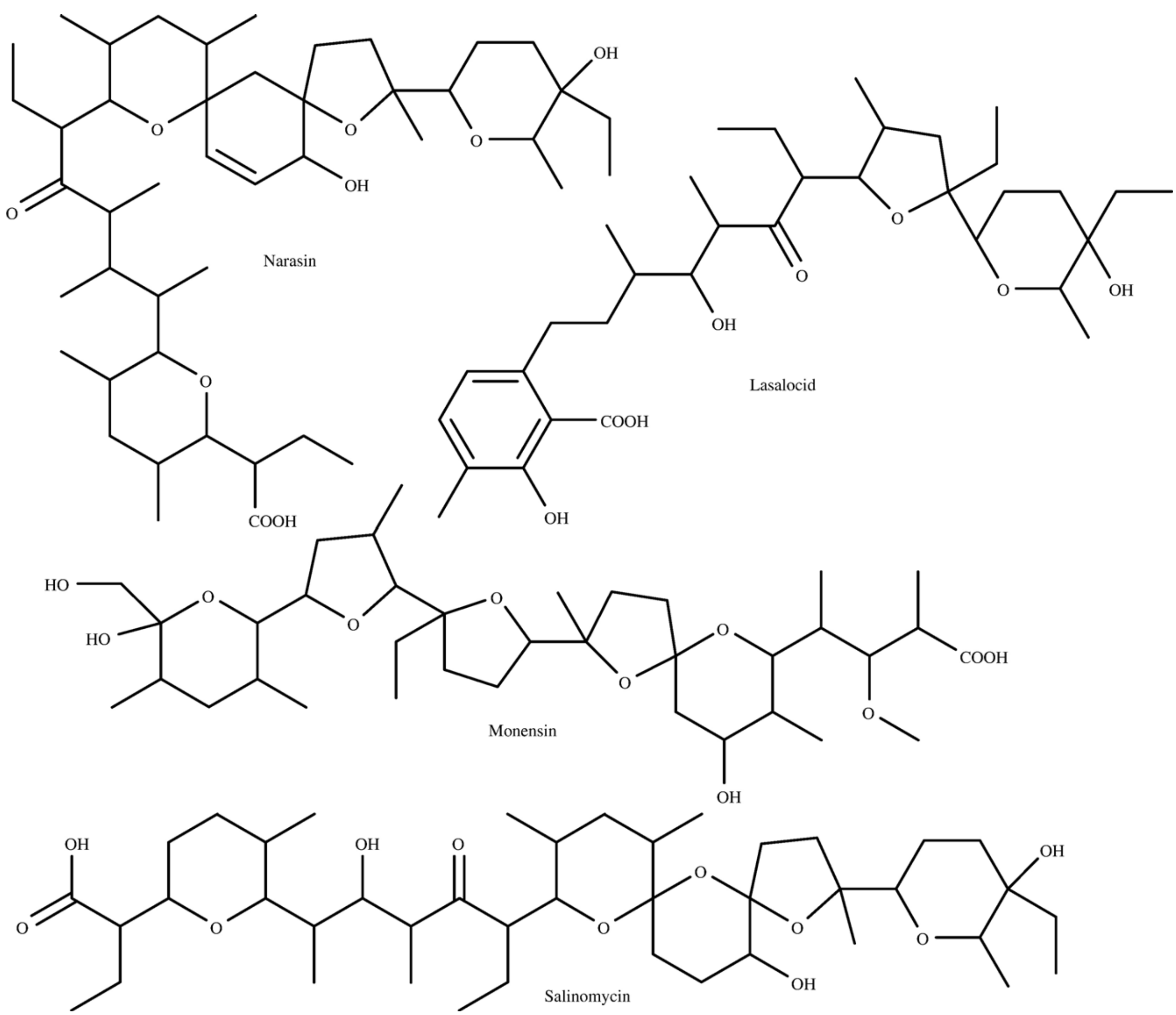

Fig. 5. Molecular structures of polyether ionophores.

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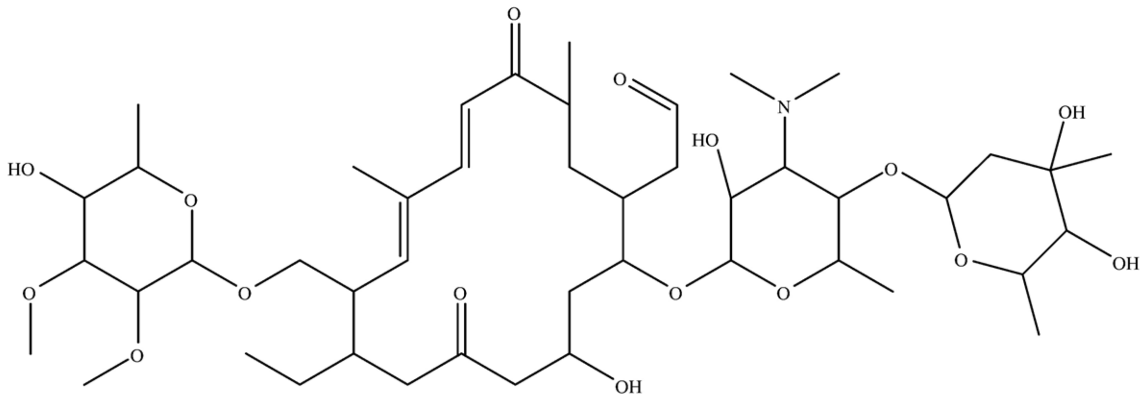

Fig. 6. Molecular structure of tylosin.

3.1. Polyether ionophores (lasalocid, monensin, narasin, and salinomycin)

Polyether ionophores (lasalocid, monensin, narasin, and salinomycin) are used to improve feed conversion efficiency in feedlot beef cattle (Page, 2003). Salinomycin is also known to increase the rate of weight gain, thus enhancing productivity. Lasalocid and Monensin are further used to improve milk production in dairy cows. Approved polyether ionophores are typically included in cattle diets at rates of 5–33 g per tonne of feed (Page, 2003). Molecular structures are provided in Fig. 5.

The polyether ionophores are toxic to many bacteria, protozoa, fungi and higher organisms. Their three-dimensional conformation creates a highly hydrophobic exterior and a hydrophilic interior, enabling the binding of one or more cations. The lipophilic nature allows ready penetration of cell membranes, enabling uncontrolled influx and/or efflux of selected ions, such as potassium and sodium, from the cell. This osmotic interference often leads to cell death. Ionophores are not currently used in human medicine since they are absorbed and are toxic.

3.2. Tylosin

Tylosin (Fig. 6) is a macrolide antibiotic agent produced from *Streptomyces* spp. bacteria. Tylosin is used in Australia for the reduction of incidences of liver abscess in beef cattle (Page, 2003) and has been shown to be around 40–70% effective for doing so (Nagaraja and Chengappa, 1998). Tylosin is typically given to Australian beef cattle at a rate of 11 g per tonne of feed (Page, 2003).

Macrolides are able to penetrate host cells, and their therapeutic action is, to some extent, a reflection of their intracellular concentration. Typically, they accumulate rapidly

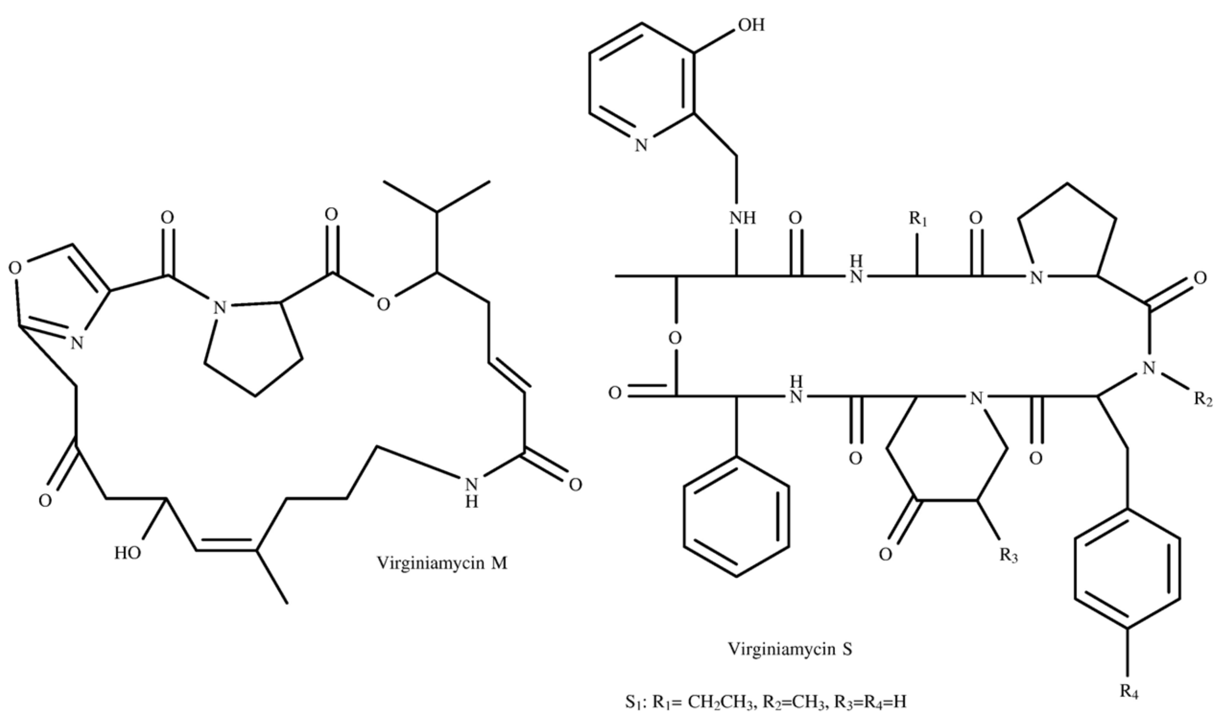

Fig. 7. Molecular structure of virginiamycin.

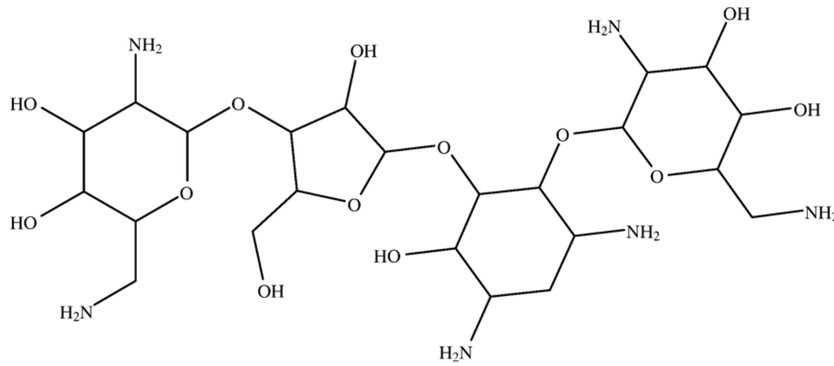

Fig. 8. Molecular structure of neomycin.

to a saturation point both in the cytoplasm and in the intracellular granules. As a group the macrolides are particularly valuable for the treatment of cell-associated pathogens such as mycoplasmas.

3.3. Virginiamycin

Virginiamycin (Fig. 7) belongs to the streptogramin class of antibiotic agents. Commercial formulations consist of a mixture of virginiamycin S and virginiamycin M which together are bactericidal. Virginiamycin acts in the 50 S bacterial ribosome at a position very close to the site of action of macrolide antibiotics. It is used in Australia to reduce the risk of lactic acidosis in cattle that are fed a high grain diet. Virginiamycin is currently approved for use in beef and dairy cattle at a rate of 20 g per tonne of feed (Page, 2003), however the approved uses are subject to on-going review in Australia (APVMA, 2003).

3.4. Neomycin

Neomycin (Fig. 8) is an aminoglycoside antibiotic. It is available in Australia as a feed additive only for the treatment of bacterial enteritus (scours). It is not registered for use as a growth promotant and treatment via feed is restricted to 3–5 days (APVMA, 2006).

3.5. Oxytetracycline

Oxytetracycline (Fig. 9) is not registered as a growth promoting agent in Australia, however it is occasionally used as a feed-additive to control infectious cattle diseases caused by the effects of transport (APVMA, 2006).

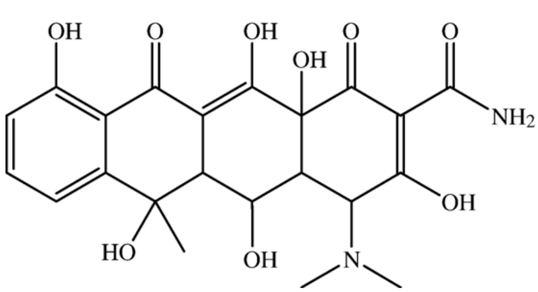

Fig. 9. Molecular structure of oxytetracycline.

3.6. Environmental and public health concerns

There are a number of concerns regarding residual antibiotics for livestock production. The primary concern is the contribution that livestock antibiotic use may make to the increasing incidence of antibiotics resistance among a wide variety of pathogenic bacteria (Barton et al., 2003). A second concern regards potential deleterious effects that these compounds may have on natural soil bacterial populations.

Antibiotic resistance is a major problem facing modern medicine. Microorganisms with increasing rates of resistance to commonly used antibiotics include enteric Gram-negative bacilli (*Klebsiella* and *Enterobacter* species) resistant to extended-spectrum β-lactams, *Streptococcus pneumoniae* resistant to penicillins, methicillin-resistant *Staphylococcus aureus*, vancomycin-resistant enterococci, and *Shigella* and *Salmonella* species resistant to multiple antibiotics (Conly, 2002; Nimmo et al., 2003). Bacterial isolates, recovered from humans, cattle, pigs and food show widespread resistance to tetracycline, sulfamethoxazole, cephalothin and ampicillin (Schroeder et al., 2002). Cattle isolates show common resistance to numerous antibiotics including sulfamethoxazole, tetracycline, ampicillin and neomycin (Cergole-Novella et al., 2006; Stephens, 2003).

Numerous studies published in the medical literature have suggested a relationship between clinical antibiotic-resistant infections and the use of antibiotics in agriculture (Phillips et al., 2004; Shuford and Patel, 2005; Smith et al., 1999), however evidence of such a relationship remains controversial (Phillips et al., 2004). Furthermore, within Australia, most of the enteric antibiotics used in cattle production are not closely related to important antibiotics for human health. This applies to the polyether ionophores (lasalocid, monensin, narasin and salinomycin) and neomycin.

Tylosin belongs to the *macrolide* class of antibiotics, which also includes some important drugs used widely for human health in Australia (erythromycin, roxithromycin, clarithromycin, clindamycin, azithromycin, and lincomycin) (Australian Government Department of Health and Aging, 2005). However, molecularly, tylosin is a somewhat 'aberrant' macrolide in that the usual 14-membered ring has been widened to a 16-membered ring (Fig. 6).

Virginiamycin is a streptogramin antibiotic. It is not used for human health, although it is very close to another streptogramin known as quinupristin–dalfopristin, which is of great significance. Quinupristin–dalfopristin is not widely used in Australia but is kept as a last-defence for the treatment of Methicillin Resistant *Staphylococcus aureus* or Vancomycin Resistant *Enterococcus faecium* infections. The veterinary use of virginiamycin has been subject to close scrutiny in Australia because of strong concerns regarding the relationship between virginiamycin resistance and conferred quinupristin–dalfopristin resistance as well as vancomycin resistance (APVMA, 2003).

Oxytetracycline is a member of the chemically homologous group of antibacterial drugs known as tetracyclines. Oxytetracycline is not used for human health in Australia, however some other very closely related tetracyclines are. These include doxycycline, tetracycline, minocycline and demeclocycline hydrochloride (Australian Government Department of Health and Aging, 2005). The potential for overexposure of some bacteria to oxytetracycline is of some concern since bacterial acquisition of resistance to this drug may provide resistance to the entire class of tetracycline antimicrobials (McDermott et al., 2003).

Bacterial populations intrinsically resistant to antibiotics exist in the environment. Exposure to antibiotic drugs may provide a selective pressure for these organisms to increase in dominance (Schroeder et al., 2002). Furthermore, exposure to antibiotics may induce genes coded for resistance that are subsequently transferred to other members of the microbial community (Hughes and Datta, 1983; Lipsitch and Samore, 2002).

Considerable increases in antimicrobial resistance have been reported in faecal samples excreted by lot fed beef cattle in Canada (Inglis et al., 2006). Importantly, substantial increases in the prevalence of resistance to the human-health antibiotics, tetracycline and doxycycline were reported in this study for animals administered chlortetracycline and oxytetracycline in feed and/or long-acting injection. Some increased resistance to erythromycin was also observed. Dairy farm manure has also been shown to contain significant concentrations of multiple antibiotic-resistant bacteria (Esiobu et al., 2002; Murinda et al., 2005).

Tetracycline resistance levels in soil are temporarily influenced by the increasing addition of pig-manure slurry (Sengelov et al., 2003). Similarly, chorotetracycline and tylosin resistance levels in soils have been shown to temporarily increase after amendment with manure (Halling-Sorensen et al., 2005). For streptomycin and erythromycin, only minor variations in resistance levels have been reported (Sengelov et al., 2003).

Microbial profiles collected from surface waters in the USA, Europe and Asia also show significant levels of resistance to diverse antibacterial agents (Arvanitidou et al., 1997; Ash et al., 2002; Park et al., 2003). This may be, in part, due to run off from manure-fertilised agricultural soils, but other sources such as sewage treatment plants may also be of significance.

The World Health Organisation discourages the enteric use of antibacterial agents that belong to classes used for human health or that could create cross-resistance to them (World Health Organization, 1997). Largely in response to this, the European Union has imposed bans on the use of avoparcin, bacitracin, spiramycin, tylosin and virginiamycin for growth promotion (Shuford and Patel, 2005).

In addition to the potential influence that these compounds may have on the development of bacterial resistance, there is also concern regarding their toxicity toward aquatic organisms (Kümpel et al., 2001). In terms of toxicity, a particular complication for antibiotics is that standardised tests, such as the OECD respiration inhibition test, may not be suitable to assess the effects of these agents on environmental bacteria (Kummerer et al., 2004). Alternatively, manometric respiration tests have been proposed to assess the effects of veterinary antibiotics in soil (Vaclavik et al., 2004). Such tests provide details of relative antimicrobial potency towards soil microorganisms and can be used to rank compounds.

3.7. Environmental fate of antibiotics

Many antibiotic compounds are only partially degraded during metabolism by humans and other animals, and thus are excreted largely unchanged. Accordingly, animal excrements following antibiotic use for treatment or growth promotion are considered to be important sources of these compounds to some affected environments (Boxall et al., 2000; Hamscher et al., 2000; Jorgensen and Halling-Sorensen, 2000; Patten et al., 1980). Some antibiotics, such as tylosin and oxytetracycline sorb relatively strongly to manure particles (Loke et al., 2002).

The storage of liquid manure prior to use as a fertiliser can enhance the degradation of some antibiotic agents, although some others are more persistent (Schlusener et al., 2006). In a 180-day study, degradation half-lives were determined for erythromycin (41 days), roxithromycin (130 days) and salinomycin (6 days). However the concentration of one compound, tiamulin (which is not administered to Australian cattle), remained unchanged during the entire storage period.

Where contaminated manure and effluent are used to fertilise agricultural soils, loads of up to kilograms of antibiotic per hectare may be reached (Thiele-Bruhn, 2003). Considerable variation exists, but some antibiotic agents appear to be very persistent in soils (Gavalchin and Katz, 1994; Hamscher et al., 2000). For example, tetracyclines have been identified on manure-fertilised fields in Germany at soil depths of up to 30 cm (Hamscher et al., 2000).

The different classes of antibiotic drugs vary considerably in their molecular structures, and thus in their physical, chemical and biochemical properties. Some substances are very hydrophobic, while others are completely water soluble (United States Pharmacopeia, 2000). Accordingly, distribution coefficients to describe the adsorption of antibiotics to soils or dissolution in water can vary across at least five orders of magnitude (Thiele-Bruhn, 2003; Tolls, 2001). For this reason, it is not possible to provide a widely generalised description of antibiotic fates in soils.

For some antibacterial agents estimation of the partitioning coefficients cannot be made from measures of hydrophobicity

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(e.g. log K_{ow}) alone (Tolls, 2001). For example, sorption of oxytetracycline appears to be strongly influenced by ionic binding to divalent metal ions as such Mg^{2+} and Ca^{2+} as well as other charged compounds in the matrix (Loke et al., 2002). Soil texture, cation exchange capacity and iron oxide content appear to be among the most significant factors determining oxytetracycline binding (Jones et al., 2005). The adsorption of some compounds is also significantly influenced by soil pH (Boxall et al., 2002) and overall ionic strength (Sithole and Guy, 1987).

Some antibiotic compounds have the potential to leach through soil or with surface run-off during rain events and contaminate local groundwater and surface waterways. For example, multiple classes of antibacterial compounds have been reported in surface and groundwater samples collected proximal to pig and poultry farms in the USA (Campagnolo et al., 2002). Agriculturally-derived antibiotics have also been identified in surface waters in Colorado, USA (Cha et al., 2005; Yang and Carlson, 2003). A US Geological Survey study on the occurrence of pharmaceuticals in surface waters identified a number of antibiotics that are not used for human therapy in the US (Kolpin et al., 2002). The relatively high frequency of detection of these agricultural antibiotics may be indicative of the potential for surface water contamination by these chemicals.

Antibacterial agents are susceptible to biotic degradation by microbial organisms present in manure and soil (Ingerslev and Halling-Sorensen, 2001). However, biodegradation rates (or half lives) vary between different classes of compounds and are also influenced by environmental conditions including available oxygen and the specific nature of the microbial community (Ingerslev et al., 2001). Dissipation half-lives vary with soil type and have been estimated for chlorotetracycline (25–34 days) tylosin (49–67 days) (Halling-Sorensen et al., 2005). Other examples of degradation rates of antibiotics in manure and soils have been compiled from the literature by Thiele-Bruhn (2003).

3.8. Antibiotic resistance genes

In addition to the influence of antibiotic agents themselves, microbial resistance to antibiotics can also be disseminated by the spread of resistant organisms and the resistance genes that they carry (Hughes and Datta, 1983). Antibiotic resistance to many agents is widespread in some environments and much of this resistance is the result of genetic coding that may be transferred between organisms (Ash et al., 2002).

For example, among sixteen US rivers, over 40% of bacteria resistant to one or more antibiotics had at least one plasmid that coded for resistance and genes resistant to ampicillin were detected in 70% of the isolated plasmids (Ash et al., 2002). Similarly, 24% of surface water *Salmonella* strains tested in Greece exhibited resistance to at least one of 20 antimicrobials and 26% were able to transfer resistance to *E. coli* (Arvanitidou et al., 1997).

Furthermore, evidence is mounting to support the hypothesis that intestinal bacteria not only exchange resistant genes among themselves, but might also interact with bacteria that are passing through the colon, causing these bacteria to acquire and transmit antibiotic resistance genes (Salyers et al., 2004; Salyers and Whitt, 2005).

The primary route of genetic transfer between different genera of bacteria (ie: horizontal transfer) is believed to be via a process known as *conjugation* (Davies, 1994; Seveno et al., 2002). Many examples have been reported of horizontal gene transfer between bacterial species, genera or families and even between bacteria and eukaryotes (Amabilecuevas and Chicurel, 1992).

An alternative pathway for gene transfer is believed to be via *natural transformation* involving the uptake of DNA present in the environment (Seveno et al., 2002). Released into the aquatic environment, DNA is rapidly degraded. However, when bound to soils, it may persist for up to a period of months (Nielsen et al., 1997; Romanowski et al., 1993).

It is believed that bacterial natural transformation may be a significant gene transfer process in nature, at least under some conditions (Baur et al., 1996; Lorenz and Wackernagel, 1994; Webb and Davies, 1993). However, little is known about the relative importance of gene transfer via transformation as the frequency of such occurrences is yet to be properly validated (Seveno et al., 2002).

Recent attention has turned to the role that cattle rumen protozoa may play in the gene transfer between microorganisms (McCuddin et al., 2006). Rumen protozoa are predators of bacteria and thus can harbour antibiotic resistance genes. Evidence has been reported for the involvement of rumen protozoa in the antibacterial-resistance gene transfer between two bacterial species, *Salmonella* and *Klebsiella* (McCuddin et al., 2006).

Animal manure is also considered to be a likely environment to favour the horizontal transfer of genes (Gotz and Smalla, 1997; Seveno et al., 2002). This is because of the combination of high bacterial survival and activity, coupled with the selective pressure resulting from the presence of antibiotics in animal faeces. Seveno et al. (2002) report that before the use of the streptothricin antibiotic 'nourseothricin' as a feed additive in some countries, resistance quotients for coliforms in manure were approximately 0.1–1%. However, after the inclusion of nourseothricin, resistance quotients of up to 80% were observed. Furthermore, plasmid-borne resistance of this drug was detected among strains isolated from pigs, and subsequently in isolates from manure, river water, food and human beings (Smalla et al., 2000; Tschape, 1994).

In soil environments, gene transfer frequencies are thought to be low, limited mainly by nutrient availability which in turn limits bacterial density and activity (Smit et al., 1998). Transfer is also affected by other factors such as soil moisture, pH and temperature (van Elsas et al., 1998, 1988). However, the areas around plant roots are often very high in the amount and diversity of nutrients and thus these sites can be 'hot spots' for gene transfer (Seveno et al., 2002).

4. Ectoparasiticides

Externally used parasiticides, known as 'ectoparasitides' are chemical formulations used to control external parasites (ectoparasites) such as ticks, flies and lice on livestock including cattle

(Waltisbuhl et al., 2005). A number of application methods are used including dips, sprays, pour-on solutions, ear tags and back rubbers (Reeves, 2005; Spence, 1994). Back rubbers are provided for self-treatment by cattle.

Many ectoparasitic infections are seasonal and therefore predictable. Accordingly, they can be countered by prophylactic use of the ectoparasiticides (Taylor, 2001). The available chemicals may act either systemically, following dermal uptake by the host, or simply by direct contact with the target parasites (Taylor, 2001).

The regulatory framework for the management of ectoparasiticides in Australia is administered by Australian Pesticides and Veterinary Medicines Authority (APVMA, 2006). Product registration involves rigorous scientific assessment aimed at ensuring effective and safe use, as well as efforts to make sure that environmental risks can be controlled (Reeves and Ashton, 2005).

There are currently more than 170 registered cattle ectoparasiticide products in Australia (APVMA, 2006). These contain a range of chemical compounds including chlorinated hydrocarbons (now discontinued), organophosphates, carbamates, synthetic pyrethroids, amidines, macrocyclic lactones and tick development inhibitors (Holdsworth, 2005b).

4.1. Chlorinated hydrocarbons

Chlorinated hydrocarbons (e.g.: DDT, benzene hexachloride (BHC), chlordane, heptachlor, dieldrin, aldrin, methoxychlor and toxaphene) were common cattle ectoparasiticides in Australia from the mid 1940s (Holdsworth, 2005a). However, their use was banned in 1962 due to their accumulation in animal tissue (particularly the fat), and the need to comply with pesticide residue standards set by countries importing Australian meat (Holdsworth, 2005a).

Since chlorinated hydrocarbons have not been registered for ectoparasiticidal use in Australia now for more than 40 years, they are not reviewed in detail here. Although these compounds are notoriously environmentally resilient, it is very unlikely that feedlot sites would be contaminated with them since almost all Australian feedlots were constructed since the ban. Furthermore, regular testing for these chemicals is undertaken at the time of animal processing and any detection above half the maximum residue limit is investigated.

4.2. Organophosphates and carbamates

The use of chlorinated hydrocarbons was predominantly replaced by the then newly available organophosphate compounds (e.g.: diazinon, dioxathion, carbophenothion, coumaphos and ethion) and carbamate compounds (e.g.: carbaryl, promacyl, and bendiocarb) (Holdsworth, 2005a). These chemicals work by disrupting the function of acetylcholinesterase enzymes, leading to neuromuscular paralysis (Taylor, 2001).

The use of some organophosphates and carbamates has receded during the last few decades due to the widespread development of resistance by many ectoparasites. However, many

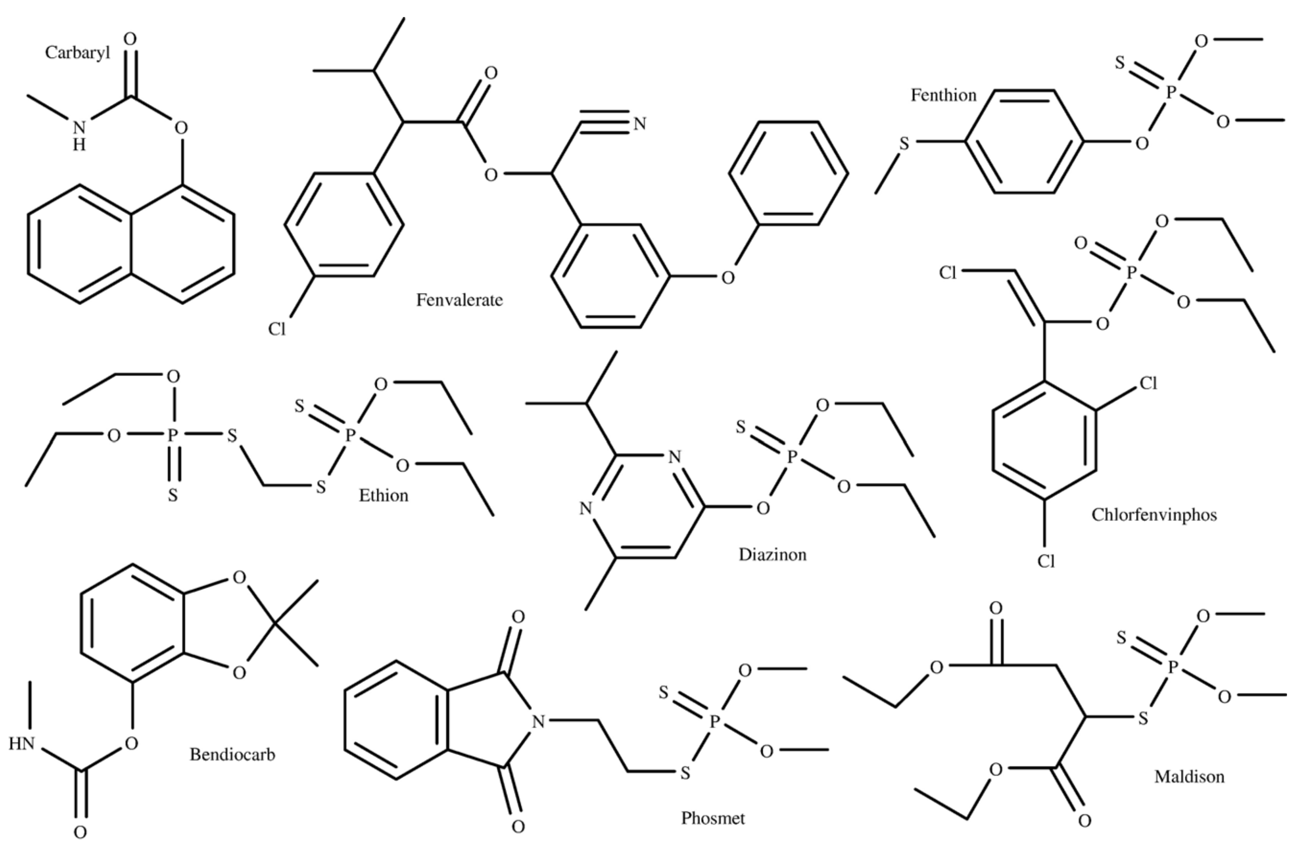

Fig. 10. Molecular structures of organophosphates and carbamates.

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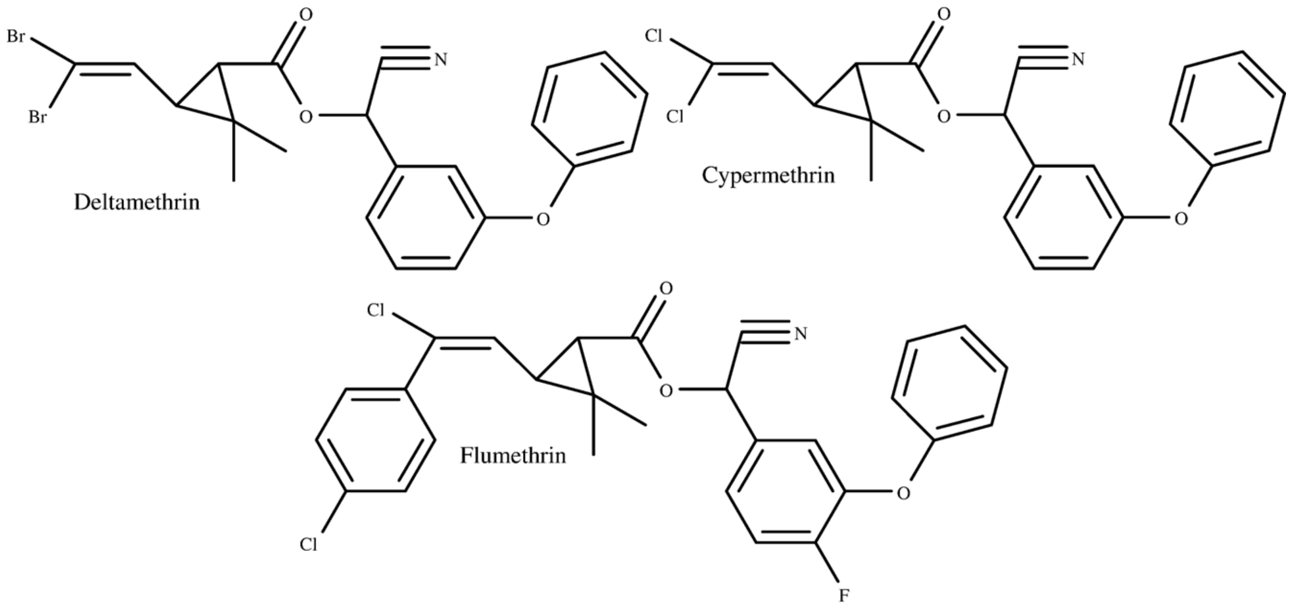


Fig. 11. Molecular structures of synthetic pyrethroids.

continue to be used and administered through back-rubbers, ear-tags, dips, pour-on preparations, and sprays (Holdsworth, 2005a). Registered organophosphate and carbamate cattle ectoparasiticides in Australia include fenthion, fenvalerate, phosmet, maldison, chlorfenvinphos, diazinon, ethion, carbaryl and bendiocarb (Fig. 10).

4.3. Synthetic pyrethroids

Synthetic pyrethroids are a relatively recent class of ectoparasiticide, having first been registered for use in Australia in 1980 (Holdsworth, 2005a). They are used on Australian cattle to control buffalo flies, lice and ticks via sprays, dips, pour-on formulations and ear tags. Synthetic pyrethroids are synthesised chemicals modelled on the natural insecticide, pyrethrin (Taylor, 2001). They are lipophilic molecules and undergo rapid adsorption, distribution and excretion. Their effectiveness is believed to be due to their ability to interfere with sodium channels of insect nerve axons resulting in eventual paralysis (Taylor, 2001). Registered synthetic pyrethroids for use as cattle ectoparasiticides in Australia include deltamethrin, cypermethrin and flumethrin (Fig. 11).

4.4. Amidines

Amidines (e.g.: chlorphenamide, clenpyrin, cymiazole and amitraz) have been used in Australia to treat lice and ticks by plunge dipping and spraying (Holdsworth, 2005a). However, amitraz (Fig. 12) is the major amidine currently used, primarily to kill ticks on beef and dairy cattle. It acts at octopamine receptor sites, resulting in hyperexcitability and death of the organism (Roeder, 1995). Amitraz is usually applied in dips, sprays and pour-on solutions.

4.5. Macrocyclic lactones

Macrocyclic lactones have been employed as ectoparasiticides in Australia since 1985 (Holdsworth, 2005a). The macrocyclic lactones registered for use on cattle include abamectin, doramectin, ivermectin, eprinomectin (Fig. 13) and moxidectin. These compounds are produced as fermentation products of the soil microorganisms *Streptomyces avermilitis* and *Streptomyces cyanogriseus* (Fisher and Mrozik, 1992; Shoop et al., 1995). Most are supplied as pour-on treatments for cattle ticks, buffalo flies, lice and mites. However, some are also approved for subcutaneous injection for the control of lice, mites, mange and ticks. Macrocyclic lactones are highly lipophilic and, following administration, are stored in animal fat tissue from where they are slowly released, metabolised and excreted, primarily in the faeces (Taylor, 2001).

4.6. Benzylphenyl ureas

Benzylphenyl ureas inhibit the production of chitin, a complex aminopolysaccharide and a major component of insect cuticles (Taylor, 2001). They are highly lipophilic molecules and accumulate in body fat from where they are slowly released into the bloodstream and excreted largely unchanged.

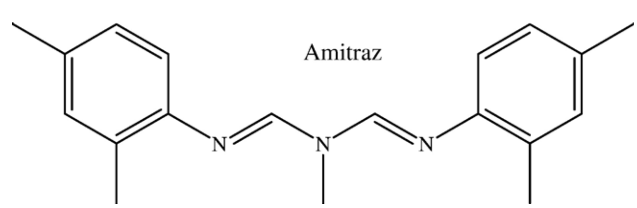
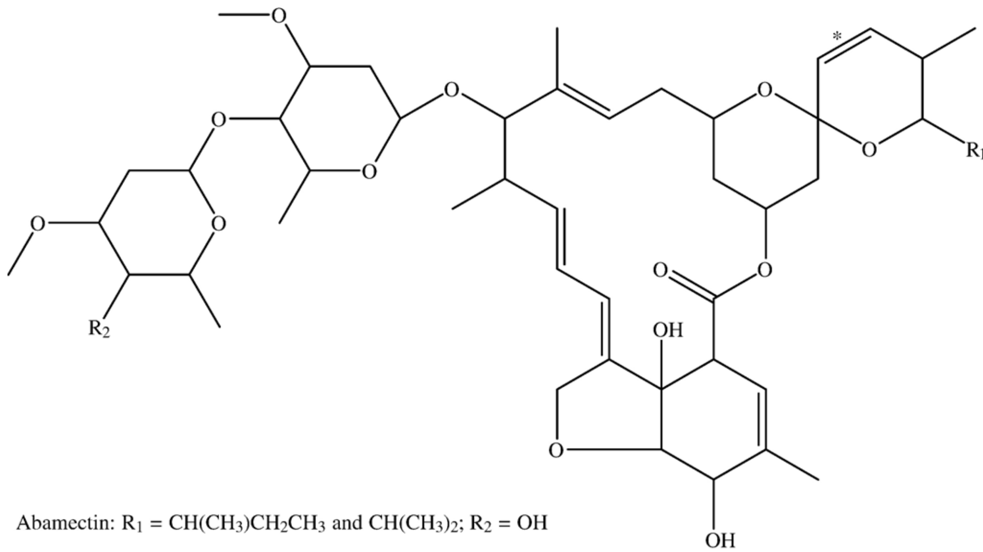


Fig. 12. Molecular structure of amitraz.



Abamectin: $R_1 = CH(CH_3)CH_2CH_3$ and $CH(CH_3)_2$; $R_2 = OH$

Doramectin: R_1 = Cyclohexyl; $R_2 = OH$

Ivermectin: $R_1 = CH(CH_3)CH_2CH_3$ and $CH(CH_3)_2$; R_2=OH; * Marked bond saturated ($-CH_2-CH_2-$)

Eprinomectin: $R_1 = CH(CH_3)CH_2CH_3$ and $CH(CH_3)_2$; $R_2 = NHCOCH_3$

Fig. 13. Molecular structures of some macrocyclic lactones.

Fluazuron (Fig. 14) is a benzylphenyl urea tick-development inhibitor used as a pour-on solution to control cattle ticks in Australia. It acts systemically on all developmental stages of ticks as they feed and breed on cattle (Holdsworth, 2005a).

4.7. Environmental and public health concerns

Due to their chemical diversity, ectoparasiticides vary considerably in their metabolism and excretion behaviour by cattle. Some organophosphate and carbamate compounds may be expected to be efficiently metabolised, while other classes including the synthetic pyrethroids, benzylphenyl ureas, macrocyclic lactones and fluazuron are known to be passed into manure largely unchanged (Fisher and Mrozik, 1992; Lumaret and Errouissi, 2002; Taylor, 2001; Vale et al., 1999; Wardhaugh, 2005).

Ectoparasiticides are, by design, highly toxic to numerous organisms and are typically selected to target the arthropod nervous system. There are significant concerns regarding the potential impacts of some compounds on non-target biota (Floate et al., 2005; Hill, 1989; Kolar and Kožuh Eržen, 2006; McKellar, 1997). Some, such as organophosphates and carbamates, are anticholinestrases and due to their wide agricultural application have frequently been associated with toxicity to wildlife (Ragnarsdottir, 2000).

Furthermore, some ectoparasiticides have been associated with concerns regarding human health (Taylor, 2001). Oral reference doses for a number of important ectoparasiticides have been allocated by the US EPA Integrated Risk Information System (IRIS) or the US EPA Health Effects Assessment Summary Tables (HEAST) as shown in Table 1 (US EPA, 2007a, 2007b). Furthermore, a cancer slope factor of 2.4×10^{-2} mg kg^{-1} day^{-1} has been allocated for the organophosphate, tetrachlorvinphos by IRIS (US EPA, 2007b). Numerous cases have been reported where farmers' health has been affected by organophosphate exposure through animal dipping and spraying of fields (Ragnarsdottir, 2000). On the other hand, a variety of *in vitro* and *in vivo* studies have indicated minimal acute toxicity of the macrocyclic lactones ivermectin and abamecton towards humans (Fisher and Mrozik, 1992).

The vast majority of ectoparasiticides are neurotoxins, working by disrupting parasite nervous systems (Taylor, 2001).

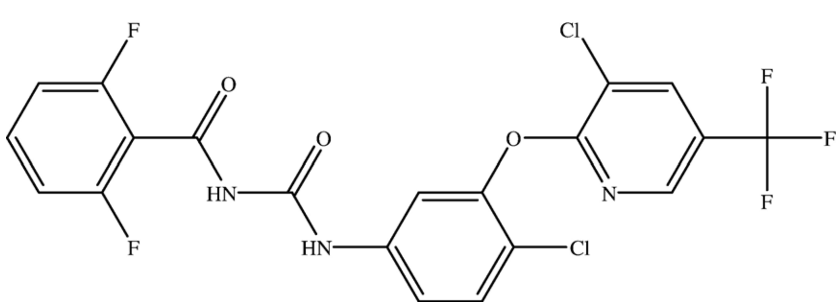


Fig. 14. Molecular structure of fluazuron.

Table 1
Oral references doses (RfDo) for parasiticides

	RfDo (mg kg^{-1} day^{-1})	Reference
Abamectin	4.0×10^{-4}	US EPA (2007b)
Amitraz	2.5×10^{-3}	US EPA (2007b)
Chlorpyrifos	3.0×10^{-3}	US EPA (2007b)
Cypermethrin	1.0×10^{-2}	US EPA (2007b)
Diazinon	9.0×10^{-4}	US EPA (2007a)
Diflubenzuron	2.0×10^{-2}	US EPA (2007b)
Ethion	5.0×10^{-4}	US EPA (2007b)
Fenvalerate	2.5×10^{-2}	US EPA (2007b)
Maldison	2.0×10^{-2}	US EPA (2007b)
Permethrin	5.0×10^{-2}	US EPA (2007b)
Temephos	2.0×10^{-2}	US EPA (2007a)
Tetrachlorvinphos	3.0×10^{-2}	US EPA (2007b)

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Accordingly, they are generally very non-specific in their activeness. Others work by the inhibition of important insect enzyme functions or by disrupting moulting processes (Taylor, 2001).

Particular concern has arisen for the unintended effects of ectoparasiticides on manure fauna (Lumaret and Errouissi, 2002; Wardhaugh, 2005). Manure fauna play a vital role in the processes of manure degradation, nutrient cycling and pasture hygiene. Macrocyclic lactones, in particular, are believed to pose a broad threat to the survival of manure-dependant organisms with reported impacts including larval mortality, mortality of immature adults, reduced egg production, and delayed reproductive development (Ridsdill-Smith, 1993; Wall and Strong, 1987; Wardhaugh et al., 1998, 1993). Toxicity towards manure-dwelling insects has also been reported after topical treatment of cattle with various synthetic pyrethroids (Kruger et al., 1999; Sommer et al., 2001; Vale et al., 1999; Wardhaugh et al., 1998). As a consequence of reduced insect activity, animal manure degradation has been shown to have been retarded in numerous studies (Lumaret and Errouissi, 2002).

Faecal residues or metabolites may be highly toxic to soil-dwelling organisms (Floate et al., 2005; Lumaret and Errouissi, 2002). The potential contamination of surface waters by ectoparasiticides may also be a significant concern since many aquatic organisms are sensitive to relatively low concentrations of pesticides (Halley et al., 1989, 1993; Kolar and Kožuh Eržen, 2006).

Exposure of some organisms to low (non lethal) concentrations of ectoparasiticides is believed to have significant potential to promote the development of resistance to these agents (Hennessy, 1994). This may be expected to have implications for future feedlot management.

4.8. Environmental fate of ectoparasiticides

Organophosphate chemicals are relatively water soluble and are transported readily through soils and into groundwater or surface waters (Ragnarsdottir, 2000). Chemical and biochemical degradation of organophosphates may occur by oxidative or reductive mechanisms or by hydrolysis or photolysis under suitable conditions (Ragnarsdottir, 2000).

Synthetic pyrethroids are non-polar compounds and sorb strongly to soil (Elliott, 1989; Pawlisz et al., 1998). They are considered to be very easily degraded in the environment, primarily by photochemical and biochemical mechanisms (Demoute, 1989). Nonetheless, persistence in soils can be quite variable depending upon soil type as well as ambient conditions (Pawlisz et al., 1998). Furthermore, synthetic pyrethroids contain a number of chiral centres and the consequent stereoisomers have been shown to be variably degraded by biological transformations (Qin et al., 2006).

Macrocyclic lactones tend to have very high organic carbon sorption coefficients and thus are highly sorbed to soils and have limited water solubility (Kolar and Kožuh Eržen, 2006). As a consequence of their characteristically tight binding to soils, the macrocyclic lactones are highly immobile and considered to be unlikely to pose a significant threat to waterways (Halley et al., 1989, 1993).

While high concentrations of abamectin and doramectin have been reported in sheep manure for numerous weeks following treatment, impacted soil concentrations were determined to be much lower and effectively dissipated after a few days, indicating that field conditions have an important role in the degradation of these agents (Kožuh Eržen et al., 2005).

Macrocyclic lactones such as abamectin and ivermectin appear to be rapidly degraded upon exposure to sunlight (Fisher and Mrozik, 1992; Halley et al., 1989). These chemicals have also been shown to be susceptible to aerobic biodegradation in soils under suitable conditions (Halley et al., 1993). However ivermectin appears to be quite persistent in manure under suitable conditions (Sommer and Steffansen, 1993).

5. Mycotoxins

Mycotoxins are toxic chemicals produced by fungi such as mould (Newsome, 2006). Thousands of mycotoxins exist and infestation of cereal grains is common (Murphy et al., 2006). It is generally assumed that mycotoxins are of greatest concern in developing countries where climatic conditions, and agricultural and storage practices, are considered conductive to fungal growth and toxin production (Aziz et al., 2006; Singhal and Kaur, 2005). However, mycotoxin contamination of forages and feeds is reported to be common on European farms (Wilde, 2005). Many mycotoxins possess acute and chronic toxicity at low concentrations and some are mutagenic, carcinogenic or teratogenic to a wide range of organisms causing hepatic carcinoma even in humans (Groopman et al., 1988).

Ingestion of contaminated feed has been associated with a range of animal disease incidences internationally (Ozsoy et al., 2005; Seeling and Danicke, 2005; Singhal and Kaur, 2005). The effects of mycotoxins in dairy cattle are reported to result in symptoms that are often non-specific or similar to other diseases and nutritional disorders and, as such, are often unrecognised (Wilde, 2005).

Ochratoxin is a mycotoxin produced by several strains of *Penicillium* and *Aspergillus* spp. Ochratoxin A (Fig. 15) is the

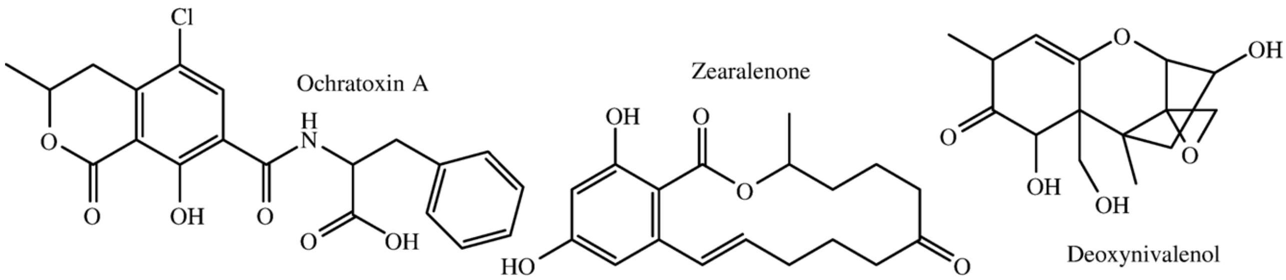

Fig. 15. Molecular structures of some common mycotoxins.

main toxin in this group. It is found in infected wheat, corn, and oats, and cheese and meat products of animals consuming ochratoxin-contaminated grains (Aish et al., 2004; Murphy et al., 2006). Although the toxin is reported to occur in foods around the world, the main regions of concern are Europe and, for some foods, Africa (Murphy et al., 2006).

Zearalenone (Fig. 15) is an estrogenic mycotoxin or “mycoestrogen” for which, genotoxicity is also a reported concern (Le Guevel and Pakdel, 2001). Occasional outbreaks of zearalenone mycotoxicosis in livestock around the world are known to cause infertility (Murphy et al., 2006; Seeling and Danicke, 2005). This toxin is found almost entirely in grains, in highly variable amounts ranging from a few ng/g up to μg/g concentrations. Deoxynivalenol (Fig. 15) is a mycotoxin associated with reduced performance and immune function in livestock (Seeling and Danicke, 2005). The kinetics, biotransformation and carry over of zearalenone and deoxynivalenol, as well as the effects of on ruminants, have recently been reviewed (Seeling and Danicke, 2005).

A number of toxinogenic fungal species, particularly producers of tremorgenic mycotoxins, have been isolated from traditional fermented meats. Tremorgenic mycotoxins are a group of fungal metabolites known to act on the central nervous system, causing sustained tremors, convulsions, and death in animals (Sabater-Vilar et al., 2003). Recent investigations indicate that many of these mycotoxins also exhibit a degree of genotoxicity (Sabater-Vilar et al., 2003). However, it is not known whether mycotoxin contamination of feedlot wastes may pose a risk to humans or the environment.

6. Heavy metals

The accumulation of heavy metals such as Cd, Pb and Hg in soils is a potential concern for Australian agriculture due to the capacity for these elements to adversely affect food quality, crop growth and environmental health (McLaughlin et al., 2000). Livestock manure is a possible source of low concentrations of some heavy metals. For example, Pb concentrations of 1.6–8.6 mg/kg and Cd concentrations of 0.1–0.7 have been reported in cattle manure in some countries (Bolan et al., 2004).

Some metals in livestock excreta may be derived from the animal diet, either intentionally or as a result of contamination. For example, some lighter metallic elements (such as As, Co, Cu, Mn, Se and Zn) may be added to livestock feeds as essential nutrients or to improve feed conversion efficiencies (Bolan et al., 2004). However, heavy metals are more likely to be derived from the ingestion of contaminated soil by the animal. For example, soil can be a significant source of Cd ingestion by Australian cattle grazing on fields amended with phosphate fertilisers containing high Cd concentrations (Loganathan et al., 1999). Dairy cattle manure has exhibited contamination with Pb and Ba via soil ingestion (McBride and Spiers, 2001). Practices such as using composted municipal waste as feedlot bedding also have the potential to contribute to the presence of heavy metals in contaminated manure (Zehnder et al., 2000). However, this is unlikely to be an issue for Australian beef feedlots since bedding is not used in outdoor feedlots and there are only a few indoor feedlots that generally use sawdust or rice hulls as bedding.

Unlike organic pollutants, metals persist indefinitely, changing only in their speciation and thus, mobility, partitioning and phytotoxicity (Nolan et al., 2003). In manure, most of the Cd is present in an organically complexed form, while other metals may be present more commonly in a ‘free’ state (Bolan et al., 2004). Biological processing (such as during anaerobic biogas production) does not significantly alter the concentrations of most metals in manure but may change its chemical form (Fares et al., 2005).

Soil properties such as organic matter content, mineralogy, salinity and pH can all have major effects on the mobility of specific heavy metals (McLaughlin et al., 2000). Sudden addition of cations such as Ca^{2+} to soils can result in cation-exchange processes, leading to significant leaching of some heavy metals from some soils (Voegelin et al., 2003).

In one study based on nutrient loading rates, it was estimated that land application of manures such as beef effluent, dairy slurry or composted cattle manure produced in England and Wales could result in typical field concentrations of approximately 0.002 kg/ha Cd and around 0.04 kg/ha Pb (Nicholson et al., 1999).

7. Dioxins

Dioxins are halogenated organic compounds derived from industrial processes. Many of this group of 210 chemicals are persistent and have become ubiquitous in the environment (Berry et al., 1993; Jones and Sewart, 1997; Vandenheuvel and Lucier, 1993). Dioxins include chlorinated dibenzo-p-dioxins (CDDs), chlorinated dibenzofurans (CDFs) and certain polychlorinated biphenyls (PCBs). The term ‘dioxin’ is commonly used to refer to the most studied and one of the most toxic

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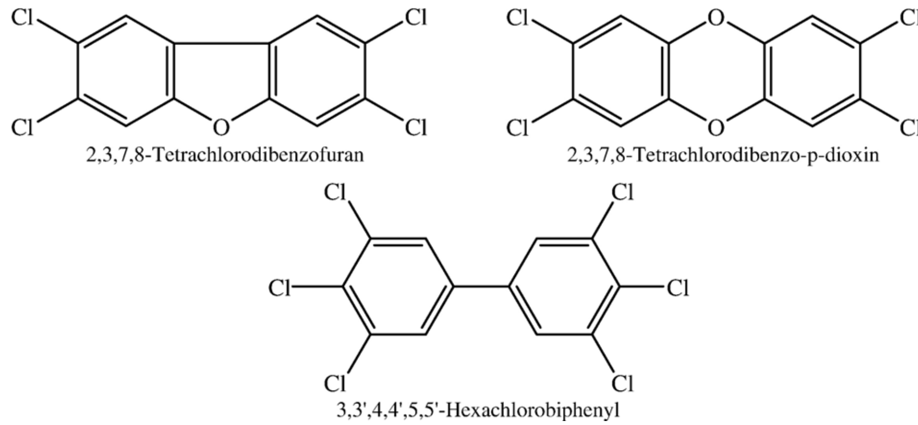

Fig. 16. Molecular structures of some dioxins.

dioxins, 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) (Schecter et al., 2006). The US EPA has assigned an oral cancer slope factor of 1.5×10^5 mg kg^{-1} day^{-1} for TCDD (US EPA, 2007a). Molecular structures of some important dioxins are presented in Fig. 16.

If animals are exposed, dioxins accumulate in fat tissue and this is among the most predominant exposure routes for most humans (Eduljee and Gair, 1996; Patandin et al., 1999; Schecter et al., 1997). Livestock are exposed to dioxins by the deposition of airborne dioxins onto plant and soil surfaces, and subsequent ingestion of this contaminated vegetation and soil (Fries, 1995b). Consumption of vegetation or roughages is considered the most important exposure factor (Fries, 1995a). Consumption of grass, hay, silage or grain can explain over 90% of dioxin concentrations in cattle (Lorber et al., 1994). Lot feeding is thought to significantly reduce dioxin concentrations in beef since the cattle are fed a predominantly grain based diet for several months before slaughter (Lorber et al., 1994). Grains contain lower concentrations of dioxins since the seed is not directly exposed to the air while growing. Livestock may also be exposed to dioxins if pentachlorophenol (PCP) treated wood is used on production facilities (Fries et al., 1996). Feedlot waste production is not known to increase nor concentrate dioxins in the environment.

8. Conclusions

While a large number of natural and synthetic chemicals are present in feedlot cattle manure and effluent, if any animal welfare, environmental or public health risks exist, they are expected to be associated with trace concentrations of some key biologically significant compounds.

Steroidal hormones (both natural and synthetic) have the potential to be significant environmental pollutants that may affect a diverse range of organisms. In particular, it has been shown that androgenic hormones such as testosterone and trenbolone are significantly active in the manure and effluent from beef feedlots in other countries. These chemicals are poorly understood in terms of fate and environmental implications.

Antibiotic agents are of interest primarily for the role that they may play in the dissemination of resistant bacterial organisms and genes. If poorly managed, manure and effluent containing residues of these chemicals may have implications for public health in Australia. Management practices involving virginiamycin, tylosin and oxytetracycline should be monitored.

Ectoparasiticides applied for the control of ticks, buffalo flies, lice and mites are toxic to a wide range of insect species and may be hazardous to important manure-borne insects such as dung beetles. Ectoparasiticides of greatest (potential) significance include synthetic pyrethroids, fluazuron, macrocyclic lactones and amitraz.

Mycotoxins, heavy metals and dioxins were also considered in this review. These include numerous toxic compounds, for which the potential for deleterious implications to animal welfare and public health is not understood. However, there is less available evidence to suggest that they ought to be prioritised for research in feedlot manure and effluent.

The vast majority of the available information has been obtained from international research and may not be representative of conditions in Australia. Very few of the individual chemical contaminants have been thoroughly investigated in Australian feedlot manure and effluent. The identification of best management practices for Australian feedlots will therefore require a comprehensive survey of the presence and fate of representative chemicals combined with investigations of their persistence and transport when manure and effluent are subjected to various management practices.

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