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Comparison of Anti-ectoparasite and Anti-endoparasite Therapies and Control Strategies

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Abstract

In this chapter, we consider the similarities and differences between management of ecto- and endoparasites. We discuss the general approaches of prevention and control of ecto- and endoparasites (historic and current chemotherapies, environmental management/host management), while considering the different challenges faced relating to lifecycle, host distribution, genetics, and selection pressure.

Introduction

The Merriam Webster dictionary defines a parasite as an organism living in, with, or on another organism. "Parasitism" refers to the intimate association between the parasite and host, whereby the parasite obtains part or all of its nutrition or needs from the host and results in an overall negative effect on the host. Simply, ectoparasites live on the outside of the animal and endoparasites on the inside. Microparasites (bacteria, viruses, protozoa) establish infections where it is hard to quantify numbers of infectious agents present, so numbers of infected hosts are quantified, rather than numbers of parasites within each host. Microparasites are small and have rapid generation times relative to their hosts. Macroparasites (nematodes, flies, ticks, etc.) are larger and can be counted; so the unit of study is the individual parasite, not the infected host. Macroparasites are also small and have rapid generation times, but there is less of a difference than between microparasites and host. Epiparasites are an interesting class of parasites whereby a parasite parasitizes a parasite in a host–parasite interaction referred to as hyperparasitism (as referred to in the well-known poem by Jonathan Swift: "a flea has smaller fleas that on him prey, And these have smaller still to bite 'em: And so proceed ad infinitum"). Examples of this are the larvae of the tapeworm,

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Dipylidium caninum, which infect fleas (*Ctenocephalides species*) and biting lice (*Trichodectes canis*). When a dog ingests a parasitized flea/louse when grooming, the tapeworm develops into an adult in the dog's intestine.

Fleas, ticks, and flies are the most visible and treated ectoparasites, but lice and mites also affect health and wellness. Infestation with ectoparasites causes many pathogenic effects, including tissue damage and blood loss due to feeding; hypersensitivity responses following exposure to ectoparasite antigens; secondary infections; and, most importantly disease transmission. Ectoparasites have evolved to fill many niches, but may be considered in terms of their host association. Many mites and lice live almost completely in permanent association with their host and, as such, have fairly low mobility and are open to risk of desiccation and death without the protection of their host. Other parasites, such as fleas, ticks, and flies, are more mobile and relatively resistant to damaging factors when off the host. As a result, the first category of organisms, mites and lice, often has a commensal relationship with the host as opposed to a parasitic interaction. The latter are able to find new hosts relatively easily, so are less impacted by death of a host and therefore likely to impose greater harm to the host. Most medically important ectoparasites have short generation times, large numbers of offspring, and very high rates of population growth [1].

Roundworms are the major infective internal parasite in both humans and animals, although cestodes (tapeworms) and trematodes (flukes) also have a significant impact on health. Helminth infections cause significant long-term, chronic debilitating disease and even death. In humans, it is estimated that around 125 000 deaths occur every year, and these are mainly due to infections with the hookworms, *Ancylostoma duodenale* and *Necator americanus*, or the roundworm, *Ascaris lumbricoides* [2]. In companion animals, endoparasite infections are primarily a disease of younger animals, with peak occurrence in dogs less than 6 months old and cats under 18 months old [3], with prevalence ranging from 5% to 70% worldwide [4]. Clinically, symptoms can vary from zero to critical (emaciation, anemia, death) and the zoonotic risks associated with some helminths are an additional concern. The economic impact of helminth infections on livestock, especially ruminant, production is well recognized [5, 6]; in pigs, it has been shown that the presence of endoparasites induces a reduction in body weight [7]. The mechanisms for the impact of helminths on production include direct tissue damage and diminished function of the affected organs; diversion of energy and protein resources of the host from production toward defense and immune mechanisms and reduced feed intake. In companion animals, there are similar adverse effects on health; unfortunately, roundworm infection is common, due to the ubiquity of infective stage larvae in the environment, and concerns are elevated due to zoonotic health risks.

Approaches for Ectoparasite and Endoparasite Control

Treatment of parasites results in removal of an existing infection, whereas prevention is a process by which infection is deterred. For dog and cat ectoparasite

infections, experts generally recommend prophylaxis (year round in some climates) over therapeutic treatment, to effectively manage control of the lifecycle, as well as to reduce the risk of disease transmission from ectoparasite vectors [8, 9]. The benefit from regular preventative treatment is particularly recognized for the control of fleas due to the nature of their lifecycle; an adult flea infestation is only a very small part of the population, which includes immature stages present in the pet's environment. It is critical to control these stages, either by the use of products that target these early lifecycle stages or by regular use of products that eliminate adult fleas on the animal, which will progressively lead to the reduction of environmental lifecycle stages. CAPC (Companion Animal Parasite Council) goes as far as to recommend "avoiding initial infestation altogether by placing pets on life-long prevention programs is the best option for pets and their owners" [8]. Transmission of diseases (i.e., *Rickettsia rickettsia* and *Borrelia burgdorferi*) by vectors, especially ticks, in dogs and cats is a major concern, and reducing the ability of a vector to attach and/or feed with an effective ectoparasite control program will reduce the risk of disease transmission. Tick-borne diseases in dogs and cats are becoming increasingly important, with several tick species responsible for the continued spread of multiple diseases. Among the other more important diseases are babesiosis, hepatazoonosis, Ehrlichiosis, anaplasmosis, cytauxzoonosis (cats), and tick paralysis. Although control of internal parasites is the primary concern for horses, ectoparasites can also impact the welfare of horses, either through dermatological effects or nuisance bites, which affect the ability of horses to thrive. The primary ectoparasites of horses are houseflies, stable flies, mosquitoes, and horse and deer flies; ticks, lice, and mites also parasitize horses. The major problem is a limited supply of effective, licensed products for horses [10], combined with the challenges of managing ectoparasite species that are able to live for extensive periods off the animal, requiring frequent treatment. Fly repellents tend to have a very short duration of efficacy, if any, and need frequent reapplication. Taylor's 2001 review [11] highlighted how few pharmaceutical agents are available for treating horse ectoparasites and this situation has improved little in the intervening years.

For livestock, as for companion animals, ectoparasite control is dependent on the parasite lifecycle – do they spend their whole life on the host, like lice; or only spend time on the animal to feed, as for some species of mites, which then return to protected spaces in the environment? For the former, treating just the animal will suffice; for the latter, the environment must also be treated. In a 1992 review [12], Byford *et al.* gave an authoritative overview of the commercial and health impact of ectoparasite infestation in the United States, focusing on the horn fly, *Haematobia irritans*, commercially the most important and widespread pest in cattle in the southern United States. Although a complicated condition, the overall implication was that the damaging effect on production and performance of cattle results from an alteration of the total energy balance following ectoparasite infestation. This is a major problem, considering the widespread resistance of horn flies to pyrethroids, probably accelerated by the use of pyrethroid-impregnated ear tags [13].

Humans are as susceptible to ectoparasite infestation as animals are, and are often affected by the same pests; for example, close contact with pets can result in infestation with fleas, ticks, lice, and mites and, although more common in animals, humans can also suffer from myiasis, especially in tropical regions. Scabies and head lice [14], as well as being socially embarrassing, can cause significant health problems. Resistance is a major issue, with multiple resistance mechanisms identified in different populations of head lice, including kdr (knockdown resistance) mutations of the sodium channel and oxidative metabolism resistance mechanisms (see chapter 6 by J. M. Clark in this volume). Although head lice are the most prevalent parasites causing pediculosis, body louse prevalence is also increasing, which heightens the public health threat due to risk of transmission of a number of diseases, including typhus (*Rickettsia prowazekii*), louse-borne relapsing fever (*B. recurrentis*), and quintana (trench) fever (*Bartonella quintana*). Tungiasis occurs in tropical and subtropical regions and is caused by the tiny flea, *Tunga penetrans*, the chigoe flea or jigger, which embeds itself under the stratum corneum and can lead to dangerous complications from secondary infections.

However, the biggest impact on human health globally is from ectoparasite vectors. Malaria, caused by the protozoan parasite *Plasmodia* spp.*,* is commonly transmitted by infected female *Anopheles* spp. mosquitoes and, in 2015, there were approximately 214 million malaria cases and an estimated 438 000 malaria deaths [15]. Ticks are becoming increasingly important as a cause of significant disease in humans, as well as their pets. Examples of disease common to both pets and humans include the bacterial Lyme disease (*B. burgdorferi*), transmitted by the deer tick, *Ixodes scapularis* (*Ixodes ricinus* in the European Union); Rocky Mountain spotted fever (*R. rickettsia*), transmitted by *Dermacentor variabilis*; and ehrlichiosis (*Ehrlichia chaffeensis*), transmitted by the lone star tick, *Amblyomma americanum* and *I. scapularis.* The protozoal disease babesiosis is caused by infection with *Babesia microti* or *Babesia equi*, transmitted by *I. scapularis* and *Ixodes pacificus.* Viral diseases can also be transmitted by ticks, for example, tick-borne encephalitis (TBE) (caused by the flavivirus, TBE virus), transmitted by *Ixodes* spp. and there are even toxins, such as the tick paralysis toxin transmitted by *Dermacentor* spp. in the United States and *Ixodes holocyclus* in Australia. Ticks and mosquitoes may cause significant disease, but fleas have also had a major effect on human history. The vector for bubonic plague, *Xenopsylla cheopis*, transmits the bacterium *Yersinia pestis* when it feeds and this was thought to be the cause of the Black Death, which killed an estimated 50 million people in the fourteenth century [16].

For helminth infections, prevention is managed by disrupting the lifecycle of the parasite, which, in humans, is usually achievable by good sanitation and hygiene; but in animals, this is often less feasible. For livestock, experts recommend combining anthelmintic control with minimizing exposure to reinfection; while in companion animals with exposure to the external environment. Where contamination of the environment with infective larvae is extensive, prevention usually requires a strict treatment regimen, combined with regular egg production

monitoring. A unique situation exists with heartworm, where a very high degree (up to 100%) of efficacy is required to control this potentially life-threatening disease of dogs and cats. Fortunately, regular dosing (1-month and 6-month products) with a macrocyclic lactone (ML)-based anthelmintic prevents development of the larval-stage heartworms. Heartworm larvae are very sensitive to ML products and until recently efficacy was thought to be 100% for the various products. However, more recent evidence of heartworm resistance to MLs has been detected in some areas of the United States (Mississippi Delta) and is a cause for concern. The American Heartworm Society [17] generates guidelines for canine and feline heartworm prevention, which it updates regularly based on the latest scientific understanding of the disease; the most recent revision was in 2014. For horses, as mentioned earlier, internal parasites are a major concern, especially as few new drugs are being approved for horses. In the face of increasing anthelmintic resistance [18], more sustainable methods for helminth control are being sought.

Ectoparasiticides

There are many mechanisms of action utilized in the management of ectoparasites in animals and humans, most older ectoparasiticides being historically leveraged from the crop protection industry. Numerous agricultural pests and veterinary ectoparasites are insects and acarines; and agrochemicals with activity against crop pests also frequently work against animal health ectoparasites. Add to this the fact that the market for Animal Health ectoparasiticides is significantly smaller than the market for agricultural pesticides, and it makes commercial sense to leverage the learnings and assets for animal health utility. Ivermectin is a major exception, being discovered by a pharmaceutical company animal health group (Merck Sharp & Dohme), and was first used on animals and later for agriculture and human medicine.

A primary driver for the development of these multiple therapies is the development of resistance. Resistance is a shift in susceptibility to a drug [19] and is recognized as a failure of drugs to control parasitism. Resistance is often measured as survival of parasites following a treatment that would be expected to be effective, or as a reduction in the protection period that a persistent treatment provides. Resistance development is multifactorial and involves parasite genetic factors (dominance of resistance alleles, gene frequency, fitness of resistant parasites, linkage disequilibrium, etc.); the host–parasite interaction (immunogenicity, pathogenicity, levels of refugia, etc.); biological factors (breeding patterns, numbers of offspring, generation time, behaviors that impact gene flow and opportunities for selection – migration, refugia, host range, etc.); and the parasite management system (method of application, frequency and timing of treatments, life cycle stage treated, selection threshold, etc.).

Insecticide resistance was first documented in 1908 by Melander [20] who noticed significant levels of survival of the San Jose scale insect, *Quadraspidiotus perniciosus* (Comstock), after exposure to lime-sulfur. In a 1984 review, Forgash [21] described the emergence of 428 resistant insect and acarine species in the

following years, with 61% having medical/veterinary importance, and numbers still growing. By 2014, the cumulative increase in species resistant to insecticides was 586 [22]. Significantly, the numbers of resistant species started to increase dramatically after the introduction of synthetic organic insecticides (i.e., DDT (dichlorodiphenyltrichloroethane), cyclodienes, and organophosphates) in the 1940s. These products had better efficacy and broader spectrum of activity and consequently were used more extensively and repetitively, a practice that likely resulted in the observed resistance.

The Insecticide Resistance Action Committee (IRAC) was set up in 1984 to provide a coordinated response from industry to delay resistance development in insect and mite pests [23]. Its primary objective is to ensure long-term efficacy of insecticides and acaricides, thereby enabling sustainable agriculture and improved public health. One of the tools used is a mode of action classification scheme [22], which classifies pesticides based on the target site of action or mode of action. This can then be utilized, along with guidance on resistance management, to support alternation or rotation-based resistance management programs. The current classification includes 25 different mechanisms of action. Although focused on the crop protection industry, these classifications are also valid for effective management of human and animal health insect and acarine infestations.

Methods of ectoparasiticide use vary depending on the parasite and the host. For animal health, convenience is a major driver of route of administration [24]. Treating livestock is a very costly and resource-intensive process, so farmers have traditionally sought methods that allow whole-herd administration, such as foggers, dusts, sprays, dips, and so on, primarily incorporating formulations of organophosphates and synthetic pyrethroids to control ticks, mites, lice, and blowflies. Dosing frequency is highly dependent on the persistence of ectoparasiticide on the skin, hair, or wool of the animal, not just the lifecycle of the parasite; duration of efficacy is therefore generally longer in sheep, as the persistence on wool is higher. Cattle ear tags, primarily formulated with pyrethroids and some organophosphates, are still used for management of biting flies; when attached to the ear, insecticide is released from the formulation and dissolves in the sebum, spreading over the whole body, likely by grooming, ear/tail flapping, and contact between animals. In the past 30–40 years, agents and formulations with systemic efficacy have been developed and have enabled easy pour-on delivery (i.e., avermectins/milbemycins, synthetic pyrethroids, and some organophosphates); and even parenteral delivery for control of some ectoparasites, primarily endectocides (i.e., avermectins/milbemycins).

Companion animal ectoparasiticide products have progressed significantly in the past 30 years. Historically, dusting powders, baths, and aerosol sprays and impregnated insecticidal collars, with organophosphates, carbamates, and synthetic pyrethroids as the active agents, were the only available control measures for fleas, ticks, mites, and lice. Efficacy was variable and often with short duration and there were higher risks associated with toxicity both for the owner applying the product and for the animal, than with products developed in recent years. Spot-on application increased in popularity in the 1990s, with formulations incorporating

the nicotinergic flea agent, imidacloprid, the GABA-gated chloride channel antagonist for flea and tick control, fipronil, and the chloride channel agonist, selamectin, for control of endo- and ectoparasites. More recently, oral ectoparasiticides such as spinosad [25, 26], a putative nicotinic acetylcholine receptor agonist; and the GABA-gated chloride channel blocking isoxazolines [27–29] have become available on the market (see chapter 15, by Woods and McTier, in this volume). The latter are potent insecticidal and acaricidal molecules which have provided safe, oral chewable tablets for the treatment and control of fleas and ticks in dogs. Interestingly, insecticidal collars, such as Scalibor® (Merck) and Seresto® (Bayer) that provide both repellency and direct killing of parasites have made significant sales in recent years, due partly to an increased concern about the spread of *Leishmania* into northern Europe [30], as well as convenient prevention of flea and tick infestations. Domestic dogs are the primary reservoirs for human visceral leishmaniasis, caused by the zoonotic protozoa *Leishmania infantum*. Control of the sand fly vectors, *Phlebotomine* spp*.,* is the primary approach to managing disease transmission and collars impregnated with pyrethroids, such as Merck's Scalibor, are able to deliver an extended duration of prevention.

As outlined, a key motivator for managing ectoparasite infestation in humans is reduction of the risk of vector-borne disease transmission [31], although we should not underestimate the potential for significant morbidity from other ectoparasites in susceptible populations [32]. Treatments are generally topical [33]. For head lice treatment, pyrethroids are the main pediculicides [14]; scabies is treated with topical scabicides (pyrethroids, lindane, malathion, crotamiton, benzyl benzoate) and off-label oral ivermectin [34]; in tungiasis, the flea (*T. penetrans*) is removed physically. The incidence of flea- and tick-borne diseases is thought to be greater than is recognized by doctors and health authorities, and hence diagnosis and treatment are often delayed as they are not initially considered when attempting to determine the cause of the illness [35]. Control of fleas on pets and in the environment is the best approach for preventing disease transmission, as discussed earlier. Similarly, preventing exposure to ticks is recommended to prevent transmission of diseases such as Lyme disease; for example, with the use of an insect repellent, either DEET (*N*,*N*-diethyl-*meta*-toluamide) or a pyrethroid spray. In areas where TBE is prevalent (central and eastern Europe and northern Asia), the World Health Organization (WHO) recommends immunization with the TBE vaccine, which has been shown to be highly effective [36], and is on the WHO list of essential medicines [37]. There is increasing interest in utilizing oral ivermectin for control of ectoparasites, especially in poor countries where populations are infected with multiple parasites and ivermectin is already used in antifilarial control programs, although spectrum gaps have been identified [38], so this is not a universal solution.

Endoparasiticides

There are fewer classes of endoparasiticides than ectoparasiticides due in part to a reduced emphasis on the discovery of endoparasiticide agents by the agrochemical industry, although there are still examples of nematicidal molecules being

discovered by crop protection companies and leveraged for development as animal health anthelmintics; emodepside being one example. In 1990, Meiji Seika Kaisha patented PF1022A, a novel cyclooctadepsipeptide anthelmintic (European patent 0382173A2) [39]. Fujisawa Pharmaceutical Co. (Japan) then filed another patent which included the bis-*para*-morphonyl derivative of PF1022A, named emodepside; which was licensed by Bayer Animal Health and developed as an anthelmintic for dogs and cats; marketed as Profender®, in combination with praziquantel [40].

Similar to the situation with ectoparasiticides, resistance is a strong driver for identification of novel endoparasiticides. However, for the major commercially important host species (cattle and dogs) nematodes have been slow to develop resistance to the endectocidal avermectins and milbemycins, so investment in novel endoparasitic drug classes has been limited. As a result, only three new drug classes have been marketed in the past 30 years [41]. Endoparasiticide resistance is, however, now being reported in cattle gastrointestinal nematodes [42–44] and heartworm (*Dirofilaria immitis*) resistance to MLs (avermectins and milbemycins) in dogs is now acknowledged to have emerged in the Mississippi Delta in the United States [45, 46]. This has no doubt stimulated investment in Animal Health endoparasiticide research. For example, more than 800 anthelmintic families were filed in animal health company patents during the past 10 years; at least 175 of these describing novel compounds. Along with increased investment in anthelmintic discovery for human filarial diseases, this is very encouraging for future management of endoparasitic diseases in animals and humans.

In animal health, anthelmintics are used therapeutically to treat existing infections or clinical outbreaks or prophylactically where treatment timing is dependent on the disease epidemiology. When viewed across all hosts, anthelmintics are primarily administered orally: as drenches in livestock, tablets for dogs and cats, and pastes for horses; but parenteral dosing, by injection or with pour-on formulations, is also widely used in cattle (and to a limited degree in dogs), to reduce time and resources needed to treat the animals. Due to increasing resistance, which is widespread and serious in sheep, multiple drug classes are used, both alone and in combination (in some cases, with multiple active agents); with the newer aminoacetonitrile derivative and spiroindole drugs being utilized increasingly in sheep to control infections in areas where all other drug classes no longer work. There is a strong advocacy among experts for more sustainable approaches to resistance management [47], and this will be discussed in more detail in the section titled "Endoparasite Challenges" Section 2.2.

Challenges for Ecto- and Endoparasite Control

Ectoparasite Challenges

Unfortunately, selective breeding for "improved" traits in livestock and companion animals has generally increased susceptibility to parasites; for example, some breeds of dog (Dalmation, American Bulldog, and American Pit Bull Terrier) appear to be more susceptible to *Demodex canis* [48]. This is exacerbated by

intensive production practices for livestock and the increasing zoonosis concerns with the growth of pets being viewed as family members; in a US 2015 Harris Poll [49], 95% of pet owners considered their pets to be members of the family (up 7% since 2007).

With the inherent variability in lifecycles, climate, and hosts, it is difficult to make broad recommendations on management of ectoparasite infestations. It can be challenging to control parasites, such as ticks and flies, that only spend part of their lifecycle on the host. Some parasite infections are seasonal (e.g., tick infections are common in spring and autumn and louse/mite infections more common in autumn and winter), allowing seasonally targeted treatments. With changes in climate, there are increasing predictions and observations of the spread of diseases from warmer to previously more temperate climates, due to movement of the vectors, as for sand flies in Europe [30]. There is also no doubt that increased international trade and travel is leading to reemergence of ectoparasite diseases; epidemiological studies support that ectoparasite diseases and their vectors are hyperendemic in the developing world [50].

At what point is intervention optimal? This is a surprisingly difficult question to answer. Logically, it makes sense to intervene before welfare is impacted, but this is not always well understood and can sometimes be difficult to measure. For example, with hypersensitivity, as in flea allergy dermatitis [51], once sensitization has occurred, recurrence of signs can be initiated by just a small number of bites, although the threshold of sensitivity varies between individual dogs [52], so preventing flea infestation with monthly treatments, either topical or oral, is recommended to break the lifecycle. There are established guidelines for companion animal parasite management [8, 9, 17], which include guidance for ectoparasites. As highlighted in the section titled "Ectoparasite Challenges", experts generally recommend prophylaxis over therapeutic treatment, to effectively manage control of the lifecycle, and to reduce the risk of vector disease transmission. For livestock, it is a continuous battle to maintain the efficacy of parasiticides [53]. There is a dichotomy between the desire of farmers for easy application and fewer interventions, and the prevention of resistance by minimizing selection pressure and maintaining refugia, thereby ensuring the population is constantly refreshed with unexposed, susceptible parasites [54]. It is no coincidence that resistance developed more rapidly in the single host tick, *Rhipicephalus* (*Boophilus*) *microplus*, than in multihost ticks [55]. Understanding population dynamics is a valuable tool, but can be challenging. Experts advocate the development of more sustainable, integrated pest management programs [1, 56], incorporating strategic, directed treatments, environmental control, disease management, and resistant breeds. However, this would require significant changes in management practices in the industry.

In humans, head lice infestations are a significant issue in developed, as well as developing countries [57–59], with evidence that prevalence is increasing around the world [60]. It is therefore surprising that monitoring and reporting are not standard practice in many countries [60]. It is clear that epidemics spring up frequently in populations of children, where, if left untreated, the infestations

spread rapidly. Unfortunately, resistance to topical ectoparasiticides is widespread [61], with many plant-based products now being used, although safety and efficacy have not been well established. A key recognition from mathematical modeling is that synchronized treatment of "potentially" infected individuals (relatives, classmates, and other close contacts) should interrupt transmission [62], with systematic treatments being another key to successful eradication of infections.

Vaccines have long been championed as the solution to parasiticide resistance; however, despite decades of investment and research into host–parasite interactions and evaluation of many putative vaccine antigens, the number of marketed ectoparasite and helminth parasite vaccines is disappointingly limited [63]. Ectoparasite vaccines are particularly challenging, as the parasites live either on the surface of the host or even off the host. Bm86 is the only ectoparasite recombinant vaccine and works by immunizing cattle with a "hidden" tick gut antigen; antibodies generated against Bm86 rupture the gut wall of the tick and give good levels of protection against tick infection, although repeated immunizations are required to maintain antibody levels [64]. The lower levels of efficacy when compared to drug treatment, requiring parallel drug treatment, led to poor sales and removal from the market. Interestingly, a new formulation, developed by the U.S. Department of Agriculture (USDA) Agricultural Research Services (ARS), USDA Veterinary Services, and Zoetis, was recently given a conditional license for management of *R. microplus* infestations in both permanent and temporary quarantine zones in Texas [65].

Environmental concerns have led in the past to removal of pesticides from the market. Everyone is familiar with DDT, once considered the solution to all our pest problems. Its use to control ectoparasite vectors undoubtedly saved many lives before tolerance/resistance started to emerge. Unfortunately, it is also now known to be a persistent organic pollutant, which is readily adsorbed to soil/sediment and is resistant to environmental degradation; and add to this a high lipophilicity, which leads to bioaccumulation in the food chain and impacts on wildlife. There was a widespread ban implemented in most countries between 1970 and 1990, although a limited supply of DDT is still used for vector control, by indoor residual spraying (spraying the inside walls of homes made of mud or wood) [66]. Environmental impact now has to be evaluated for every new antiparasitic drug; ectoparasiticide, anthelmintic, or endectocide. This is generally not an issue for the newer oral companion animal ectoparasiticides, where environmental exposure is very limited, but can require a considerable program of work for livestock ectoparasiticides. Some ectoparasites, such as red mites on chickens, spend the majority of their lifecycle in crevices in buildings, only leaving to feed on the host for a short period at night. In this case, spraying the buildings with pesticide is the most effective control method; however, the products are considered biocides and require an extensive environmental program for approval.

Endoparasite Challenges
In this section we focus on the management of helminth parasites in livestock where drug resistance is a major challenge for both the control and prevention of endoparasite infections.

Both internal and external parasites of grazing livestock are ubiquitous and therefore all grazing livestock should be considered an at-risk or an exposed population to infection and infestation [67]. Even light-to-moderate infections and infestations negatively impact the welfare, thrift, and production efficiency of grazing animals. Approaches to minimize parasite infections and infestations include husbandry practices founded on an understanding of the epidemiology of the organism, chemotherapeutic interventions to prevent or remove the effect of the organism on the host, or a combination of both. Prior to the 1960s the use of chemotherapeutic agents to achieve modern-day expectations for animal welfare and production efficiency was virtually nonexistent. Livestock producers were essentially dependent on the genetic resistance or tolerance of the host to sustain body growth and reproduction, often at a high cost to the welfare and production efficiency of the animal. Furthermore, little was known about the epidemiology and biology of internal and external parasites, which could assist management decisions to moderate parasite infections. Even with the current knowledge base of parasite epidemiology and host genetics and breeding techniques, improved husbandry and hygiene practices and genetic selection as stand-alone methods for parasite control are far from achieving the level of animal welfare and production efficiency expected from modern livestock producers and society [68].

Since the 1960s, global beef production has more than doubled and carcass weights have increased by approximately 30% [69]. Improvements in animal welfare gained from effective parasite control by the three main classes of anthelmintics (benzimidazole, imidazothiazole/tetrahydropyrimidines, and MLs) have contributed to the efficiency of livestock production. The endectocidal characteristic of the MLs has also enabled livestock producers to depart from the once common use of plunge dipping or bath treatments for external parasite control; now limited primarily to tropical and subtropical regions of the world. Livestock managed under effective internal and external parasite control programs founded on chemotherapeutic control are more efficient converters of feedstuff to meat, enabling more efficient utilization of land and feed resources.

Inherent with the administration of any anthelmintic is the genetic selection of the subpopulation of organisms that are genetically tolerant or resistant to the active ingredient. Anthelmintic resistance, at least to the major classes of compounds, is conferred by multiple alleles and therefore constitutes a small percentage of a naive parasite population. As selection pressure is increased on a parasite population, the proportion of resistant parasites increases until they are the dominant genotype in the parasite population. In addition to the frequency of exposure/selection pressure, underdosing (exposing parasites to subtherapeutic levels of a drug) will also increase the resistant population by further selecting parasites that are genetically tolerant to the active pharmaceutical ingredient (API). This phenomenon has been observed for all classes of anthelmintics and will likely be the case if other classes of anthelmintic compounds become commercially available [42].

The greatest prevalence of anthelmintic resistance in livestock has been observed in the sheep industry where frequent anthelmintic administrations were common

practice in internal parasite control programs. The frequency of administration was driven primarily by high mortality and clinical morbidity associated with parasitic gastroenteritis and anemia due in large part to the hematophagous parasite *Haemonchus contortus. H. contortus* is often the most prevalent internal parasite in sheep due to its high fecundity (egg shedding) relative to other species. The practice of frequent anthelmintic application and resistance development in sheep has often been inaccurately extrapolated to characterize the use pattern of anthelmintics and endectocides in cattle parasite management programs. This is an unfortunate and mistaken characterization of the industry as a whole. There are likely cases of misuse, overuse, or erroneous application of anthelmintics; but without empirical evidence demonstrating common practice, such statements are mere generalizations. It would not be possible to have 30 years of effective use of these compounds in the cattle industry if frequent indiscriminate or misuse was common practice in the industry. A brief review of the scientific literature will demonstrate that anthelmintic resistance is now, however, emerging in all species of livestock that are exposed to the current classes of compounds [42, 67, 70]. Industry and scientific leaders in conjunction with veterinarians and producers are working toward solutions to maintain the longevity of existing anthelmintics. For example, there is a slow shift in some market segments away from pour-on formulations to injectable formulations to ensure proper dose rate and application and the goal is to continue this trend. The introduction and proper use of combination products or concurrent use of anthelmintics with disparate modes of action [71] are also being introduced to producers along with education on use patterns.

Targeted selective treatments have long been advocated as a refugia-based approach to resistance management [47]. A number of biomarkers have been proposed, including Famacha® for haemonchosis [72] and measures of health and/or performance [73–75]. However, even the advocates recognize that it can be a challenge to convince farmers of the value of these approaches, when weighed against the additional time, energy, and costs required to implement [76, 77].

The availability of all of the existing classes of anthelmintics is vital to maintaining the current level of health and welfare of livestock in modern production systems, with a need to introduce new classes to support and maintain these levels, alongside helminth management programs that include anthelmintic resistance management as a variable.

Perspectives on Current and Future Strategies for Ecto- and Endoparasite Control

It is evident that antiparasitic agents greatly enhance the welfare and subsequently, for livestock, the production efficiency of the host. Unfortunately, these advantages diminish over time if parasiticide-susceptible parasite populations are not maintained within the environment where the host–parasite interaction occurs. When consistent genetic selection pressure by an antiparasitic agent is maintained on a parasite population, a threshold is reached where the parasite population is no longer susceptible to the antiparasitic agent and its benefits are

no longer observed. Such is the case for small ruminants and horses, and the situation is now emerging in cattle. Researchers in more recent years have been evaluating mechanisms to manage resistance development and keep products effective for longer; but there is a mismatch between the reality of dosing regimens for animals and for humans in the developing world and the reality of what is required for "best practice" for management of resistance development.

Challenges of Bringing New Antiparasitic Drugs to the Market

In earlier sections we highlighted the importance of agrochemical pesticide development for leveraging substrate for animal health application, particularly for ectoparasiticide drugs, with the isoxazoline class highlighting the value of this resource; as well as the importance of animal health drug development as a source of human health antiparasitic medicines. We have previously shown figures highlighting the consolidation of the animal health industry over time [78, 79]. As a consequence of acquisitions and mergers, the overall resources available for antiparasitic discovery have reduced considerably over the past 25 years. We have updated the figure for this chapter (Figure 1.1) to show that the recent acquisition of Novartis by Elanco, and the acquisition of Merial by Boehringer Ingelheim Vetmedica have only intensified this consolidation. Although antiparasitic drugs are core to the success of animal health companies, these changes have resulted in fewer players, reduced competition, and potentially less opportunity for the discovery of novel antiparasitic molecules. A reduction in resources available for research and development (R&D) will impact availability of scientists and funds to discover and develop new products.
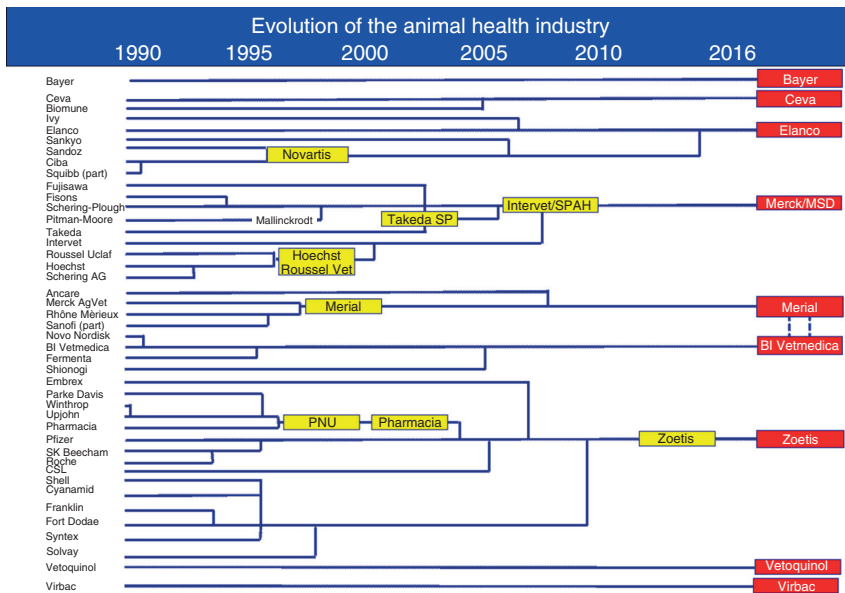

Figure 1.1 Consolidation of animal health companies 1990–2016.

As discussed in previous reviews [41, 78, 79], identifying molecules that kill parasites *in vitro* is the easiest part of the R&D process. A major challenge is delivering the drug with the optimal pharmacokinetic profile for efficacy, via the preferred, convenient route of administration. The drug has to be safe, both to the animal and the handler; with additional regulatory hurdles for human food safety (and environmental safety) for livestock products. Added to this are increasing regulatory pressures on new and even on existing products. For example, European Medicines Agency (EMEA) has concerns about persistent, bioaccumulative and toxic (PBT) or very persistent and very bioaccumulative (vPvB) substances in veterinary medicinal products [80]. Finally, the product has to be manufactured to high levels of quality, both for the API and the formulated product. All of these challenges increase the time and resources required to bring a product to the market. Companies then have to balance the chance of success and R&D costs against the potential value and return on investment (ROI) long term. If this is low, or negative, the product will not be developed. For human health the business model is different, with nongovernmental (nonprofit) organizations (NGOs) increasingly investing in neglected parasite diseases, including helminths. Although they still face all the challenges of the animal health industry, there is no expectation of ROI, but still a proven route to success is partnering with animal health companies. Historically, there are good examples of pharmaceutical companies partnering with the WHO to develop products for human health use. Mectizan® is a great example in which Merck led a collaboration with WHO in the late 1980s, running field studies to demonstrate the efficacy and safety of ivermectin for the treatment of onchocerciasis (river blindness). They have continued to commit to the Mectizan Donation Program [81] to provide ivermectin to treat both onchocerciasis and lymphatic filariasis (elephantiasis) in Africa. DNDi (Drugs for Neglected Diseases Initiative) has also recently partnered with Bayer Animal Health to evaluate the anthelmintic emodepside for macrofilaricidal activity against onchocerciasis [82].

Prevention and Control – Balancing the "Ideal" Against the "Real World"

In an ideal world, we would manage parasite infections and infestations with good hygiene and husbandry but, with the realities of the conditions of the developing world for humans and the intensive farming required to feed the world's population, this is never going to be possible, even for infections that could be managed this way. Parasiticides are therefore a fact of life, and it is to everyone's advantage to extend the lifespan of existing and new drugs. There is a clash, however, between treatment regimens that minimize resistance development and the desire for convenience in dosing and duration of efficacy, both for humans in the developing world, where access to treatments may be limited and challenging to reach, and for animals where reducing handling is a major driver. Farmers and pet owners now expect treatments and preventatives that meet their needs, be they chewable monthly flea and tick products for dogs (Simparica®,[1] NexGard®, and Bravecto®) or long-acting products (heartworm preventatives for dogs (ProHeart® 6 and 12) and anthelmintics for cattle (LongRange®)). Indications are that the

market is progressing even further down the path of convenience, to meet the demands of their customers. So, how do we balance the demands of the customer with responsible parasite management?

Integrated parasite management (IPM) has been championed for many years as a responsible tactic to minimize the impact of parasite resistance [83, 84]. IPM aims to improve host resistance by combining multiple approaches, both chemical and nonchemical (targeted use of parasiticides, improved monitoring of resistance and infection levels, and incorporation of nonchemical control methods, e.g., fungi). There are examples of initiatives to utilize agroecological approaches in developing countries [85]. However, there is reluctance among end users in the developed world to accept the increased costs and resources to implement such schemes and the reality that there will likely be some loss in production and a level of parasitism present.

One approach being used successfully in sheep for helminth control is development of combination products. Historically there have been concerns about combinations increasing parasite selection, but modeling has shown that combining a new drug (with low resistance frequency and very high efficacy) with another class of anthelmintic will delay development of resistance to the new drug [71, 86]. The modeling showed that resistance to even a new active drug can develop rapidly if it is used in an inappropriate manner. However, although the benefit of the anthelmintic in slowing resistance development to the new entity is influenced by the level of resistance to the older drug, Leathwick's model [71] predicted that even at 50% efficacy of the older drug, the development of resistance to the novel drug should be slowed in a combination and vice versa. Nonetheless, best practice would be to use the combination while the older drug still has relatively high efficacy and resistance genes are still infrequent. The model also illustrated that a large percentage of the population must remain unexposed to the treatment – as refugia decreased, resistance developed more rapidly, reducing the benefit of the combination; encouraging management strategies such as rotational grazing. Another important observation was that resistance was still delayed even when resistance to one of the drugs was functionally dominant, as long as a high level of refugia was maintained. This is likely to be due to fully overlapping generations and small proportions of populations exposed to each treatment.

For insecticides too, modeling shows that mixtures are effective at delaying resistance (even better than alternation), as long as a proportion of the population is not exposed to the treatment [87]. In order for these mixtures to be effective for delaying resistance, the initial resistance frequencies should be low, the agents should be close to 100% effective against treated susceptible homozygotes, and the combination components should be nearly equal in persistence.

There have been some efforts to identify and validate nutritional supplements for sustainable control of gastrointestinal nematodes in livestock [77]; both targeting direct anthelmintic effects and the indirect effect of supplementary feeding improving an animal's resilience against gut nematode infections (nutraceuticals).

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[1] SIMPARICA is a trademark or registered trademark of Zoetis Services LLC in the United States and other countries.

Tannin-rich plants (TRP), for example, may have a direct effect on reduction of larvae establishment in the host, as well as benefit for the host from the nutrients in the fodder [88]. This approach however depends on supporting the level of resilience and resistance against gastrointestinal nematode infections, which varies among ruminant species and also among and within breeds.

Biological control is well established for control of agricultural pests, where a range of control methods are used, including introduction of pathogens (bacteria, fungi, viruses, etc.), predators (insect larvae (ladybugs), entomopathogenic nematodes, predatory mites), and parasitoids (wasps and flies). More recently, RNA interference (RNAi) is also being evaluated. Transgenic plants offer the opportunity to express pathogens/toxins and this has been incredibly successful, especially with the use of *Bacillus thuringiensis* (Bt) toxin, which has replaced chemical insecticide use for many crops. This strategy is now being applied to RNAi, the latest tool for pest management [89, 90], with an RNAi-enhanced corn engineered to contain RNA devised to kill rootworms in development by Monsanto. RNAi sprays are also being developed, and could be on the market by 2020 [89]. However, there are concerns about effects on biodiversity and a need to evaluate potential levels of risk posed to nontarget species by biological control strategies [91]. Although there are studies evaluating fungi for veterinary control of livestock gastrointestinal nematodes [92–94], biological control strategies have yet to make an impact on the management of veterinary parasites.

The reality for veterinary and human parasite control is that there will continue to be an expectation for ectoparasiticides that rapidly clear existing infestations and prevent reinfestation for extended periods of 1 month and longer. For helminth control in livestock, the aim is to keep the challenge to young livestock at a minimum rate by both periodic and strategic deworming. For companion animals, the recommendation is year-round broad-spectrum parasite control with efficacy against roundworms, hookworms, and whipworms [8].

It is clear that parasites, both internal and external, have a major impact on the health and well-being of humans, both directly and through their effect on companion animals (with associated zoonotic diseases) and livestock (influencing the efficiency of food production). Building our understanding of the biology of the responsible organisms will help in the development of new drugs, vaccines, and control strategies.

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