Veterinary Pharmacology in Animal Health Discovery: Structure and Activity of Avermectins and Milbemycins in Animal Health

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Merck & Co. is one of the few pharmaceutical companies that has a full basic discovery research group concentrating on animal health. This group is a part of Merck Research Laboratories, which is a division separate from the marketing and technical services divison of Merck AgVet. Basic research has been instrumental in the success of animal health products at Merck and was responsible for the discovery of ivermectin and the associated avermectins. This paper focuses on veterinary pharmacology and discovery research related to avermectins and milbemycins for animal health.

The avermectins and milbemycins are closely related 16-membered macrocyclic lactones (Burg *et al.*, 1979; Takiguchi *et al.*, 1980). Both chemical groups are produced through fermentation by soil dwelling actinomycetes from the genus *Streptomyces* and have similar biological activities. The most important structural difference between the two groups is a disaccharide, bisoleandrosyloxy, substituent found at the 13-position of the macrolide ring of the avermectins. The milbemycins have no substituent at the 13-position. Thus, one can think of the avermectins as glycosylated milbemycins, or of the milbemycins as deglycosylated avermectins.

Although the milbemycins were discovered in 1973 as acaricidal and insecticidal compounds for crop protection by Sankyo scientists, the potential of this group was not realized until the discovery of the acaricidal, insecticidal, and nematocidal activities of the avermectins by Merck scientists in 1975 (Egerton *et al.*, 1979; Ostlind *et al.*, 1979). The addition of this latter property started a new chapter in the treatment of endoparasitic infections and ectoparasitic infestations in animal and human medicine. It was the unique combination killing of endo- and ectoparasites by the avermectins that gave rise to the name 'endectocide'.

Avermectins have been reported to exert numerous pharmacological effects on different animals (Turner & Schaeffer, 1989). Recent evidence shows that they interact stereoselectively and with high affinity to an invertebrate specific glutamate-gated chloride channel distinct from GABA-sensitive chloride channels (Schaeffer & Haines, 1989; Arena *et al.*, 1991; Arena *et al.*, 1992). The subsequent chloride ion flux is presumed to cause the observed paralysis and death in nematode and arthropod species. All available information indicates that the milbemycins have the same mode of action as the avermectins (Conder *et al.*, 1993; Shoop *et al.*, 1993; Arena *et al.*, 1994)

Curious is the fact that although avermectins and milbemycins have been used intensively over the last decade to control parasites and pests of man, animals, and crops, it still is not known whether these molecules confer any protection to the very microbes that make them or whether they are but fortuitous metabolic by-products.

Structures of naturally occurring avermectins and milbemycins

The naturally occurring avermectin and milbemycin molecules are composed of a 16-membered macrocyclic backbone to which is fused a hexahydro-benzofuran unit from C-2 to C-8a and a spiroketal unit from C-17 to C-25 (Figure 1). The furan ring is not closed in some milbemycins. There is a bisolean-drosyloxy substituent attached at the C-13 of avermectins, whereas that position is unsubstituted in milbemycins. There can be several different alkyl substituents at C-25.

The avermectins are produced as a mixture of eight different components from fermentation of *S. avermitilis*. The natural components are denoted A_{1a}, A_{1b}, A_{2a}, A_{2b}, B_{1a}, B_{1b}, B_{2a}, and B_{2b}. The A-components have a methoxy group at the 5-position, whereas the B-components have a hydroxy group; the 1-components have a double bond between the 22- and 23-position, whereas the 2-components have a single bond with a hydroxy group at the 23-position; and the a-components have a secondary butyl sidechain at the 25-position, whereas the b-components have an isopropyl substituent at the 25-position.

The naturally occurring milbemycins from fermentation of *S. hygroscopicus* and *S. cyaneogriseus* fall into a similar pattern. They too could be subdivided into A- and B-components based on hydroxy or methoxy groupings at the 5-position. Rather than having a 1-component, however, they are found with a single bond between the 22- and 23-position similar to the avermectin 2-components. Naturally occurring C-25 substituents include groupings like methyl, ethyl, or sidechains with a trisubstituted double bond (Takiguchi *et al.*, 1980; Carter *et al.*, 1987), and mutant *S. hygroscopicus* have been found with isopropyl substituents at C-25 like the avermectin b-components (Mishima *et al.*, 1983).

Structure/Activity Relationships

With respect to the avermectins, of the eight natural components produced by *S. avermitilis* only A_{2a}, B_{1a}, and B_{2a} are produced in quantity in fermentation and, of those, B_{1a} possesses the highest potency and breadth of spectrum to include nematode, insect, and acarine species. For example, anthelmintic potency and spectrum were determined against six species of nematodes in experimentally infected sheep (Table 1). It was data such as these that revealed the combination of B- and 1-components provided the potency and spectrum desired.

Consequently, it is not surprising that the majority of effort on the avermectins has been directed toward members in the B_1 series. These components were found subsequently to be the most important in the milbemycins as well. It should be noted that separation of a- from b-components in large scale fermentation is impractical and, fortunately, unnecessary because these two homologs have virtually identical activities. Therefore, the avermectin literature often refers only to A_1, A_2, B_1, and B_2 and it is usually inferred, if not explicitly stated, that each one of these occurs as a mixture of a- and b-components; and because the a-component is produced in greater proportion during fermentation, terminology such as avermectin B_1 consists of not less than 80% a-component and not more than 20% b-component' is often used. These verbalized descriptions of the mixtures can lead to confusion because typically only the more abundant a-components are shown in structure drawings (as is done in Figs. 1 and 2).

Ivermectin, 22,23-dihydro-avermectin B_1, was the first avermectin or milbemycin to be commercialized (Chabala *et al.*, 1980; Egerton *et al.*, 1980). It was synthesized by selective hydrogenation of the planar 22,23-double bond of avermectin B_1, which gave it the same chair

conformation found in the B_2 series, and was released for use in animals in 1981. Its wide spectrum, unprecedented potency, good safety margin, and new mode of action quickly made it the treatment of choice for nematode and arthropod parasitisms in cattle, sheep, goat, swine, and horses (Campbell *et al.*, 1983; Campbell & Benz, 1984). Various formulations have been optimized for oral, subcutaneous, ruminal, and topical deliveries in veterinary medicine. Ivermectin was later found to have very important activity against filarid nematode infections of dogs (heartworm) and humans (onchocerciasis or river blindness).

The discovery and successful commercialization of ivermectin led to major chemical and microbiological efforts to explore the avermectins. Convenient chemical procedures for the stepwise removal of the bisoleandrosyl moiety were already at hand and it was of great interest to examine the deglycosylated avermectin derivatives to find how they compared to the milbemycins. For example, removal of the distal oleandrose from ivermectin (22,23-dihydro-avermectin B_1) via acid hydrolysis produced an ivermectin monosaccharide (22,23-dihydro-avermectin B_1 monosaccharide) that is only slightly less potent as an anthelmintic than the parent, while removal of both oleandroses produced an ivermectin aglycone (22,23-dihydro-avermectin B_1 aglycone) containing a 13-a-hydroxy group (Figure 2), which was surprisingly almost devoid of useful antiparasitic activities in animal health (Chabala *et al.*, 1980). This led to the question whether the 13-deoxy-ivermectin aglycone (22,23-dihydro-13-deoxy-avermectin B_1 aglycone) and the closely related milbemycins would possess any nematocidal activity at all.

The major structural difference between the ivermectin aglycone (22,23-dihydro-avermectin B_1 aglycone) and the milbemycins that was determined to be responsible for the lack of activity was the a-hydroxy group at the 13-position. Removal of that 13-a-hydroxy structural entity yielded the less polar and more milbemycin-like 13-deoxy-ivermectin aglycone (22,23-dihydro-13-deoxy-avermectin B_1 aglycone) and restored potent anthelmintic activity (Table 2). In fact, the 22,23-dihydro-13-deoxy-avermectin B_{1b} component is Milbemycin D (Mrozik *et al.*, 1983).

Subsequently, a number of lipophilic 13-substituents such as chloro, fluoro, methoxime, or certain alkoxy groups were also found to restore activity (Mrozik *et al.*, 1989). Polar substituents at the 13-position such as the hydroxy and amino groups showed diminished activity. It is sometimes assumed that the bisoleandrosyl or disaccharide moiety at the 13-position would make the molecule more polar. However, close scrutiny of the bisoleandrosyl group reveals it to possess only a single hydroxy group, which is atypical of most sugars, and thus this disaccharide moiety is more lipophilic than might first appear.

Little modification has occurred in the hexahydrobenzofuran region. It was realized early that the hydroxy at the 5-position was essential for high potency and broad-spectrum. Any removal of that hydroxy and replacement with a methoxy or ketoxime, irrespective of whether it is an avermectin or milbemycin, has generally led to compounds with reduced potency and spectrum (Mrozik *et al.*, 1982).

Chemical modification in the spiroketal region was first rewarded when the double bond between the 22-23-position of avermectin B_1 was saturated to produce ivermectin (Chabala *et al.*, 1980). This modification maintained the excellent potency against helminths of avermectin B_1 while providing improved insecticidal spectrum and modest gains in safety. Modifications such as the 23-hydroxy group of Avermectin B_2 or the 23-oxo or -methoxime of milbemycin have yielded spectrum shifts, but no overall potency gains relative to avermectin B_1. Other modifications to the spiroketal group, such as that between B_{1a} and B_{1b}, have almost no impact on activity. Likewise, lipophilic substituents at the 25-position such as cyclohexyls or cyclopentyls have shifted spectra only modestly and have shown no major improvement over ivermectin (Mrozik, 1994). In general, polar substituents at the 25-position have produced diminished activity.

Avermectins and Milbemycins as Narrow- or Broad-Spectrum Compounds: Theoretical Considerations

More than a decade after the commercialization of ivermectin, chemical and microbiological efforts have resulted in seven avermectins and milbemycins that have entered the markets as broad-spectrum endectocides in animal health and/or as acaricidal and insecticidal compounds for crop protection (Figures 3 and 4). These marketed compounds provide great insight into the fundamental question as to whether one can truly radically alter activity of a molecule through careful structural changes or whether one can only tweak the activity with structural changes. The answer, interestingly, depends on whether you view the avermectins and milbemycins as broad-spectrum or narrow-spectrum compounds.

With the commercial success of ivermectin, we, and others, tested hundreds of avermectin and milbemycin analogs in narrow-spectrum *in vitro* and *in vivo* tests. We also tested several hundred analogs for broad-spectrum nematode, and to a lesser extent, arthropod activity in sheep, cattle, and dogs. Not surprisingly, a shift in potency has been consistently observed within the typical nematode/acarine/insect spectrum with slight modification to the molecules. And that shift has been more important to compounds emphasizing narrow, specific bands of the spectrum. For instance, if we were analyzing a series of 100 avermectins and milbemycins against *Haemonchus contortus* in an *in vitro* test, it would not be unusual to find that activity varied within the series by two orders of magnitude. Undoubtedly, we would be able to pick an optimized compound from *that* series against *that* specific nematode species, and it would not be too surprising to find that it may actually be more potent against that one species than any of the presently commercialized avermectins or milbemycins. If the potency of this compound was verified in sheep and if *H. contortus* represented a stand-alone market, then this would be a very valuable finding.

However, with respect to broad-spectrum activity, experience has shown that if we experimentally infect sheep with six different nematode species, including the one species in the case above, and then administer the same 100 avermectins and milbemycins, we will find that Compound #1 kills *H. contortus* at 200 µg/kg, and then kills the other five species at dosages varying between, say, 2 and 200 µg/kg. Compound #100 kills the *H. contortus* at 2 µg/kg, and then kills the additional five species at dosages varying between 2 and 200 µg/kg as well. The other 98 compounds each kill the *H. contortus* in rank order as predicted from the *in vitro* test and produce their own unique 'spectral fingerprints' against the other species between 2 and 200 µg/kg. Although each avermectin and milbemycin maintained the same relative potency *in vivo* as in the narrow spectrum *in vitro* test, all required at least 200 µg/kg to eliminate the dosage-limiting species for the full broad-spectrum claims.

What this contrived data set demonstrates is that the specific chemical properties necessary for absorption, transport, and killing of one parasite species in the abomasum are rarely the same properties necessary to kill the other five inhabiting other parts of the body. This is true with ectoparasites as well. When dealing with broad-spectrum compounds, we are held at the mercy of the dosage-limiting organism and our compound's potency is defined by that organism.

This example, although theoretical and simplistic, represents in essence the history of the search for superior avermectins and milbemycins. The question we posed as to whether there can be fundamental change of activity through alteration of the molecule is clearly in the affirmative when we view the avermectins and milbemycins in a narrow-spectrum perspective. However, the reality of animal health medicine is that there are few parasite species that, on their own, can

justify the development of any avermectin or milbemycin. The developmental costs of these compounds for specific narrow-spectrum uses, like ivermectin against heartworm (*Dirofilaria immitis*) or onchocerciasis (*Onchocerca volvulus*), have been subsidized largely by their primary use as broad-spectrum drugs.

When our question of 'whether there can be fundamental change of activity through alteration of the molecule' is viewed from the broad-spectrum perspective, the collective experience of several major pharmaceutical companies has unfortunately been no. We have already noted 1) the chemical properties necessary to kill one parasite are not the same required to kill the others and 2) the difference in dosage between the most susceptible and the dosage-limiting species is often more than an order of magnitude, and these factors have acted in concert to create a consequence that we now introduce as 3) no avermectin or milbemycin has been able to break through the 200 µg/kg barrier against the approximately two dozen or more species that comprise the nematode/acarine/ insect spectrum in the core cattle and sheep markets. As a result, the markets have not seen fundamental changes in broad-spectrum avermectins and milbemycins over the last decade from when the original avermectin was introduced. The standard set in 1981 with the release of ivermectin consisted of a dosage of 200 µg/kg that was fully active against virtually every conceivable nematode species and most of the important acarine and insect species in the cattle and sheep markets. Recent avermectin and milbemycin additions to the broad-spectrum armamentarium each have emphasized a narrow band of the 'spectral fingerprint', but they have essentially the same overall spectrum and a dosage of 200 µg/kg.

We now illustrate these concepts with concrete examples from the marketed avermectins and milbemycins.

Broad-Spectrum Compounds for Large Animals

The avermectins were discovered in 1975, and six years later the semi-synthetic ivermectin was first introduced commercially for animal use. Ivermectin's novel mode of action made it fully active against parasites known to be resistant to other antiparasiticides. To say that ivermectin's potency and spectrum were revolutionary would be an understatement. Table 3 presents only the species in cattle for which there are registered claims with a single subcutaneous dosage at the manufacturer's recommended use level of 200 µg/kg. This dosage in cattle was established when it was found that nematode targets such as adult *Cooperia oncophora* and *Nematodirus helvetianus* required that delivery rate for high efficacy.

Since 1981, ivermectin has been released in over 60 countries for use not only in cattle, but also in sheep, goats, horses, swine, dogs, camels, reindeer, bison, and man. Large scale use of ivermectin over the last decade has not resulted in the development of resistance in any cattle, horse, or swine parasites, while laboratory selection or intensive abuse in sheep and goats has resulted in development of a limited number of nematode strains which show resistance (Shoop, 1993).

Abamectin, avermectin B_1, is a natural product from fermentation of *S. avermitilis* and is the starting material for ivermectin. Its simpler production combined with excellent potency and spectrum of its own against nematodes and acarines could not be ignored, and it too was subsequently released for use in cattle at a 200 µg/kg dose. The species spectrum for abamectin in cattle can also be seen in Table 3. Since abamectin is more potent against nematodes than ivermectin (Egerton *et al.*, 1979; 1980) and less potent against some arthropods, its dosage was selected by not only taking data on

nematodes into consideration, but also data on sucking lice, *Linognathus vituli*, and cattle ticks, *Boophilus microplus*, were considered as well (Benz & Cox, 1989). Abamectin remains the only avermectin or milbemycin that is used in both animal health and crop protection. Its use in crop protection is as an acaricide and insecticide.

The third avermectin or milbemycin released as an endectocide was moxidectin, a 23-methoxime LL-F28249a milbemycin. Moxidectin is a semi-synthetic whose starting material, nemadectin or LL-F28249a, is a fermentation product from *S. cyaneogriseus*. It was introduced at a recommended dosage of 200 µg/kg for use in sheep and cattle. Within the typical spectrum of claims for avermectins and milbemycins, moxidectin is more similar to abamectin than ivermectin in that its anthelmintic properties are superior to its insecticidal properties; for example, there is little or no activity for moxidectin at 200 µg/kg against *Dermatobia* or *Damalinia*. Thus, although technically an endectocide, moxidectin's activity has made it more of a narrow-spectrum compound for helminths than the more balanced endectocidal capabilities of ivermectin. The dosage-limiting nematode species in cattle for moxidectin appear to be *Cooperia oncophora* (Ranjan *et al.*, 1992) and *Nematodirus helvetianus* (Cyanamid Product Manual Technical Bulletin, 1994), both of which require the full dose for complete efficacy.

Moxidectin has a 'spectral fingerprint' in sheep in which *H. contortus* and *Ostertagia circumcincta* appear to be among the most susceptible nematodes. When use-level moxidectin was tested in sheep against mildly ivermectin-resistant *H. contortus* it was found to be efficacious against them (Craig *et al.*, 1992; Pankavich *et al.*, 1992). This led to the erroneous conclusion that this milbemycin may have a different mode of action from ivermectin. This error was corrected when careful titration of moxidectin and ivermectin was made against ivermectin-resistant and -susceptible worms. It was shown that more moxidectin was required to remove ivermectin-resistant worms than was required to remove ivermectin-susceptible worms (Conder *et al.*, 1993; Shoop *et al.*, 1993). It is concluded that use of moxidectin against ivermectin-resistant worms as a general practice is irresponsible.

Consequently, there is no evidence that the milbemycins have a different mode of action from the avermectins. To the contrary, all data and logic support the presence of a similar mechanism of action whether they be from structural comparison of the molecules which show them to be almost superimposable (Shoop, 1993), electrophysiological studies indicating similar chloride ion channel-based pharmacologic effects (Conder *et al.*, 1993; Arena *et al.*, 1994), mutual resistance in several species of nematodes (Shoop *et al.*, 1993; Conder *et al.*, 1993), competitive inhibition of ivermectin binding to *Caenorhabditis elegans* membranes by moxidectin (Arena *et al.*, 1994), similar toxic signs in mammals (Sasaki *et al.*, 1990), and virtually identical spectrum of activity against a phylogenetically unusual assortment of nematode, insect, and acarine parasites.

Since the activity against an ivermectin-resistant strain by moxidectin can no longer be ascribed to a different mode of action, one now finds in the literature general statements that moxidectin must then be a more potent compound than other avermectins and milbemycins (Kieran, 1994). It must be remembered that a broad-spectrum compound is defined by its dosage limiting parasite, not by the most susceptible. Moxidectin, like all of the registered compounds, requires its full dose for its broad-spectrum claims. Any inference that moxidectin is more potent can only be considered if one views it as a narrow-spectrum compound and not the endectocide it was meant to be. Intuitively, if moxidectin were 2-fold more potent than other avermectins and milbemycins, then its use level would be 100 µg/kg, not 200 µg/kg.

Doramectin, 25-cyclohexyl-avermectin B_1, was the fourth avermectin or milbemycin endectocide to be introduced at 200 µg/kg for production animals. It is a fermentation product from a mutant *S. avermitilis* strain. Its spectrum of activity is very similar to ivermectin, abamectin, and moxidectin (Jones *et al.*, 1993; Logan *et al.*, 1993).

Narrow-Spectrum Compounds for Dogs

Four avermectins and milbemycins are registered for use in dogs at present. What all have as their core claim is the nearly absolute activity against developing larvae of heartworm, *Dirofilaria immitis*, and all are used in monthly programs for prophylaxis against this parasite. There is no endectocidal activity in dogs with these compounds such as occurs in production animals. In fact, not only has there been no insect or mite claims, but even within the nematodes there is only a very narrow spectrum. For example, none of the compounds has any useful activity at their marketed dosages against common nematodes such as *Toxascaris leonina, Uncinaria stenocephala, Strongyloides stercoralis*, any of the spirurids, or adult *D. immitis*, and certainly there is no suggestion that they would be active against tapeworms common in dogs.

One reason activity against nematode species in the intestinal tract has been limited is because certain genetic lines of collies were found to be sensitive to the avermectins and milbemycins (Pulliam *et al.*, 1985; Paul *et al.*, 1987). The desire to use these compounds safely in all dogs, including collies, has required the use of dosages below those levels necessary for complete broad-spectrum activity against species in the intestinal tract.

Ivermectin is given orally at 6 µg/kg for heartworm prophylaxis and at this dosage is safe in all dogs including those genetic lines of collies defined as ivermectin sensitive. Thus, in the sensitive collie, ivermectin has approximately a 20-fold safety factor. At 6 µg/kg, ivermectin has partial activity against one hookworm (*A. caninum*) but not enough for a claim (Egerton *et al.*, 1985; Clark *et al.*, 1992). Additional claims have been gained by combining ivermectin with pyrantel pamoate (Clark *et al.*, 1992).

Milbemycin D or 22,23-dihydro-13-deoxy-avermectin B_{1b} aglycone has only been available in Japan. It is a fermentation product from a mutant *S. hygroscopicus* strain and is given to dogs orally at a 1000 µg/kg dosage. Although it has been tested in some collies for toxicity (Sasaki *et al.*, 1986c), it has not been tested in collies defined to be ivermectin sensitive. Milbemycin D has been reported to be prophylactic against developing heartworm and active against adult *A. caninum* and *Toxocara canis* (Sakamoto *et al.*, 1984; Sasaki *et al.*, 1986a; 1986b).

Milbemycin oxime is a semi-synthetic derivative of the natural product, milbemycin A_3/A_4. Replacement of the parent hydroxy with a ketoxime at the 5-position has produced a less potent compound than the parent, but it also provided a commensurate increase in safety. As a result, this compound is used orally at 500 µg/kg and, like ivermectin, has been reported to have an adequate safety margin even in ivermectin sensitive collies (Tranquilli *et al.*, 1991). At that 500 µg/kg dosage, milbemycin oxime is prophylactic against heartworm and has activity against *A. caninum, Toxocara canis*, and *Trichuris vulpis* (Bowman *et al.*, 1988; Wade *et al.*, 1991; Stansfield & Hepler, 1991; Blagburn *et al.*, 1992).

Moxidectin has recently been developed for use in dogs. An oral dosage of 3 µg/kg is predicted to be prophylactic against developing heartworm and is reported safe in ivermectin sensitive collies. There likely will be no additional claims against helminths in the gastrointestinal tract.

Narrow- or Broad-Spectrum Compounds: Epilogue

After looking at the marketed avermectins and milbemycins we find in practical terms the effects of the theoretical problems mentioned earlier. With regard to the broad-spectrum issues, there are avermectins and milbemycins that specialize in certain narrow bands of the helminth/insect/ acarine spectrum and there are avermectins and milbemycins that find a balance amongst this spectrum. Each compound has its own strengths, its own unique 'spectral fingerprint', and its own dosage-limiting species, but what is most interesting is how exact the final dosages and how similar the overall claims of these marketed compounds are for broad-spectrum use.

With regard to narrow-spectrum avermectin and milbemycins, we must look to dogs for comparison. There it is the activity against developing *D. immitis* which reveals the several orders of magnitude difference between avermectin and milbemycin compounds that we had theorized previously with the *Haemonchus contortus* model. Marketed dosages for avermectins and milbemycins for dogs range from 3 to 1000 μg/kg to achieve this claim against *D. immitis*. It would be very instructive to titrate the various avermectins and milbemycins against the entire immature and adult nematode spectrum in dogs to identify the various dosage-limiting worms. It would not be too risky to predict that each avermectin and milbemycin would have its own individual 'spectral fingerprint' and its own dosage-limiting worm. The interesting question is whether they would all require a similar dosage for broad-spectrum control.

Conclusions

The avermectins and, to a much lesser extent, the milbemycins, have revolutionized antiparasitic and antipest control over the last decade. Much effort has been expended searching for broad-spectrum second generation products, but none has exceeded the original in any fundamental way. Newer avermectin and milbemycin compounds that have appeared claim niches in the marketplace based on emphasis of certain narrow parts of the overall spectrum. Fundamental changes such as appeared in the benzimidazole broad-spectrum class of anthelmintics cannot be pointed to; for instance, within a similar period of time after the introduction of thiabendazole, newer benzimidazoles were brought to market with potency several times that of thiabendazole and with spectrum changes adding new phyla to the claims.

Why this has not happened with the avermectins and milbemycins is difficult to say. Perhaps a different story would be told if A_1 or a weaker milbemycin had reached the market first.

It is concluded that, at present, there are no second generation avermectins and milbemycins. All avermectins and milbemycins marketed subsequent to ivermectin are viewed as siblings of the first generation. Fundamental changes that could make the next leap toward a true second generation avermectin or milbemycin include analogs offering tapeworm and/or fluke activity, complete spectrum of intestinal nematodes in dogs and cats, activity against adult heartworm in dogs, activity against ectoparasites in companion animals, increased spectrum for broad-spectrum use in man, and reduced tissue residues allowing zero milk discard in lactating animals.

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Table 1. Efficacy and spectrum of avermectin derivatives against 6 species of nematodes in sheep.

Structure	Dose (mg/kg)	Efficacy					
		Hc	Oc	Ta	Tc	Csp	Oec
Avermectin A_1	0.1	2	2	0	0	2	0
Avermectin A_2	0.1	3	3	3	3	0	3
Avermectin B_1	0.1	3	3	3	3	3	3
Avermectin B_2	0.1	3	3	3	3	2	3

Hc = Haemonchus contortus, Oc = Ostertagia circumcincta, Ta = Trichostrongylus axei, Trichostrongylus colubriformis, Csp = Cooperia spp., Oec = Oesophagostomum columbianum.
0 = <50%, 1 = 50-74%, 2 = 75-90%, 3 = >90% efficacy.

Table 2. Efficacy and spectrum of avermectin derivatives against 6 species of nematodes in sheep.

Structure	Dose (mg/kg)	Efficacy Hc	Oc	Ta	Tc	Csp	Oec
22,23 dihydro Avm B_1	0.1	3	3	3	3	3	3
22,23 dihydro Avm B_1 Monosaccharide	0.3	3	3	3	3	2	3
22,23 dihydro Avm B_1 Aglycone	3.0	1	2	3	3	1	3
22,23 dihydro-13-deoxy Avm B_1 Aglycone*	0.1	3	3	3	3	3	3

Hc = Haemonchus contortus, Oc = Ostertagia circumcincta, Ta = Trichostrongylus axei, Trichostrongylus colubriformis, Csp = Cooperia spp., Oec = Oesophagostomum columbianum.
0 = <50%, 1 = 50-74%, 2 = 75-90%, 3 = >90% efficacy.
* 22,23 dihydro-13-deoxy Avm B_{1b} aglycone can be obtained synthetically from ivermectin starting material (Mrozik et al., 1983) or from fermentation of mutant S. hygroscopicus (Mishima et al., 1983); when obtained from the latter it was given the name Milbemycin D.

Table 3. Species in cattle for which there are efficacy claims for ivermectin given subcutaneously at 200 ug/kg.

Nematoda	Arthropoda
Gastrointestinal worms	Cattle grubs
*Bunostomum phlebotomum**	*Dermatobia hominis**
*Cooperia oncophora**	*Hypoderma bovis**
*C. pectinata**	*H. lineatum**
*C. punctata**	
*C. sp.**	Screworm fly larvae
*Haemonchus placei**	*Chrysomya bezziana*
Mecistocirrus digitatus	*Cochliomyia hominivorax*
*Nematodirus helvetianus**	
N. spathiger	Sucking lice
*Oesophagostomum radiatum**	*Haematopinus eurysternus**
*Ostertagia lyrata**	*Linognathus vituli**
*O. ostertagi**	*Solenopotes capillatus**
*Strongyloides papillosus**	
Toxocara vitulorum	Biting Lice
*Trichostrongylus axei**	*Damalinia bovis*‡
*T. colubriformis**	
Trichuris spp.*	Mange mites
	*Psoroptes ovis**
Lungworms	*Sarcoptes scabiei* var *bovis**
*Dictyocaulus viviparus**	*Chorioptes bovis*‡
Skin worms	Ticks
Parafilaria bovicola	*Boophilus microplus**
	B. decoloratus‡
Eye worms	*Ornithodoros savignyi*
Thelazia spp.	

‡ Species in cattle for which ivermectin has an 'aid in the control of' claim.
* Species in cattle for which there are efficacy claims for abamectin given subcutaneously at 200 ug/kg. Abamectin also has efficacy claims against *Cooperia surnabada* and *C. spatula*.

Figure 1. Avermectin B_{1a}, the most abundant component from fermentation of Streptomyces avermitilis.

Figure 2. Sequential removal of sugar groups from ivermectin and then removal of the 13-alpha hydroxy substituent.

Figure 3. Avermectins commercialized for animal health or crop protection.

Figure 4. Milbemycins commercialized for animal health or crop protection.

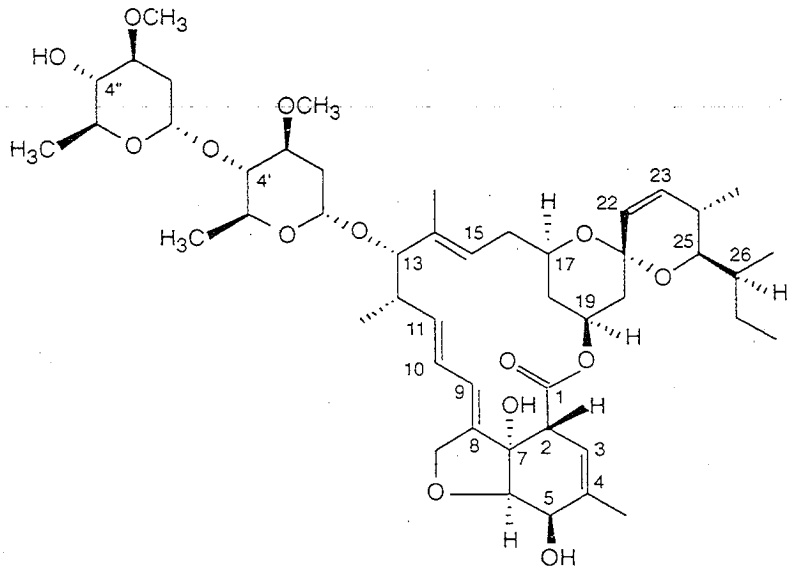


Figure 1. Avermectin B_{1a}, the most abundant component from fermentation of Streptomyces avermitilis.

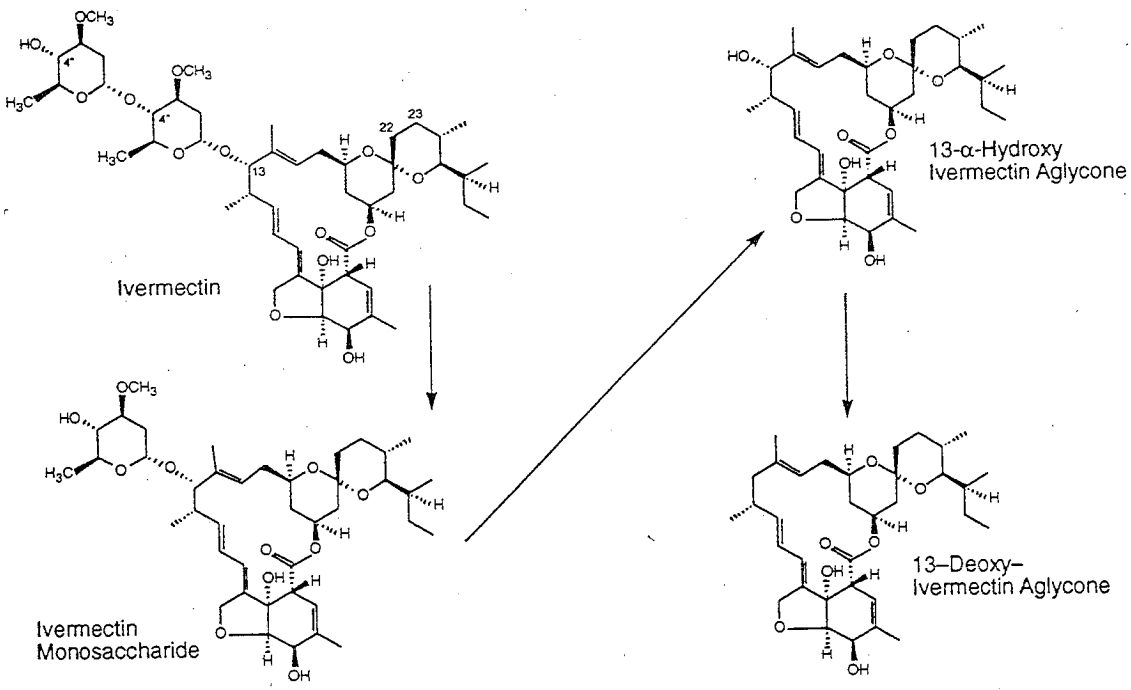
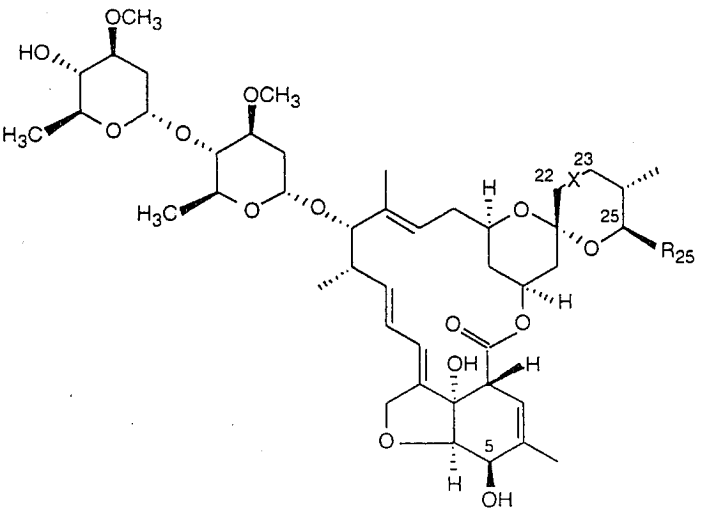
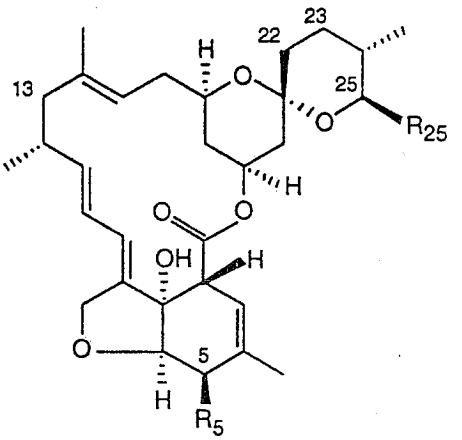


Figure 2. Sequential removal of sugar groups from ivermectin and then removal of the 13-alpha hydroxy substituent.



Ivermectin: X = $-CH_2CH_2-$ R_{25} = $CH(CH_3)CH_2CH_3$ and $CH(CH_3)_2$
Abamectin: X = $-CH=CH-$ R_{25} = $CH(CH_3)CH_2CH_3$ and $CH(CH_3)_2$
Doramectin: X = $-CH=CH-$ R_{25} = Cyclohexyl

Figure 3. Avermectins commercialized for animal health or crop protection.



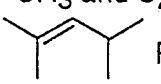
Milbemycin D: R_5 = —OH R_{25} = $-CH(CH_3)_2$
Milbemycin A_3/A_4: R_5 = —OH R_{25} = CH_3 and C_2H_5
Milbemycin A_3/A_4 5–Oxime: R_5 = =NOH R_{25} = CH_3 and C_2H_5
Moxidectin: R_5 = —OH R_{25} =  R_{23} = $=NOCH_3$

Figure 4. Milbemycins commercialized for animal health or crop protection.