

**Chemicals for control of the Old World screw-worm fly  
*Chrysomya bezziana* in Australia**

**PJ James<sup>1</sup>, PE Green<sup>1</sup>, R Urech<sup>1</sup> and JP Spradbery<sup>2</sup>**

<sup>1</sup>Animal Research Institute, Department of Primary Industries and Fisheries, 665  
Fairfield Road Yeerongpilly Qld 4105

<sup>2</sup>XCS Consulting Pty Ltd, GPO Box 2566, Canberra, ACT 2601

## Executive Summary

The Old World screw-worm, *Chrysomya bezziana* (OWS) is considered to be the most serious exotic pest threatening Australia's livestock industries and is endemic in a number of our neighbouring countries. New World screw-worm (*Cochliomyia hominivorax*) (NWS) and Palaeartic screw-worm (*Wohlfahrtia magnifica*) (PSW) are other potential invaders but considered less of a threat. The use of chemical pesticides to treat screw-worm fly (SWF) infestations and for strike prophylaxis would be a key component of any response plan to address a screw-worm incursion.

This review, prepared for Animal Health Australia, considers chemicals with potential for use against OWS in the event of an incursion. Implications for the conduct of an eradication program and aspects including the possibility of development of resistance, potential for residues in produce and environmental impact are considered and other potential control strategies are noted.

Chemicals are considered in the five following categories:

- products currently registered with Australian Pesticides and Veterinary Medicines Authority with a claim against OWS,
- products registered in Australia for animal use and with known efficacy against OWS but no current claim,
- products registered for animal use and likely effectiveness against OWS but with no current claim,
- chemicals not registered in Australia, but with likely effectiveness for screw-worm control and
- other chemicals that might be effective but have yet to be evaluated.

Known efficacy is defined as the availability of published information or chemical company data that demonstrate efficacy against OWS.

All formulations currently registered with a claim against OWS in Australia are based on ivermectin, administered by subcutaneous injection to cattle. All of these formulations have meat withholding periods of one month or longer and most are not registered for use on animals producing milk for human consumption. There are no products with a claim against screw-worm registered for use in sheep.

In the event of a screw-worm fly incursion into Australia, chemical control products registered for animal application and with known efficacy against SWF but without a specific claim could be approved for use at short notice under an Emergency Use Off-Label Permit. Products currently registered in Australia which have shown efficacy against screw-worm infestations can be divided into those that are primarily curative in their action and those that provide extended protection and may be able to fulfill a prophylactic role.

Insecticides that could be used for the treatment of animals with SWF infestations include the organophosphates diazinon, chlorfenvinphos and fenthion, macrocyclic lactones, applied topically or systemically, spinosad and possibly some synthetic pyrethroids. Ivermectin

administered systemically is effective against early OWS larvae but may not reliably kill older larvae. Topical application of ivermectin may be more effective against late stage larvae. Spinosad is a relatively newly registered compound that has the attraction of a nil withholding period and which is approved for use on organic properties by a number of certifying bodies. It could also be used to provide short term protection for animals during transport to market.

The list of chemicals shown to provide significant prophylactic effect against OWS is limited to macrocyclic lactones (MLs), closantel and zeta-cypermethrin formulated in ear tags. Studies with the currently registered injectable formulations of ivermectin indicated protection periods varying between two weeks and one month, a pour-on formulation of eprinomectin protected for 3 days but failed at 7 days and a doramectin pour-on protected for 7 days but failed at 14 days. Zeta-cypermethrin ear tags provided up to four months protection although low level strike was recorded during the later part of this period and the likely efficacy of the tags in protecting more severe predisposing lesions such as castration wounds has been questioned. These tags could play a significant role in integrated approaches and may be of particular use on dairy enterprises as they have a nil milk withholding period. The dose of closantel required for extended protection against OWS is higher than presently registered for use in sheep and there is a risk of inducing optic neuropathy, particularly in young animals, at these higher rates.

A number of more recently registered ML products have shown encouraging results against other myiasis species but are yet to be tested against OWS. These include injectable doramectin, which gave superior protection to ivermectin in studies against NWS, long acting, capsule and bolus formulations of ivermectin and injectable abamectin.

Reliance on only one chemical class for prophylaxis is considered inadvisable because of the pressure it would exert for the development of resistance and the implications for a control program should this occur. The insect growth regulator dicyclanil and the 6% cyromazine spray on formulation, both registered for application to sheep, require testing. Dicyclanil provides up to 6 months protection against wool myiasis and has been shown to protect against NWS in castration wounds for up to 16 days, the period required for healing. Dicyclanil could provide a valuable tool for use against SWF in the event of an incursion.

A range of other chemicals and biopesticides likely to have effect against OWS, but not currently registered for animal use in Australia and yet to be evaluated against OWS are also considered. It is unlikely that companies will be willing to undertake the safety and toxicological studies required to register a new compound in Australia for use against screwworm alone. Fipronil has shown effectiveness against NWS and has been registered overseas for the control of ticks and other parasites in ruminants. Fipronil appears to have the most potential for future use of those insecticides not presently registered in Australia.

All future testing against OWS should be conducted according to the guidelines recently developed by the World Association for the Advancement of Veterinary Parasitology (Holdsworth *et al.* 2005). The chemicals given highest priority for future testing include injectable doramectin, capsule, bolus and long acting formulations of MLs, dicyclanil, injectable abamectin and 6% cyromazine spray.

## Chemicals for the control of Old World screw-worm fly *Chrysomya bezziana* in Australia

### 1. Introduction

The Old World screw-worm fly, *Chrysomya bezziana* (OWS), is considered to be the most serious exotic insect pest threatening Australia's livestock industries. It would also have medical implications for the Australia's human population should it become established. Screw-worm fly is endemic in our closest neighbouring countries, Papua New Guinea and Indonesia. While the likelihood of introduction of OWS to Australia is considered to be low, the risk is real, especially with increasing live animal exports to the SE Asian region and subsequent return of transport vessels to northern Australian ports.

The OWS is an obligate parasite of warm-blooded animals, the females laying batches of eggs at the edge of wounds caused accidentally or through management practices such as castration and de-horning. The navels of the new born and the vulva region of their mothers are highly susceptible to strike. Body orifices such as nose and ears are also prime targets for ovipositing screw-worm flies (SWF). During the 6-7 days of feeding, the larvae burrow deeply into the host tissue, destroying muscle and causing severe blood loss. Reduction in productivity, maiming or even death can result from screw-worm infestations.

The strategic use of insecticides was pivotal to the successful eradication of the New World screw-worm fly, *Cochliomyia hominivorax* (NWS), from the USA, Central America and Libya and use of chemical pesticides to treat screw-worm infestations and protect wounds from strike will be a significant component of any response plan invoked following an incursion of SWF into Australia. A wide range of chemicals has been used to treat screw-worm infestations, particularly for NWS in the USA and Central and South America (reviewed in part by Graham 1979; Drummond *et al.* 1988; Spradbery 1994). Studies on chemical control of OWS have been less intense but a range of insecticides (Spradbery *et al.* 1976, 1991), acaricides (Spradbery *et al.* 1983), the salicylanilide, closantel *per os* (Spradbery and Owen 1990) and macrocyclic lactones (Spradbery *et al.* 1985, Perkins 1987, Wardhaugh and Mahon 2002, Wardhaugh *et al.* 2001a) have been evaluated. Many of these products are not now available and many chemical products studied and found to be effective for control of screw-worm have not been registered for such use in Australia. In particular, coumaphos, which was hailed by Reichard (1999) as the 'work horse' insecticide for NWS control and the standard for any new treatments, is no longer registered for animal use in Australia.

This review, commissioned by Animal Health Australia, discusses both registered (Table 1) and unregistered chemicals with demonstrated or likely efficacy against OWS, with a focus on production animals. It should be emphasized that in the event of a SWF incursion into Australia, chemical control products with known efficacy but without a specific SWF claim could be approved at short notice following application for an Emergency Use (Category 43, off-label permit) from the Australian Pesticides and Veterinary Medicines Authority

(APVMA). Such an application attracts no fee and there is no set period for assessment. For this reason chemical products are considered in five main categories:

- products registered with APVMA with a claim against OWS,
- products registered for animal use and known effectiveness against OWS but with no current claim,
- products registered for animal use and likely effectiveness against OWS but with no current claim,
- chemicals not registered in Australia, but with likely effectiveness for screw-worm control and
- other chemicals that might be effective but have yet to be evaluated.

All information on the registration status in Australia of chemical products considered in this review and indications for their use was sourced from Infopest (Department of Primary Industries and Fisheries Queensland, October 2005) and only commercially available products were included.

### **1.1. Other myiasis flies**

The later part of this review deals with compounds not tested against OWS, but of likely efficacy. This often involves a consideration of chemicals that have been shown to be effective against other myiasis flies. The two species most similar in biology to OWS are the NWS, *Cochliomyia hominivorax*, (Diptera: Calliphoridae) and the Palaearctic screw-worm (PSW) *Wohlfahrtia magnifica*, (Diptera: Sarcophagidae) alternatively referred to as Wohlfahrt's wound myiasis fly or the fleshfly (Sotiraki *et al.* 2005a). Despite being in different genera and geographically separated, these species are remarkably similar to OWS in their biological and behavioural characteristics, including responses to treatment and prophylaxis (Reichard 1999) and results against these species are considered to gain an indication of chemicals not tested against OWS, but which are likely to be effective against this species. A brief description of the distribution, impact and biology of the other two main wound myiasis flies is given below.

***Cochliomyia hominivorax***. Historically the range of NWS extended from the southern states of the USA through Mexico, Central America and the Caribbean Islands to northern Chile, Argentina and Uruguay (Hall and Wall 1995) and covered an area of some 23 million km<sup>2</sup>. However, through a successful eradication campaign based mainly on the use of the sterile male technique, though also with extensive use of chemical treatments, NWS has been eradicated from the USA, Mexico and a number of Central American countries. Currently NWS is endemic throughout most of South America, although not Chile, with most of the Caribbean countries naturally NWS-free or free through eradication. NWS continues to be endemic in Cuba, the Dominican Republic, Haiti, Jamaica, Trinidad and Tobago (Vargas-Teran 2002).

NWS is an obligate parasite of mammals and, rarely, birds. The inability of NWS and OWS to survive low temperatures largely determines their geographic ranges (Spradbery 1994) which could extend as a result of global warming (Sutherst 2001). Currently there is no

overlap in the ranges of OSW and NWS but if left uncontrolled they would overlap considerably (Sutherst *et al.* 1989).

***Wohlfahrtia magnifica*** The PSW is largely responsible for wound myiasis in southern and eastern Europe, Russia, the Near East and North Africa (Hall, 1991, 1997; Colebrook and Wall, 2004) where it predominately affects sheep and camels, although other hosts such as goats, horses, cattle and pigs are also attacked. Unlike the two Calliphorids, PSW females are larviparous and infestations can develop rapidly. The genitalia of healthy animals are major infestation sites and PSW infestation can lead to significant reproductive problems (Sotiraki *et al.* 2005b). Similar to NWS and OWS, the larvae burrow deeply into the host's tissues so that only the posterior spiracles are exposed. Unlike NWS and OWS, the *in vitro* rearing of this species has not been satisfactorily achieved, which has retarded detailed studies in a number of areas, including chemotherapy.

Products effective for the treatment and prophylaxis of NWS and PSW infestations would be expected to have similar efficacy against OWS because of the close biological and behavioural similarities of these species. However some caution is required in extrapolating from one species to the other. For example, ivermectin, is not considered effective either as a prophylactic or curative treatment for PSW in sheep (Farkas *et al.* 1996; Sotiraki *et al.* 2005b) although it has good effect against OWS and NWS.

There are also a number of other myiasis flies, most of which have biology that differs more markedly from OWS, but from which indications about the likely efficacy of chemical therapeutics can sometime be drawn. These include the wool myiasis flies *Lucilia cuprina* and *Lucilia sericata*, various facultative myiasis flies, most in the families Calliphoridae and Sarcophagidae and botflies (Oestridae) such as *Hypoderma* spp., *Oestrus ovis* and *Dermatobia hominis*. Results against these species are also considered where relevant.

## **2.0. Assessment of chemical products for OWS control**

### ***2.1. Products registered with the APVMA with a claim for OWS control***

The only products currently registered with a claim against OWS in Australia are all based on ivermectin and are restricted to use on cattle (Table 2). Ivermectin is a macrocyclic lactone with a class 6 mode of action. Although ivermectin is registered in Australia for use against internal and some external parasites of sheep, there is currently no product registered with an OWS control claim for sheep.

Soon after the introduction of ivermectin, Spradbery *et al.* (1985) and Perkins (1987) demonstrated its persistent efficacy against OWS in laboratory and field trials with cattle in PNG and Malaysia. Comprehensive *in vitro*, pen and field trials, including dose titration studies, demonstrated the efficacy of injectable ivermectin (Spradbery *et al.* 1985). At 200µg/kg body weight, the residual protection provided against OWS was a minimum of 14 days in pen trials and 16-20 days in field trials with cattle with castration and branding wounds. Treatment of OWS strikes containing larvae 2 to 5 days old demonstrated complete

mortality of larvae up to 2 days old and a progressive decline in mortality with age of larvae to 21% at 5 days old.

Injectable ivermectin at 200µg/kg body weight protected new-born calves against navel strike and prevented re-infestation of OWS wounds for 10-11 days (Perkins 1987). Ahmad (2002) claimed up to 28 days protection in cattle against OWS from a single ivermectin injection, but no substantiating data were presented. In addition to the ivermectin formulations currently available for use in beef and dairy cattle (Table 2), a long-acting (LA), injectable ivermectin formulation is registered, although its efficacy against OWS (Table 3) is yet to be evaluated.

Ivermectin has been recommended in AUSVETPLAN as the prophylactic drug of choice for any OWS incursion into Australia (ARMCANZ 1996). It is expected that other macrocyclic lactone products registered for animal use in Australia would have similar efficacy against OWS, but these are not currently registered for OWS control and are discussed in the next section.

## ***2.2. Products registered for animal use with known effectiveness against OWS but no current claim***

Known efficacy is defined for this review as the availability of published information or chemical company data that demonstrate efficacy against OWS. Prophylactic products containing a range of active constituents (Table 3) and wound treatments (Table 4) fall into this category. Very often efficacy is dependent on the mode of application and as far as possible this is taken into account in the following assessment. Where a chemical has been shown to be effective against OWS and is registered for use within Australia, but does not occur in Tables 3 or 4, this is often because the registered mode of application differs from that used for OWS testing.

***Macrocyclic lactones (MLs).*** Since the introduction of ivermectin in 1981, a series of highly efficient macrocyclic lactone endectocides have come onto the market. Although ivermectin administered as a subcutaneous injection is the only ML currently registered for use against OWS in Australia, other MLs and methods of administration have been shown to be effective. Wardhaugh *et al.* (2001a) evaluated pour-on formulations of doramectin, eprinomectin and moxidectin and a sustained-release (SR) bolus of ivermectin, against OWS on cattle in Malaysia. Doramectin and eprinomectin provided 7-14 and 3-7 days protection respectively while no activity, even at 3 days, was shown for moxidectin. No myiases were established on the ivermectin SR bolus treated cattle tested at 14, 21, 28 and 102 days after treatment. In studies with NWS Lessa and Lessa (1999) showed that an ivermectin sustained release bolus applied 15 days prior to castration provided complete protection until healing of wounds was completed 16 days later. Two controlled release ivermectin capsule formulations are registered (Table 5) for sheep in Australia but have not been evaluated against OWS.

***Pyrethroid Insecticides (SPs).*** Pyrethroids, applied as dips, sprays, pour-ons and ear tags are registered for ectoparasite control in both cattle and sheep. Many pyrethroids exhibit both repellent and insecticidal properties. While there is some evidence of efficacy of pyrethroids against adult NWS (Elowni *et al.* 1995, 1999), less evidence is available for OWS. Ahmad

(2002) reported high efficacy of cyphenothrin in treating OWS infestations, but provided no supporting data. In addition, Kadhim *et al.* (2000) reported that a cypermethrin pour-on was effective for the treatment of OWS in sheep and Sotiraki *et al.* (2003) indicated that 12 days protection was provided against PSW by a cypermethrin pour-on.

Although Barricade S (cypermethrin and chlorfenvinphos) was effective in treating infested wounds (Spradbery *et al.* 1991), this may have been largely due largely to the OP component of the formulation. Tozer and Spradbery (2002), using zeta-cypermethrin ear tags, demonstrated significant reductions in OWS strikes for up to 4 months in two field trials in Malaysia. However, complete protection was not achieved. In Australia these tags are registered for the control of buffalo flies and concurrently reduce *Stephanofilaria* lesions, a predisposing cause of OWS infestations. Strategic combinations of ear tags with systemic products such as the MLs, might provide effective OWS control.

Spradbery and Kirk (1992) reported observations in the United Arab Emirates where a combination of SP applications appeared to reduce OWS myiasis. Few myiasis were observed in cattle following the introduction of insecticidal ear tags (10% permethrin), use of a pour-on systemic SP (5% cypermethrin) and twice daily fogging of the cattle yards with deltamethrin whereas at nearby sheep pens, myiasis due to *C. bezziana* continued to be a problem.

**Salicylanilides.** A number of closantel products are registered at 7.5–10 mg/kg bodyweight for internal parasite control in sheep (Table 3). Closantel, like other salicylanilides, binds to plasma proteins and is effective against blood sucking helminths such as *Haemonchus contortus* for 4 and 6 weeks at 7.5 and 10 mg/kg respectively. Closantel controls all stages of *O. ovis* in sheep (Arundel 1985) and Guerrero (1984) has reviewed its systemic activity against *C. hominivorax* in sheep. Spradbery and Owen (1990) demonstrated residual protection in sheep against OWS for 8–15 days at 15 mg/kg, but lower dose rates (5 and 10 mg/kg) were ineffective. However, there are a number of reports of blindness in young sheep and goats associated with high doses of closantel (Button *et al.* 1987, Obwolo 1989, Gill *et al.* 1999, Borges *et al.* 1999, Barlow *et al.* 2002) and in at least one instance this extended to older sheep (4 years) and at dose rates as low as twice the recommended rate (Gill *et al.* 1999). Further safety testing would be required before closantel at the dose rates found to be effective by Spradbery and Owen could be recommended for OWS control.

**Organophosphorus insecticides (OPs).** Organophosphorous insecticides are cholinesterase inhibitors and of a different mode of action group (1B) to the macrocyclic lactones. Reichard (1999) suggests that the OP coumaphos is the standard for comparison of other treatments, but as previously noted it is not currently registered in Australia. Laboratory and *in vivo* testing of insecticides and acaricides included a number of OP products (Spradbery *et al.* 1976, 1983, 1991). Of these, only diazinon, chlorfenvinphos, fenthion and trichlophon are still registered for use on production animals in Australia.

Diazinon applied as a 0.5% liquid formulation resolved 89% of artificial wounds while when applied as a 1.5% powder formulation with synergized pyrethrins it was 100% effective (Spradbery *et al.* 1991). Although numerous formulations of diazinon (e.g. sprays, dips,



wound treatments, ear tags) are still registered, apart from the fly strike powder (Table 4), most indications are for application at lower concentrations than utilized in these studies. Label recommendations for diazinon applications vary from 2% for powder formulations to 0.1% for wound dressings to 0.05% for sprays and jetting fluids. In studies with *C. hominivorax*, Benitez Usher *et al.* (1997) used 2% diazinon powder to give reliable resolution of wounds in calves with navel and scrotal myiases.

Chlorfenvinphos at 0.05%, fenthion at 0.2% and chlorfenvinphos in combination with cypermethrin, available in Australia as Barricade S or Blockade S, all gave 100% strike resolution (Spradbery *et al.* 1983, 1991). In studies with NWS chlorfenvinphos at 0.05% gave results that were comparable with coumaphos (Graham 1979). Trichlorphon gave comparatively poor results against OWS (Spradbery *et al.* 1983). The topical formulation tested is no longer available, but trichlorfon is registered for oral administration and is considered in a later section of this review.

Spradbery *et al.* (1991) tested the residual efficacy of a number of OPs and concluded that it was generally poor. Organophosphates as jetting products currently give periods of protection of 3 to 6 weeks against *L. cuprina* and much longer periods were provided before the development of resistance (Levot 1995). However extended periods of protection against wool myiasis were probably the result of persistence of the chemical in the fleece. It is therefore unlikely that OPs, formulated as jetting products for *L. cuprina*, would provide comparable periods of protection against OWS (Wardhaugh and Mahon 2002). However, chlorfenvinphos, applied as an overspray or by backrubber for buffalo fly control and fenthion as a spot-on for lice control, may have some prophylactic as well as treatment properties against OWS. It appears that the primary role of OPs in any future screw-worm eradication or control program will be as wound dressings and treatments for OWS infestations. However, they may also be required to provide short term prophylaxis in some systems where more persistent chemicals are unacceptable because of residue or environmental concerns.

**Spinosyns.** Spinosad, a relatively new natural insecticide, is a neurotoxin and acts as a contact and stomach poison (Salgado 1998; Salgado *et al.* 1998; Sparks *et al.* 2001). Two products containing spinosad, one for the treatment and prevention of blowfly strike and one an aerosol for wound treatment are registered for use on sheep in Australia. Trials in South America (NWS) and Malaysia (OWS), with the spinosad aerosol spray formulation, have indicated efficacy in the treatment and prevention of screw-worms in castration wounds, navel strikes and wounds in cattle (L.B. Lowe, pers. comm.). The formulations registered for application by jetting or as a flystrike or mulesing dressing have a lower concentration of spinosad than the aerosol, but may also be effective against OWS. Spinosad has also been shown to control the cattle tick *Boophilus microplus* (Davey *et al.* 2001) and could also contribute to protection against OWS by reducing tick bite lesions that are common predilection sites for screw-worm invasion. In addition, because spinosad is a naturally derived compound it has been approved for use on organic properties by a number of certifying bodies and may provide an option for controlling OWS infestations on organic production properties. It would also be useful for treating animals during transport to market.

***Insect Growth Regulators (IGRs).*** IGRs registered for application to production animals in Australia include diflubenzuron, triflumuron, cyromazine and dicyclanil. Although all of these chemicals are classified as IGRs, the mode of action of diflubenzuron and triflumuron differs from that of cyromazine and dicyclanil and they should be considered as functionally different groups.

Cyromazine was the first IGR introduced for the control of wool myiasis. When applied as a spray to sheep and cattle at 0.2% it was ineffective against OWS (Spradbery, Tozer and Drewett unpublished data cited by Wardhaugh and Mahon 2002). However, a number of higher concentration (6%) spray-on products are now registered for application to sheep (Table 5). These carry a registered claim for 11 weeks protection against wool myiasis and should also be tested against OWS. When supplied daily in medicated feed to sheep and cattle at 1.0 or 1.5 mg/kg bodyweight respectively, cyromazine provided protection against OWS (Tozer and Drewett unpublished data cited by Wardhaugh and Mahon 2002) and when administered systemically in a slow release intraruminal capsule, cyromazine provided 12 weeks protection against larval implants with *L. cuprina* (Anderson *et al.* 1989). Neither of these formulations of cyromazine is registered for sheep or cattle in Australia and this also appears to be the case overseas. However, a feed-through for fly control in poultry is available in Australia (Larvadex) and a feed-through for horses is available in the United States (Solitude). It is notable that the horse formulation is restricted to animals not to be used for food purposes. The development of a bolus formulation of cyromazine at the dose rate required for OWS prophylaxis is held to be unlikely (J.F. Graf, pers. comm.)

### ***2.3. Products registered for animal use and likely effectiveness against OWS, but with no current claim***

Products which are registered for animal use in Australia and have been shown to be effective against NWS or PWS (but not OWS) are discussed in this section (Table 5). Some of these products are likely to also be effective against OWS and constitute a promising pool of new products for OWS treatment in Australia.

***Macrocyclic lactones.*** Macrocyclic lactone products presently registered for cattle within Australia, but that have not been tested against OWS include pour-on formulations of ivermectin, injectable doramectin, abamectin and long-acting (LA) ivermectin. Little additional work has been published on the use of MLs for OWS in production animals since that of Wardhaugh *et al.* (2001a). However, as previously indicated, products for the treatment and prophylaxis of NWS and PSW infestations are likely to have similar efficacy against OWS in most instances.

A number of reviews have been published on the use of MLs for OWS and NWS (Hall 2004, Wardhaugh and Mahon 2002, Eddi *et al.* 2002) and extensive general information on the MLs has been compiled in the books by Campbell (1989) and Vercruyse and Rew (2002). Eddi *et al.* (2002) provided a summary of South American NWS trials from 1993 to 1999, reporting efficacy of 100% for injectable ivermectin and doramectin and an ivermectin slow-release (SR) bolus and of 96.5% for injectable abamectin. However, other South American trials have demonstrated lower efficacy, particularly for ivermectin. The results not cited by Eddi *et al.* (2002) are summarized in Table 6.

Caproni *et al.* (1998) compared the efficacy of injectable doramectin and ivermectin in preventing NWS infestations in various breeds of cattle castrated 10 days after treatment in an extensive series of 91 replicate field trials in Brazil (Table 6). Mean efficacy was 94.6% (53.3-100%) for doramectin and 43.7% (0-100%) for ivermectin when cattle were examined 3-7 days post-castration. Doramectin had an efficacy higher than 90% in 80% of the trials, whereas ivermectin was higher than 90% in only 3% of the trials. The 10-day lag period between ML injection and castration may partially explain the lower efficacy of ivermectin.

Moya-Borja *et al.* (1997) compared injectable doramectin and ivermectin in two studies with experimentally infested (30 first instar NWS) calves in Brazil. Calves were challenged with larvae 2 hours post-treatment and their status evaluated daily for 7 days. Doramectin and ivermectin prevented myiasis on 100% and 50% animals respectively on day 2. Doramectin provided complete protection for 21 days and partial protection (56%) at 28 days post-treatment. Anziani *et al.* (2000), also using experimental infestations of NWS, showed that doramectin provided a reduction in NSW infested wounds of 90.0% and 83.3%, 12 and 15 days after treatment respectively. Two long acting ivermectin formulations evaluated showed no effect. Egg masses were found on treated animals in a number of the ML studies, suggesting that protection was conferred by direct larvicidal effects rather than repellency (Benitez Usher *et al.* 1997).

Benitez Usher *et al.* (1997) indicated that efficacy of therapeutics varies depending on type and size of the wound, treatment time after birth or castration and the severity of challenge with NWS. However, the variability observed in results from the South American trials could also be explained in part by differences in experimental design, test animals or formulations.

Overall these studies suggest that injectable doramectin provides superior protection to ivermectin against NWS. Although it is difficult to predict the efficacy of a particular ML based solely on plasma pharmacokinetic profiles (Wicks *et al.* 1993), doramectin demonstrated a higher bioavailability than ivermectin (Hennessy and Alvinerie 2002) and this could be the reason for its superior effect. Doramectin is registered as an injectable in Australia (Dectomax), and given its superior effect against NWS, should receive priority for testing against OWS. Even though the ivermectin pour-ons have not been evaluated against OWS, given their pharmacokinetic characteristics (Hennessy and Alvinerie 2002) it is unlikely that they would provide improved persistence in comparison with the injectable products.

Anziani *et al.* (1996) showed that abamectin applied by injection was >96% effective in protecting against myiasis due to NWS in post-castration wounds in cattle. Abamectin is registered for application to cattle in Australia and should also be tested against OWS.

Macrocyclic lactones registered for application to sheep include ivermectin (drench, capsule, jetting, dipping), abamectin (oral, injection) and moxidectin (oral, injection). By far the most common application in sheep is as an oral drench but there appears to have been no studies of the efficacy of oral drenches against screw-worm myiasis. No ivermectin injection is registered for sheep.

Ivermectin has a label claim for protection of sheep against blowfly strike (*L. cuprina*) for 12 weeks when applied as a jetting fluid. However, *L. cuprina* is a wool myiasis fly and the difficulties in extrapolating from wool myiasis to wound myiasis with topically applied agents have been previously indicated. Ivermectin controlled release capsules proved an effective aid for control of breech strike but were less efficacious against body strike (Rugg *et al.* 1998). It is likely that some protection resulted from systemic delivery. However, breech strike usually follows faecal or urine staining of wool and active levels of ivermectin can be excreted in the faeces (Wardhaugh *et al.* 2001b). It is likely that part of the observed efficacy against breech strike resulted from topical delivery of excreted ivermectin in the faeces.

In view of the absence of information on the efficacy of oral application in sheep, it is notable that orally administered ivermectin has been effectively used to control myiasis of humans. Jelinek *et al.* (1995) reported on the oral application of ivermectin to treat NWS myiasis, while de Tarso *et al.* (2004) used a single oral dose of ivermectin (200ug/kg) to successfully eliminate NWS larvae from a case of orbital myiasis. Spontaneous emigration of the larvae occurred leading to a complete resolution of the myiasis within a 48-h period. Oral formulations of MLs are designed for maximum availability over a short period (Verduyck and Rew 2001) and are likely to be useful for short term control. Topical application of 1% ivermectin in propylene glycol also successfully cured NWS myiasis in 4 patients (Victoria *et al.* 1999). Direct application to the affected area at a dose of 400ug/kg killed the majority of larvae within one hour and no live larvae were seen after 24 hours.

Even though injectable ivermectin and moxidectin are not generally considered effective against the larvae of PSW in naturally infested sheep, Farkas *et al.* (1996) indicated the potential value of application of low-concentration aqueous solutions of ivermectin for wound treatment and prophylaxis. Ruiz-Martinez (1995) produced evidence of some protection for up to 12 days against early instars of PSW in sheep from injectable ivermectin.

Coop *et al.* (2002) reviewed the use of MLs against ectoparasites of sheep and goats but with no mention of screw-worm. They indicated that Oestrosis, at all larval stages, could be treated effectively with injectable ivermectin, doramectin or moxidectin or with an oral drench of ivermectin. A controlled-release ivermectin bolus also controlled and prevented *O. ovis* infestations. Moxidectin was considered inefficient against NWS and shows little or no activity against *Dermatobia* (Shoop *et al.* 1995). No information is available on moxidectin LA or abamectin formulations registered for sheep.

Taken overall, these results suggest that amongst the parasiticides currently registered in Australia, macrocyclic lactones, when administered in formulations that provide systemic activity, are likely to be amongst the most effective preventative and therapeutic agents. In common with most other screw-worm treatments, when administered topically, penetration to provide action against deep seated infections may be a problem. Injectable, oral (particularly for sheep) and controlled release capsule formulations seem to offer the best potential and should receive priority for testing.

**Synthetic pyrethroids.** A range of synthetic pyrethroids other than those discussed in section 2.2 are also registered for use on cattle and sheep in Australia. For cattle these include deltamethrin, fenvalerate and cypermethrin sprays, a deltamethrin dipping formulation and deltamethrin, zeta-cypermethrin and permethrin pour-ons. For sheep they include deltamethrin, cypermethrin and alpha cypermethrin backliners and pyrethrins as a spray. With the exception of cypermethrin, discussed in an earlier section there are few studies against OWS. Various deltamethrin and cypermethrin products have been employed in the Middle East for OWS control but little published evidence is available on their efficacy (Spradbery unpublished data 2005). Although tested against OWS, fenvalerate is not included in Table 5 because it was ineffective (Spradbery *et al.* 1991).

Silva *et al.* (1991) tested various combinations of permethrin, ethion (an OP), chloramphenicol (an antibiotic) and gentian violet against natural and experimental infestations of NWS. The formulations were highly effective in expelling and killing NWS larvae. Anziani *et al.* (1998) described treating wounds infested with second or third stage NWS larvae with repeated applications of cypermethrin spray until the wounds healed, but no observations were made on possible residual protection. Elowni *et al.* (1995, 1999) have shown that a range of SPs, including deltamethrin, cypermethrin and cyfluthrin are toxic to adult NWS and suggested that SP pour-ons could potentially confer whole body protection because of formulation to enhance their peripheral migration. Wardhaugh and Mahon (2002) reviewed evidence for dermal absorption and short term systemic activity of some SPs and postulated that in addition to repelling adult flies, absorbed SPs might provide a degree of residual protection against new infestations in cattle. However, they note that because of the relatively thick lipid layer covering sheep skin, there is likely to be little dermal absorption and systemic activity in sheep.

Registration indications are that the long wool application of alpha cypermethrin as a backline spray at 50g/l (Vanquish) can give up to 10 weeks protection against wool myiasis. Orton and Shipp (1983) showed that deltamethrin, cypermethrin and permethrin suppressed oviposition by *L. cuprina* and had varying degrees of repellency and it is likely that alpha-cypermethrin has similar activity. Although there may be some residual protections provided by particular pyrethroid formulations, it seems that with the exception of SP ear tags, the role of pyrethroids in the event of an incursion of OWS would be limited to wound treatment and disinfestation in most instances.

**Organophosphates.** Many of the organophosphorous compounds tested by Spradbery *et al.* (1983, 1991) are no longer registered in Australia and there are only a few that were either not tested by these authors or that have been registered subsequently.

Although a topical formulation of trichlorfon was tested, trichlorfon is currently registered in Australia as an oral drench for intestinal helminth control in cattle and stomach bot control in horses. Trichlorfon is known to have systemic properties (Graham 1979) and NWS larvae in wounds were killed after oral administration to sheep at 20mg/kg (McGregor and Bushland 1956). Trichlorfon, temephos and crotoxyphos have also been shown to be effective against PSW by topical treatment of wounds (Hall and Wall 1995). Temephos is registered as a 20% pour-on formulation for cattle and for application to sheep by dipping, showering or and

jetting for lice control, although the sheep formulation is not currently marketed. Crotoxyphos, introduced for cattle tick control in 1967, is no longer registered.

In the former Soviet Union, a range of OP insecticides (crotoxyphos, trichlorfon, dichlorvos, propoxur, phosalone, temephos, chlorpyrifos, iodofenphos and diazinon) were found to be effective against PSW (Hall and Wall 1995). Most are only suitable for topical treatment and exhibit little persistence, requiring repeated treatments until wounds have resolved. Farkas *et al.* (1996) noted that a number of OP compounds have been found effective for the topical treatment of active PSW strikes in sheep and some have also been proposed for prophylaxis. They noted that in Hungary, the most common control method was frequent topical application of OP or pyrethroid insecticides, in particular diazinon. In treating PSW infested wounds with diazinon (0.01%) they observed that treated areas became free of live maggots within 1 day but wound healing was not observed, requiring repeated treatments at 2-3 day intervals due to re-strikes.

Propetamphos is also registered in Australia for flystrike control as a jetting and dipping formulation and as a flystrike and wound dressing. Although cross-resistance exists, propetamphos provides longer periods of protection against flystrike than diazinon (Levot 1990). There appear to have been no studies of the efficacy of propetamphos against OWS, NWS or PWS and inclusion in future tests may be warranted.

***Insect growth regulators.*** As IGRs act only at moulting, take longer to kill all larvae than insecticides with other modes of action and may not prevent pathogenic effects of late instars, they are generally not used for treating existing strikes. In fact, label indications for a number of IGR wool myiasis products are that existing strikes should be treated with a registered flystrike dressing. Thus any role of IGRs against screw-worms will probably be primarily as prophylactics.

Dicyclanil, a pyrimidine derivative closely related to cyromazine, is particularly efficient against early instars of Diptera (Graf, 1999) and has shown *in vitro* activity of more than 10 fold higher than cyromazine or diflubenzuron (Schmid 1999). Dicyclanil is registered for application to sheep, but not to cattle or other animals. A 5% spray-on formulation is registered in Australia with a claim for protection against wool myiasis in sheep for 18-24 weeks (Bowen *et al.* 1999, Nottingham *et al.* 2001). In the Netherlands, prevention of natural blowfly strike for at least 16 weeks was obtained from the application of dicyclanil to lambs (Schmid *et al.* 1999) and Sotiraki *et al.* (2005b) claimed 100% efficacy for 24 weeks against PSW in sheep. In a more extensive field trial, Sotiraki *et al.* (2005a) applied dicyclanil strategically to male and young non-lactating females. Protection of males, which normally have a higher incidence of PSW than females, was 100% for at least 12 weeks while the females were protected for up to 20 weeks.

Dicyclanil also has a registered claim for the protection of mulesing and marking wounds against flystrike in sheep and has also been shown to be effective as a wound myiasis prophylactic in cattle. Direct application of 5% dicyclanil to castration wounds in calves prevented the development of NWS myiasis for 16 days in greater than 93% of cases (Anziani *et al.* 1998). Protection was described as equivalent to that of abamectin. The

incidence of egg masses on treated and control animals was similar, indicating no repellent activity of the dicyclanil.

Diflubenzuron is a relatively stable and persistent IGR and is registered for application to sheep by dipping, jetting and pour-on for the control of wool myiasis and lice infestation and for application to cattle as a pour-on for lice control. Label indications are for protection periods of 12 weeks against wool myiasis, substantiated by experimental studies (Hughes and Levot 1987, Fahy 1994). Diflubenzuron was found to inhibit egg hatch after exposure of NWS females (Crystal 1978) and to affect chitin production in OWS larvae in laboratory tests (Subramanian 2002, 2003) but there is no information available on its effectiveness against OWS in animal challenge situations. A spray-on formulation that contains both dicyclanil (5%) and diflubenzuron (1.5%) (Magic) is registered for application to sheep within 7 days of shearing, for lice and blowfly control. Whether or not the addition of diflubenzuron would enhance the efficacy against OWS in comparison with dicyclanil alone is not clear.

#### **2.4. Chemicals not registered with the AVPMA but reported elsewhere as effective for OWS, NWS or PSW control**

Chemicals within this section can be divided to two groups, those that were previously registered in Australia, but where registration has lapsed or been withdrawn and those that have never been registered for application to production animals in Australia. The first group consists mainly of the organophosphates. While a number of organochlorine insecticides have been shown to be effective against screw-worm (Spradbery *et al.* 1991, Graham 1979, Drummond *et al.* 1988), this group has been prohibited from use in Australia since 1962 and is not considered in this review.

Various formulations, such as aerosols, dusts and sprays of OP insecticides including coumaphos, fenthion, dichlofenthion, fenchlorphos (ronnel), ethion, bromophos plus chlorfenvinphos, crotoxyphos and dioxation have been shown to be effective against SWF but are not currently registered in Australia. In many cases, sprays of these compounds were recommended for SWF control at higher concentrations than the rate previously registered in Australia. In spite of resistance to coumaphos being reported in the early 1980s, it remains one of the more commonly used insecticides for the treatment and prophylaxis of SWF infestations overseas (Hall, 2004). With the high level of efficacy provided by the MLs it is unlikely that any of the OPs currently not available in Australia, with the possible exception of coumaphos, would be re-introduced for screw-worm control.

Injectable ivermectin and doramectin are in the second category and have not been registered for use on sheep in Australia. Doramectin provided full protection for up to 22 days against PSW (Sotiraki *et al.* 2003) and greater than 14 days against NWS following castration of sheep (Sanavria and Prata 1996). Both ivermectin and doramectin have been shown to be effective against OWS and NWS on cattle (see sections 2.1 and 2.3) and are clear candidates for control of OWS in sheep.

Fipronil, first used in Australia as an agricultural chemical in 1994 and as a veterinary chemical in 1995 is available as a spray-on and spot-on for flea control in cats and dogs. Fipronil has been evaluated as a 1% pour-on, and was shown to be effective for the treatment

and prophylaxis of NWS myiasis of post-castration wounds of cattle in Brazil (Lima *et al.* 2004). In their study, greater than 95% of the treated animals were protected from NWS for at least 17 days post-treatment. Most of the oviposition in these trials occurred within 7 days of castration, and the egg masses seen on treated animals suggested that fipronil was not inhibiting oviposition by NWS flies. In addition, Smith *et al.* (2000) examined the *in vitro* insecticidal effects of fipronil on larvae of the blowfly *L. sericata* and concluded that this compound may represent a potential new insecticide for development against sheep blowfly strike. Fipronil is also known to be effective against the cattle ticks *B. microplus* (Davey *et al.* 1998) and *B. annulatus* (Davey *et al.* 1999), the horn fly *Haematobia irritans* (Guglielmone *et al.* 2000) and against fleas (*Ctenocephalides felis felis*) infesting cattle (Araujo *et al.* 1998). A 1% fipronil pour-on formulation afforded protection against larval tick reinfestation for 8 weeks post-treatment (Davey *et al.* 1998). No fipronil based products have been registered in Australia for application to production animals to date. However, fipronil is registered as Topline for pour-on application to beef cattle, although not lactating dairy cows, for the control of *B. microplus*, NWS, *D. hominis* and *H. irritans* in Brazil. With widespread resistance to a range of chemicals present in Australian populations of *B. microplus* there is some likelihood that it could also be registered for use in Australia and fipronil should be considered for inclusion in any future tests against OWS.

Ahmad (2002) reported high efficacy of cyphenothrin in treating OWS infestations in Malaysia. Although a number of cyphenothrin-based products are registered as household insect residual sprays in Australia, no products are available for use on animals.

### **2.5. Other chemicals that might be effective against SWF but have yet to be evaluated**

The focus of insecticidal discovery and development efforts of the major chemical companies tends to change over time. Graf *et al.* (2004) noted that the animal health industry has tended to shift focus towards companion animal products and away from the production animal market. This shift has resulted in a decline in the availability of new chemical groups for large animal ectoparasite control.

More recent insecticides for the pet animal market have included nitenpyram (a nicotinoid), lufenuron (a benzoylurea) and fipronil. The nicotinoids may contain useful insecticides for SWF control and nitenpyram, for example, has been reported as an effective larvicide against NWS infested wounds in dogs (Machado and Rodrigues 2002). The IGRs show an excellent potential for highly specific, targeted management of several important myiasis (Colwell and Dorchie 2004) and Hall and Wall (1995) noted that the systemic insect growth regulators, such as lufenuron, may prove an important future direction for myiasis control. The IGR dicyclanil, reported to be effective against NWS and PSW, has been discussed earlier.

Triflumuron is registered for backline application for control of lice in sheep in Australia. Triflumuron also has larvicidal effects and application by jetting at 0.1% gave protection against wool myiasis for up to 12 weeks (Hopkins *et al.* 1983). However, it has never been registered for this usage. In addition, triflumuron causes ovicidal effects following the exposure of female flies and Smith and Wall (1998) evaluated triflumuron coated targets for *L. sericata* control. Triflumuron is similar in its action to diflubenzuron and could be expected to have similar effects against screw-worm.



Repellents can be considered to consist of compounds with a range of activities including vapour repellents, contact repellents, locomotor stimulants and feeding and oviposition deterrents. Parman *et al.* (1927) tested the repellent action of 353 compounds against NWS and concluded that products obtained from pine trees including pine oil and pine tar were amongst the most effective. Neem has been shown to deter oviposition by *L. cuprina* (Rice *et al.* 1985) and when included in gel formulation with a number of other plant extracts was said to be effective in curing screwworm infestations (Agrawal 1997). However, in most instances repellents are unlikely to be sufficiently persistent or provide an acceptable level of protection to be considered as a practical option. Neither neem nor any of its bioactive extracts is presently registered for animal application in Australia.

### **3. Other aspects of OWS control in Australia**

#### ***3.1. Treatment of companion animals***

Companion animals are also susceptible to SWF attack but little published information is available on treatment or protection. Dogs have been treated by various combinations of larval removal, wound debridement, antibiotics and topical and systemic insecticides. Ivermectin (200-400ug/kg) has been successfully used in combination with topical wound treatments such as dichlofenthion, diazinon and pyrethrins (McNae and Lewis 2004) and household insecticides such as cyfluthrin and transfluthrin (Chemonges-Nielsen 2003). Nitenpyram as a single dose (1-3.5 mg/kg) has been reported to be an efficient larvicide against NWS in dogs (Machado and Rodrigues 2002). Wounds infested with OWS in dogs were reported to have responded well to topical application of neem in combination with eucalyptus oil and extracts of *Cedrus deodara* and *Acorus calamus* (Agrawal 1997).

#### ***3.2. Disinsection***

There is the possibility that areas where inspections are being performed, livestock holding facilities and livestock transport vehicles could be contaminated by larvae evacuating the wounds of infested stock. The current AUSVETPLAN (1996) contains instructions on how to disinsect these areas and there currently seems little reason to modify the instructions presently in place. These instructions state:

"To minimise the likelihood of these larvae burrowing into soil and successfully pupating, as far as possible all inspections and cleaning of transport vehicles should be conducted in yards or washdown areas with concrete or otherwise toughened floors. Because of their maturity, evacuating larvae are more likely to resist the toxic effects of many of the commonly used insecticides/acaricides. Of the chemicals currently registered in Australia, the combination of chlorfenvinphos and cypermethrin (e.g. Barricade S Cattle Dip and Spray, Blockade S Cattle Dip and Spray) has known (significant but not absolute) efficacy for OWS larvae and could be used for spraying livestock transport vehicles and examination areas.

Inspection areas likely to be contaminated with evacuating larvae should be regularly steam cleaned and sprayed with an appropriate insecticide. Faeces and soil deposits in livestock

transports are best treated with pesticide and/or by steam followed by removal by high pressure hosing to destroy any larvae or pupae."

### 3.3. *Insecticide resistance*

Resistance could impact on screw-worm eradication or control programs in two major ways. A strain that entered Australia could already be resistant to one or a number of insecticides or, alternatively, resistance could develop following introduction as the result of the control or eradication strategy invoked.

Of the more than 500 species of arthropods known to be resistant to one or more groups of insecticides, only a small percentage are of veterinary interest (Kunz and Kemp, 1994) and despite widespread chemical use in the control of screw-worms there has been only limited development of resistance. In NWS, Knipling (1942) reported resistance to phenothiazine in a laboratory-selected population. Phenothiazine was a component of smears widely used in early years for control of NWS in the United States. No indication of resistance to lindane, which was used to control in NWS the US for over 10 years, was found in field strains assessed by McDuffie (1960) or Harris *et al.* (1965) despite reports of poor control. Brown and Pal (1971) documenting some 104 cases of resistance in arthropods in their extensive review, did not report any other instances of resistance in screw-worms. Rawlins *et al.* (1983) surveyed NWS populations from various geographical regions for resistance. More than 50 strains were collected from wounds on cattle, sheep and burros at various sites in the US and Mexico as well as single strains from El Salvador and Jamaica. Most of the strains had been exposed to coumaphos in the field and although there was no indication of resistance in two US populations, most other strains exhibited low level resistance with resistance ratios in the range of 5-12X. However, the Jamaican strain had a resistance ratio of 78X. Nolan and Schnitzerling (1986), in an update on resistance in veterinary pests, also noted the presence of resistance to coumaphos in NWS in the Americas. The macrocyclic lactones have been used extensively against a range of parasites in South America for the last 15 years and to date there has been no indication of the development of resistance in screw-worms (Eddi *et al.* 2002).

In OWS, Kunz and Kemp (1994) listed South African strains as resistant to organochlorines (BHC, aldrin and dieldrin), but there are no other reports of resistance. No resistance to any chemical has been found in PSW (Farkas *et al.* 1996). However, it should be noted that *L. cuprina* has a well documented resistance history in Australia with reports of resistance to organochlorines, organophosphates, carbamates and diflubenzuron (Levot 1995) and similar resistance could develop in screw-worms if high selection pressure is applied.

With only limited documentation of any resistance elsewhere it seems that the chance of an introduced strain already having developed resistance to insecticides likely to be used for control is small. If the incursion is only short term, it also seems unlikely that new resistance would have time to emerge. However, all of the currently registered products with a claim against OWS in Australia are based on one ML, ivermectin and it is likely that long acting MLs would play a key prophylactic role in any OWS control program. Relying exclusively on one chemical group would exert strong selection for resistance and could jeopardise the success of any eradication program should resistance occur. Priority should be given to the

testing and registration of insecticides from other chemical groups that may be able to fill a prophylactic role in OWS control or eradication programs.

The impact of widespread chemical use for control of OWS on resistance in other parasites should also be considered. Resistance to chemicals is widespread in ticks, sheep gastrointestinal parasites and sheep blowfly. Although probably not a primary consideration in the event of a screw-worm incursion, chemical options are already limited for a number of parasites and where possible treatments should be chosen that are unlikely to contribute to further selection for resistance in these pests.

### **3.4. Insecticide residues**

Amongst other issues that will require consideration in the choice of a chemical control is the potential for residues in animal products. This will be a particular issue in lactating dairy cattle and will severely limit chemical options. However, dairy cattle are yarded frequently, can readily be inspected and any new infestations treated with non-persistent chemicals. Zeta-cypermethrin ear tags have a nil milk withholding period and may be a particularly useful tool for managing screw-worm in dairy cattle.

Withholding period and time to market will also be an important determinant of chemical choice in some meat production systems. Most meat withholding periods are 6 weeks or less and probably manageable in an incursion situation, although the ivermectin bolus has a 180 day withholding period. Fortunately the systems most sensitive to residues are also generally the most intensively managed, for example feedlot animals. In these systems animals can be more closely monitored and time to market is often well known. Thus insecticide can be chosen to meet the required withholding period, and repeated topical application of low residue products with short protection periods, coupled with regular monitoring, may be a viable control option.

### **3.5. Environmental impact**

The potential for a treatment strategy to impact on the environment may also affect chemical choice in some instances. Control of OWS on organic production enterprises could be particularly problematic. Some chemicals, notably MLs and SPs, and particularly when administered orally or systemically, can affect dung breeding fauna (Wardhaugh *et al.* 2001b, Floate *et al.* 2005). Short term use of these therapeutics is unlikely to affect ecosystems irrevocably and in most cases fauna will recover once treatment stops. However, where slow release formulations are used or extended periods of chemical use is anticipated, the potential for impacts on local ecosystems needs to be considered. Where possible, chemical strategies should be chosen to minimise the impact on dung breeding invertebrates. If the incursion is large, OWS becomes established in Australia, or there are particular local environmental sensitivities, more benign topical application strategies may be required. Notably, there does not appear to be any information available on the likely impact of dicyclanil in dung inhabiting ecosystems.

### 3.6. Other control strategies

As well as the development and evaluation of new chemical groups and associated application techniques for wound myiasis control, other approaches should also be considered.

**Sterile Insect Technique:** This method is considered by most to be a key element in any OWS eradication program in Australia. A series of papers in the Proceedings of the Screw-worm Fly Emergency Preparedness Conference held in Canberra in 2001 (OCVO 2002) describe the technique, its past use and its likely role and operation in event of an OWS incursion and readers are referred to this document and the Screw-worm Fly Disease Strategy within AUSVETPLAN for further information.

**Vaccines:** Vaccination against animal ectoparasites, including myiasis causing flies, while under development for a number of years, has met with variable degrees of success (Kay and Kemp 1994, Willadsen 1999, 2002). The only metazoan vaccine that has been commercially produced in Australia is for the cattle tick, *B. microplus* (Willadsen 2004). Following on from the work on the sheep blowfly, *L. cuprina* (East and Eisemann 1993, Tellam and Bowles 1997), Sukarsih *et al.* (2000) have evaluated a novel form of vaccination against the OWS in sheep and have shown significant *in vitro* and *in vivo* effects on OWS larvae. However, the viability of vaccination as a potential control strategy for OWS remains unresolved and there is little likelihood of a practical vaccine becoming available in the near future.

**Biopesticides:** Resistance development in veterinary ectoparasites has reduced the chemical control options (Casida and Quistad 1998) making biopesticides, such as those based on *Bacillus thuringiensis* (Pinnock 1994) increasingly attractive. Goug *et al.* (2002) evaluated new isolates of *B. thuringiensis* for control of the livestock ectoparasites, *L. cuprina*, *Haematobia exigua* and *Bovicola ovis*. Heath *et al.* (2004) evaluated a number of strains toxic to *L. cuprina* larvae for their ability to prevent experimentally induced fly strike. Results, while variable, demonstrated prevention of fly strike on sheep for 3-6 weeks. In a single field trial, a *B. thuringiensis* preparation provided less protection from naturally acquired fly strike than a commercially available dip. Lyness *et al.* (1994) also demonstrated protection against fly strike due to *L. cuprina* for up to 11 weeks. Activity of *B. thuringiensis* has also been shown against *L. sericata*, *Calliphora stygia* (Chilcott *et al.* 1998), *Chrysomya albiceps*, and *Musca domestica* (Johnson *et al.* 1998). While these and other results are encouraging, with the exception of aquatic applications for the control of mosquitoes and blackflies, no commercial formulations are yet available for control of livestock pests.

Entomopathogenic fungi such as *Metarhizium anisopliae* and *Beauveria bassiana* have also been considered as potential biocontrol agents for veterinary ectoparasites (Samish *et al.* 2004, Butt *et al.* 2001). Wright *et al.* (2004) obtained high levels of fatal infections after exposure of *L. sericata* adults to suspensions of fungal spores and discussed the implications of these results for the biocontrol of blowflies. Entomopathogenic nematodes (Bedding 1983), viruses (Thomson and Bushell 1983) and entomopathogenic fungi (D.M. Leemon pers. comm.) have also been investigated for use against sheep blowflies in Australia. Again no commercial preparations have yet been developed for use on animals.

**Traps:** Fly traps will also play a role in case of a SWF incursion to Australia. A non-sticky trapping system optimised for catching OWS (Urech *et al.* unpublished) is currently used for OWS surveillance in far-northern Australia and around major shipping ports. In the case of an OWS incursion into Australia, the traps would be used to monitor the flies' distribution and to assess the impact of control measures. An improved OWS trap could possibly also be used for population suppression to assist other control or eradication measures (Hall, 1995), similar to LuciTrap for the Australian sheep blowfly (Urech *et al.* 1998).

#### 4. Discussion

Currently the active constituent in all products registered in Australia with a claim against OWS is ivermectin, administered by subcutaneous injection. There are no registered products for sheep. However, there are many products containing a range of chemical classes that are registered for the control of other parasites and that could be utilised in the event of a screw-worm fly incursion. These include products based on OPs, SPs, IGRs, MLs, closantel and spinosad.

In a consideration of chemical control it is important to distinguish those products that are primarily curative in their action and those that provide residual protection. Many of the above-noted chemical classes have shown activity as screw-worm treatments and although most are not currently registered with a claim against OWS, there appears to be a wide range of available options for treating OWS strikes. The organophosphates listed in Table 4 are effective, spinosad a biologically derived pesticide with low mammalian toxicity is an attractive new option and ivermectin applied systemically will kill earlier larval OWS instars although it may not reliably kill older larvae. Ivermectin is registered for topical application to sheep for treatment of flystrike and may be more effective against older stage OWS larvae when applied by this means. Some IGRs may also be effective as wound treatments, but as they only act when larvae moult and may not kill all larvae immediately they are generally less attractive as treatment options.

The list of registered chemicals shown to provide significant residual protection is more restricted and presently limited to MLs, in particular injectable ivermectin, closantel and zeta-cypermethrin formulated in ear tags. These products could play a key prophylactic role in the event of an incursion. Pour-on formulations of eprinomectin and doramectin have also been shown to provide some short term protection and may be useful in some circumstances. However, there are a number of more recently registered ML products that have shown promise against other myiasis species and that should be tested against OWS. Key amongst these are injectable doramectin, which appeared to give better effect than ivermectin in studies with NWS, and long acting formulations of ivermectin. Pour-on formulations of ivermectin have not been tested but are unlikely to give better protection than injectable formulations. However, pour-on formulations are often significantly easier to administer than injectable formulations and testing may be warranted on these grounds. Slow-release bolus formulations of the MLs which can give more than 100 days protection in cattle are registered in New Zealand, although not currently on the market. If approved for use in

Australia they could play a key role in any eradication or containment program. Slow release capsules are registered for endoparasite control in sheep and offer the likelihood of prolonged protection. Testing of these capsules for protection of sheep against OWS would seem worthwhile. Injectable abamectin formulations are available for both sheep and cattle. There is only limited information regarding the efficiency of abamectin against NWS, but these reports are sufficiently positive to warrant its inclusion in future studies with OWS.

Slow release ear tag formulations of zeta-cypermethrin, presently registered in Australia could play an important protective role in cattle. However, in studies to date they did not provide complete protection and their likely usefulness against castration, dehorning or other wounds has been questioned (Wardhaugh and Mahon 2002). It is likely that ear tag formulations will be of most effect when used together with other methods or chemicals as part of an integrated approach. Zeta-cypermethrin tags are the only product known to have substantial prophylactic effect against OWS that are currently registered for use on dairy farms.

Pour-on formulations of SPs have been suggested to give short term prophylactic effect against OWS. In view of the possibility that these products may provide some systemic activity (Wardhaugh and Mahon 2002) they should also be considered for inclusion in future testing with cattle. With sheep the situation is not as clear. Sotiraki *et al.* (2003) suggested that cypermethrin pour-on gave protection against PSW for 12 days and long wool formulations applied to sheep can give protection against wool myiasis for extended periods of time. However, as discussed by Wardhaugh and Mahon (2002), because of the thick wax layer covering the skin of sheep there is unlikely to be much absorption or systemic activity and whether there would be any substantial protection on the non-woolled parts of sheep is questionable.

The IGRs are a group growing in prominence because of their low mammalian toxicity and there are a number of chemicals within this group that could play a role in protecting against OWS. Dicyclanil, registered for control of wool myiasis in Australia, provided prolonged protection against NWS in cattle and long term protection against Wohlfahrtiosis in sheep and is likely to be similarly effective against OWS. Although it cannot be used on milk producing animals and has a 28 day meat withholding period when used on sheep, of those chemicals outside of the ML group it would seem to offer the most promise as an OWS prophylactic.

Although cyromazine can provide protection when applied as a feed-through, it is not registered for this method of administration in Australia for species other than poultry. It also appears that there is no current registration for oral administration in large animals overseas, other than for horses where the label indication is that it should not be used in food producing animals. Applied topically to sheep, cyromazine provides prolonged periods of protection against wool myiasis. However, protection is aided by the ability of the wool grease to 'hold' the insecticide. In addition, *L. cuprina* larvae feed on the skin surface and subcutaneous tissues amongst the wool fibres where they would readily contact insecticide. In contrast OWS tend to create deep wounds where exposure to topically applied insecticide may be more limited. Indications to date are that cyromazine has limited value as a preventative in

cattle and sheep when applied topically at concentrations up to twice the label-recommended jetting concentration (0.2%). However, newer higher concentration (6%) spray-on formulations of cyromazine are now available and may be effective.

Diflubenzuron is a relatively stable and persistent IGR and is known to affect chitin synthesis in OWS. However, systemic action has not been reported and testing in a field situation is required to assess its likely usefulness.

In summary, there is a clear need for the registration, or at least testing of new chemical groups that can be used in the event of a screw-worm fly incursion. The lack of a product with a registered claim for sheep is a particular omission. Although there are a number of chemistries that are effective as wound treatments, products that can give significant periods of prophylaxis, a key consideration in Australia's extensive production systems, are more problematic. Clearly those products already registered or likely to be registered for other animal uses should receive priority for testing as it is unlikely that companies will be willing to undertake the full range of studies and assessments required to register a completely new chemistry just for use in the event of a screw-worm incursion.

## 5. Further testing

Any future testing of SWF control products will need to comply with the WAAVP guidelines provided in Holdsworth *et al.* (2005). These guidelines form part of an ectoparasiticide series dealing with efficacy data generation and will provide the basis for the harmonization of studies performed in various countries. It should also be borne in mind that, although OWS clearly poses the greatest risk, NWS has been previously brought into Australia (Searson *et al.* 1992) and should also be taken into account.

Previous testing has indicated a number of chemicals which are presently registered for use on production animals in Australia and that can provide successful therapeutic treatment of screw-worm myiases in most instances when applied topically. However, Spradbery (2002) makes the point that not all screw-worm infestations are overt and readily accessible to topical applications and treatment with a systemically active insecticide may be required for full resolution. This situation is likely to be more problematic in sheep where covert strikes associated with *L. cuprina* may outnumber overt strikes by as much as 13:1 (Wardhaugh and Dallwitz 1984). Spinosad is a valuable addition to these compounds as it has low mammalian toxicity, has a nil meat withholding period and is permitted for use in most organic production systems. Studies to date have shown good effect against both OWS and NWS (L.B. Lowe pers. comm.) and whether or not further testing will be required awaits the publication and assessment of the work conducted to date.

It is in the area of effective prophylactic approaches that further testing is most clearly required. In the extensive production systems operative through much of Australia intensive inspection and re-treatment is not possible. Effective prophylactic products to prevent infestations and for the treatment of animals in barrier areas for screw-worm containment are essential. There are a number of compounds now registered for use in Australia that could

provide prolonged period of protection and fill a critical prophylactic role but which require testing against OWS. These are listed below in order of priority for testing:

***Injectable doramectin:*** Injectable formulations of doramectin provided superior protection to ivermectin against NWS. Injectable doramectin is registered for treatment of cattle as Dectomax Injectable and should receive priority for testing.

***Capsule and bolus formulations of MLs:*** Ivermectin capsules are registered for control of gastrointestinal helminths in sheep for 100 days and a cattle bolus formulation (registered although not currently marketed in New Zealand and not registered in Australia) gave protection against OWS for in excess of 100 days (Wardhaugh *et al.* 2001a). The sheep capsule should be tested for effectiveness and the feasibility of registration or granting of an emergency permit for the cattle bolus in the event of an incursion should be investigated.

***Long-acting MLs:*** New long-acting formulations of ivermectin have come onto the market and offer the promise of extended protection periods. Long acting formulations of ivermectin and any other MLs that come onto the market should be tested.

***Dicyclanil:*** Although registered for use in sheep in Australia, there is currently no registration for cattle. Dicyclanil provides long term protection against wool myiasis in sheep, has shown good protection against NWS in cattle, appears to have some systemic effect and represents a completely different chemical class to the ML's. Dicyclanil should be tested for use in both sheep and cattle.

***Injectable abamectin:*** Injectable abamectin is registered for application to cattle and sheep in Australia as a number of products, has shown good effect against NWS and should also be tested

***Cyromazine 6% spray:*** Although lower concentration topical applications (0.2%) were ineffective, cyromazine provided in feed did provide protection. The 6% formulation may also provide protection.

***Sheep ML formulations:*** A number of MLs are registered for oral administration to sheep as anthelmintics. These could confer short term systemic protection and should be tested. An oral doramectin drench (Dectomax) is registered, although apparently not presently marketed and should be included in any tests.

***Organophosphates:*** Of the organophosphates currently registered, fenthion and trichlorphon appear to have some systemic action and may fill a practically significant role in the absence of coumaphos. Propetamphos has not been tested against OWS and could provide therapeutic action, but priority for testing is lower than the compounds discussed above.

***Spinosad:*** This chemical provides an attractive option for control of OWS because of its favourable residue profile and potential for use in chemical sensitive situations. Whether or not further testing is required will depend on an assessment of results from studies that have already been conducted.



**Synthetic pyrethroids:** Pour-on formulations of cypermethrin, deltamethrin and alpha cypermethrin may provide some systemic action against OWS. However, priority for testing is lower than for the above mentioned compounds.

**Others:** Of the other products considered, diflubenzuron applied topically may provide some prophylactic effect because of its stability and persistence. Fipronil is a further compound that could be considered if it was to become registered for animal use in Australia. However, fipronil is currently under review by the AVPMA and the likelihood of registration for production animal use is unknown.

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**Table 1. Australian registered parasiticides with known efficacy against OWS**

Active Constituent	Class/ MOA	Formulation	Registered Claim for OWS	Registration Status Protective Period (days)	
				Cattle	Sheep
<b><i>Prophylactic efficacy</i></b>					
ivermectin	ML/ 6	injection	Yes	R 16-20	NR
	ML/ 6	LA injection	No	R NA	NR
	ML/ 6	SR Bolus	No	NR <sup>a</sup> 135	-
	ML/ 6	CR Capsule	No	-	R 100
doramectin	ML/ 6	pour-on	No	R 7-14	NR
closantel	Salicylanilide / NA	oral	No	NR	R 8-15 <sup>b</sup>
zeta-cypermethrin	SP/ 3	ear tag	No	R 150	NR
<b><i>Treatment of existing strikes (little residual protection)</i></b>					
diazinon	OP/ 1B	wound dressing	No	R	R
chlorfenvinphos	OP/ 1B	wound dressing	No	R	R
spinosad	Spinosyn/ 5	wound dressing	No	NR	R
<b><i>Concurrent treatment: acaricides (little residual protection)</i></b>					
Cypermethrin + chlorfenvinphos	SP + OP/ 3 + 1B	dip/spray	No	R	R

MOA: Mode of Action classification, see IRAC website [www.irac-online.org](http://www.irac-online.org) for further detail; ML: macrocyclic lactones; SP: synthetic pyrethroids; OP: organophosphates; R: registered; NR: not registered; NA: not available; <sup>a</sup> Registered in New Zealand, not in Australia; <sup>b</sup> 15mg/kg dose rate

**Table 2: Australian registered products with an OWS claim**

<b>Active Constituent</b>	<b>Dose Rate</b>	<b>Formulation</b>	<b>Meat WP</b>	<b>Milk WP</b>	<b>Manufacturer</b>	<b>Product Code</b>	<b>Product Name</b>
<i>Cattle: Prophylactic products</i>							
ivermectin	200µg/kg bw	injection	28	SL	Merial	46570	Ivomec Antiparasitic Injection
					Eco Animal Health	52528	Ecomectin Antiparasitic Injection
					Pharm Tech	54915	Cevamec Antiparasitic Injection
					Bomac Lab.	56846	Bomectin Antiparasitic Injection
ivermectin + clorsulon	200µg/kg bw	injection	42	SL	Norbrook Lab.	56769	Noromectin Antiparasitic Injection
			28	nil	Merial	45359	Ivomec Plus (Ivermectin plus Clorsulon) Broad Spectrum Injection
			28	SL	Virbac Ancare Aust.	56755 55699	Virbamec Plus Injection Endectocide Genesis Ultra Injection
<i>Cattle: Treatment of screw-worm strikes</i>							Nil
<i>Sheep: Prophylaxis and/or treatment</i>							Nil

WP withholding period (days); SL: see label before using on lactating dairy cattle/sheep; bw: body weight

**Table 3: Products registered in Australia for animal use with known effectiveness against OWS but with no current claim: Prophylaxis**

Active Constituent	Dose Rate	Formulation	Meat WP	Milk WP	Manufacturer	Product Code	Product Name
<b>Cattle:</b>							
doramectin	500µg/kg bw	pour-on	42	SL	Pfizer	49665	Dectomax Pour-on Endectocide
eprinomectin	500µg/kg bw	pour-on	nil	nil	Merial	49105	Ivomec Eprinex Pour-on
ivermectin	1.72g/bolus	bolus	180	180	Merial NZ <sup>a</sup>	A6873	Ivomec SR Bolus
ivermectin	200µg/kg bw	injection	42	SL	Virbac	55727	Virbamec LA Injection
zeta-cypermethrin	10%	Ear tag	nil	nil	Flycam	48148	Y-TEX Python Insecticidal Ear Tags
<b>Sheep:</b>							
closantel <sup>b</sup>	7.5mg/kg bw	oral	28	SL	Western Stock	46165	WSD Closantel Sustained Action
					Virbac	49808	Closicare Sustained Action
					4Farmers	54371	4Farmers Closantel
closantel+albendazole					Schering-Plough	38298	Closal Broad Spectrum Oral
					Norbrook	49707	Caddy Broad Spectrum Oral
closantel+albendazole+levamisole					Jurox	56573	Q-Drench Multi-Combination
closantel+oxfendazole					Virbac	49934	Closicomb Sustained Action Oral
					Virbac	56827	Rotafluke Rotational Flukicide
closantel+selenium					Jurox	48471	Sustain+SE Sustained Action
					Schering-Plough	47055	Seponver SE Sustained Action
					Virbac	50323	Closicare with Selenium
closantel+abamectin	10mg/kg bw	oral	49	SL	Ancare	57844	Genesis Xtra Drench

WP withholding period (days); SL: See label before using on lactating dairy cattle/sheep; bw: body weight;

<sup>a</sup>Registered in New Zealand, not in Australia; <sup>b</sup>15 mg/kg required for efficacy

**Table 4: Products registered in Australia for animal use with known effectiveness against OWS but with no current claim: Treatment of existing strikes**

Active Constituent	Dose Rate	Formulation	Meat WP	Milk WP	Manufacturer	Product Code	Product Name
<b>Cattle/sheep</b>							
diazinon+	1.5%	wound/strike	14-S/3-C	SL	Western Stock	39574	WSD Mulesing Powder
pyrethrins+	0.1%	dressing					
piperonyl butoxide	0.08%		14-S/3-C	SL	Western Stock	39573	WSD Fly Strike Powder
diazinon	2%		14-S/3-C	SL	Coopers	46231	Coopers Fly Strike Powder
chlorfenvinphos +	0.064%	wound	14-S	SL	Western Stock	39575	WSD Aerosol Sheep Dressing
dibutyl phthalate	2%	dressing	14-S	SL	David Grays	42259	Sheep Dressing (Aerosol)
chlorfenvinphos	0.25%	wound	3-S/3-C	SL-S	Fort Dodge	45736	Defiance S Insecticidal Dressing
		dressing		Nil-C			
chlorfenvinphos +	0.055%	spray/dip	8	SL	Fort Dodge	45211	Barricade S Cattle Dip and Spray
cypermethrin	0.01%		8	SL	Coopers	46815	Blockade S Cattle Dip and Spray
spinosad	0.28%	wound	nil-S	SL	Elanco	56734	Extinosad Aerosol
		dressing					
spinosad	0.0125%	wound	nil-S	SL	Elanco	56875	Extinosad Lice, Fly and Maggot Eliminator
		dressing					

WP withholding period (days); S-sheep; C-cattle; SL: See label before using on lactating dairy cattle/sheep

**Table 5: Products registered in Australia for animals use with probable effectiveness against OWS but no current claim**

Active Constituent	Dose Rate	Formulation	Meat WP	Milk WP	Manufacturer	Product Code	Product Name
<b><i>Cattle: Prophylaxis</i></b>							
doramectin	200µg/kg bw	injection	42	SL	Pfizer	46128	Dectomax Injectable
abamectin	200µg/kg bw	injection	30	SL	Virbac	47652	Virbamec Injection - Cattle
			30	SL	Youngs	51817	Vetmec - Cattle Injection
			30	SL	Novartis	48506	Rycomectin - Cattle Injection
			42	SL	Ancare	49703	Genesis Injection Abamectin
			30	SL	Dover	49917	Paramectin
abamectin+ cyanocobalamin		injection	49	SL	Ancare	56315	Genesis Injection Abamectin Plus Vit B12
<b><i>Cattle: Treatment</i></b>							
chlorfenvinphos	0.4%	spray	nil	nil	Fort Dodge	45594	Supona
fenthion	200g/L	spot-on	10	SL	Bayer	33520	Tiguvon Spot-On
deltamethrin	15g/L	pour-on	nil	nil	Intervet	55098	Arrest Easy-Dose
			nil	nil	Fort Dodge	55308	Bombard
			nil	nil	Coopers	54096	Easy-Dose
			nil	nil	Virbac	55679	Deltamethrin Pour-On
	25g/L	pour-on	nil	nil	Coopers	33468	Coopafly
deltamethrin + ethion	0.005% 0.025%	dip/spray	nil	SL	Coopers	45981	Tixafly
cypermethrin	0.095%	spray	3	SL	Novartis	52995	Cypafly
zeta-cypermethrin	25g/L	pour-on	nil	nil	Elanco	50781	Demize
permethrin	8.7%	pour-on	7	(SL) SL	Y-Tek	56165	Brute Pour-On For Cattle

WP withholding period (days); S-sheep; C-cattle; SL: See label before using on lactating dairy cattle/sheep

Table 5 (continued)

Active Constituent	Dose Rate	Formulation	Meat WP	Milk WP	Manufacturer	Product Code	Product Name
<i>Sheep: Prophylaxis</i>							
cyromazine	6%	spray	7	SL	Ancare	52729	Cyrazin Spray On
			7	SL	Jurox	56440	Cyro-Fly 60
			7	SL	Norbrook	52745	Venus Spray-On
			7	SL	Novartis	39979	Vetrazin Spray-On
			7	SL	Youngs	58519	Cyromazine Spray-On
dicyclanil	5%	spray	28	SL	Novartis	50005	Clik Spray
dicyclanil + diflubenzuron	5%	spray	21	SL	Novartis	58306	Magik Spray
ivermectin	160mg/cap	capsule	126	SL	Merial	48688	Ivomec Maximizer (Adult Sheep 40-80kg)
			80mg/cap	126	SL	Merial	48689
abamectin	200µg/kg bw	injection	35	SL	Ancare	49703	Genesis Injection Abamectin
abamectin+ cyanocobalamin	200µg/kg bw	injection	35	SL	Ancare	56315	Genesis Injection Abamectin Plus Vit B12
<i>Sheep: Treatment</i>							
alpha-cypermethrin	5%	spray	nil	SL	Coopers	38354	Vanquish Long Wool
cypermethrin	25g/L	pour-on	3	SL	4Farmers	54370	Cypermethrin 25
			3	SL	Virbac	39538	Cypercare
			nil	SL	Fort Dodge	47845	Outflank
			3	SL	WSD	39065	Spurt
deltamethrin	10g/L	backline	3	SL	Coopers	36340	Clout-S

WP withholding period (days); S-sheep; C-cattle; SL: See label before using on lactating dairy cattle/sheep

**Table 6: Results from South American trials in which macrocyclic lactones were tested against New World screw-worm (not listed in Eddi *et al.* 2002)**

Active Constituent (Reference)	Country	Infestation	Myiasis type	Efficacy
Ivermectin (Moya-Borja <i>et al.</i> 1997)	Brazil	Experimental 120 larvae / animal in 4 wounds	Wounds infested 2 hours post- treatment	29% of wounds infested (cf 100% control for doramectin)
Ivermectin (Benitez Usher <i>et al.</i> 1997)	Argentina Brazil	Natural 8 trials	Navel <sup>1</sup> Scrotal <sup>2</sup> Examined daily for 14 days	Navel: 87.5-100% Scrotal <sup>3</sup> : 100% Scrotal <sup>4</sup> : 96.4%
Ivermectin <sup>5</sup> (Anziani <i>et al.</i> 2000)	Argentina	Experimental 100 larvae	Wound challenge 12 and 15 days post-treatment	d 12: 27-36%, d 15: 8-17%
Doramectin (Anziani <i>et al.</i> 2000)	Argentina	Experimental 100 larvae	Exp'tal wounds Challenge 12 and 15 days post-treatment	d 12: 91% d 15: 83%
Ivermectin (Caproni <i>et al.</i> 1998)	Brazil	Natural 91 trials, 2718 cattle	Castrated 10 days post-treatment. Examined 3 and 7 d post-castration	43.7% (0-100%) >90% in 3% trials
Doramectin (Caproni <i>et al.</i> 1998)	Brazil	As above	As above	94.6% (53.3-100%) >90% in 80% trials

<sup>1</sup> treated within 24 h of birth; <sup>2</sup> treated immediately post-castration;

<sup>3</sup> 2 month old calves; <sup>4</sup> 4 months or older calves; <sup>5</sup> long acting formulations;