The European Agency for the Evaluation of Medicinal Products
Veterinary Medicines and Inspections

EMEA/MRL/838/02-FINAL
May 2002

COMMITTEE FOR VETERINARY MEDICINAL PRODUCTS

ABAMECTIN
(Revision of the acceptable daily intake)

SUMMARY REPORT (4)

1. Abamectin is a fermentation product produced by the soil actinomycete, *Streptomyces avermitilis*. Abamectin consists of a mixture of avermectin B1a (at least 80%) and avermectin B1b (not more than 20%). It is an endectocide with a broad spectrum of activity against nematode and arthropod parasites of animals and plants. In veterinary medicine it is administered to cattle as a single subcutaneous injection of 0.2 mg/kg bw and as a single oral dose of 0.2 mg/kg bw abamectin to sheep, for the treatment of gastro-intestinal nematodes, lungworms and nasal bots.

   Abamectin is not used in human medicine.

   A toxicological ADI of 0.25 µg/kg bw (i.e. 15 µg/person) was previously established by the Committee for Veterinary Medicinal Products (CVMP) by applying a safety factor of 200 to the NOEL of 0.05 mg/kg bw/day in a study in CF-1 mice, based on maternotoxicity.

   Currently, abamectin is included in Annex I of Council Regulation (EEC) No 2377/90 as follows:

Pharmacologically active substance(s)	Marker residue	Animal species	MRLs	Target tissues	Other provisions
Abamectin	Avermectin B1a	Bovine	20 µg/kg 10 µg/kg	Liver Fat	

   Abamectin was previously included in Annex III of Council Regulation (EEC) No 2377/90 as follows:

Pharmacologically active substance(s)	Marker residue	Animal species	MRLs	Target tissues	Other provisions
Abamectin	Avermectin B1a	Ovine	20 µg/kg 50 µg/kg 25 µg/kg 20 µg/kg	Muscle Fat Liver Kidney	Provisional MRLs expire on 1.1.2001

   The CVMP recommended the inclusion of abamectin for sheep in Annex I of Council Regulation (EEC) No 2377/90. This recommendation has not yet been adopted by the Commission.

2. In April 2001 the European Commission requested that the CVMP re-consider the ADI that was established 1993 in light of additional data that was made available to the WHO/FAO Joint Meeting on Pesticide Residues (JMPR) in 1997. The Commission also asked questions related to whether account was taken of the dual use of abamectin in veterinary medicines and pesticides in setting the MRLs. The data that formed the basis of the JMPR decision were made available to the CVMP in February 2002 and have now been assessed.

7 Westferry Circus, Canary Wharf, London, E14 4HB, UK
Tel. (44-20) 74 18 84 00 Fax (44-20) 74 18 84 47
E-mail: mail@emea.eu.int http://www.emea.eu.int

3. In the original assessment of abamectin, the following NOELs were identified: chronic toxicity in the mouse: 4 mg/kg bw; repeated dose toxicity in the rat: 1.5 mg/kg bw; repeated dose toxicity in beagle dogs: 0.25 mg/kg bw; reproductive toxicity in the rat: 0.12 mg/kg bw; maternotoxicity in developmental toxicity studies in CF-1 mice: 0.05 mg/kg bw. The last NOEL was identified as the basis for the ADI by CVMP and JMPR.

4. The JMPR re-evaluation was based on data indicating that CF-1 mice showed a genetically pre-disposed sensitivity to the effects of avermectins, and neonatal rodents showed hypersensitivity relating to development of the blood-brain barrier that was atypical of primates including humans. These effects were related to P-glycoprotein expression in the small intestine and brain capillary endothelium.

5. A study of tissue distribution of the 8,9-Z isomer of avermectin B_{1a} in CF-1 mice genotyped for P-glycoprotein expression indicated that levels of the isomer were 60 times higher in the brains of -/- mice than +/+ mice and also higher in +/- mice than +/+ mice. Uptake of the 8,9-Z isomer by CF-1 foetuses was highest in -/- foetuses, intermediate in +/- foetuses and lowest in +/+ foetuses.

   P-glycoprotein was only observed in placentas of mice with a + allele. Levels of P-glycoprotein in the brains of day 20 foetuses and neonates were found to increase with time reaching levels, as a percentage of adult values, of l9%, 37% and 89% on days 14, 17 and 20 *post partum*, respectively. Studies from human and monkey foetuses indicated expression of P-glycoprotein occurs prenatally in primates, with comparable levels of P-glycoprotein mRNA in 28-week old human foetuses as adults.

6. Acute toxicity studies in CF-1 mice indicated that abamectin was about 2 times less toxic in +/+ mice (LD_{50} 28 mg/kg bw) than +/- mice (14 mg/kg bw) and 5-10 times less toxic than previous values in -/- mice (0.3-0.4 mg/kg bw). The oral LD_{50} of the 8,9-Z isomer was 217 mg/kg bw in female CD-1 mice and about 20 mg/kg bw in male CF-1 mice.

7. In a four day repeated oral dose study, abamectin was severely toxic to 12/50 female and 5/49 male CF-1 mice at a dose of 0.8 mg/kg. Insensitive CF-1 mice tolerated doses of up to 10 mg/kg bw and no toxicity was observed at this dose in CD-1 mice. P-glycoprotein was observed in the brains and small intestines of the insensitive CF-1 and CD-1 mice, but only in small amounts in only one of the sensitive CF-1s.

8. A multigeneration reproduction study was conducted in rats using ivermectin. Effects on pup mortality were observed at doses greater than or equal to 0.4 mg/kg bw. In a cross-fostering study increased mortality and decreased bodyweight was observed only in pups reared by treated dams, and not those reared by untreated dams. Metabolic studies indicated that uptake of ivermectin and distribution in the brain, liver and carcass during the first 10 days *post partum* decreased, particularly from days 6 to 10.

9. In a developmental toxicity study, groups of CF-1 mice were given daily oral doses of 0.015 to 0.06 mg/kg bw/day 8,9-Z avermectin B_1 through days 6 to 15 of gestation. No treatment-related effects were observed in dams or foetuses. In a second study, using doses of 0.015 to 0.5 mg/kg bw/day, maternotoxicity was observed at 0.5 mg/kg bw/day and increased incidences of cleft palate in foetuses at greater than or equal to 0.1 mg/kg bw/day. The NOELs were 0.1 and 0.03 mg/kg bw/day for maternal toxicity and teratogenicity, respectively.

10. Female CF-1 mice were tested for sensitivity to abamectin toxicity, and then groups of sensitive and insensitive animals were mated and dosed orally with the 8,9-Z isomer from days 6 to 15 of gestation. P-glycoprotein was found in the brains of the insensitive, but not the sensitive dams. Only one of the treated sensitive dams, dosed with up to 1 mg/kg bw had live foetuses at termination, cleft palate was observed in 45% of these, compared to none in the corresponding controls. In the groups of insensitive dams treated with 0.5 to 1.5 mg/kg bw/day, a dose-related increase in cleft palate was seen; this was not considered unexpected as the dams would have included individuals +/+ and +/- for P-glycoprotein, so all three genotypes would have been possible in the foetuses. No significant maternal or teratogenic effects were observed in CD-1 mice dosed from days 6 to 15 of gestation with 0.75 to 3 mg/kg bw/day 8,9-Z isomer. All adults

were +/+ for P-glycoprotein. Groups of adult male and female CF-1 were genotyped for P-glycoprotein expression, mated and the dams were dosed with 1.5 mg/kg bw/day 8,9-Z isomer from days 6 to 15 of gestation. The incidence of foetuses with cleft palate in the +/- x +/-, and -/- x -/- control groups were 0.83% and 0%. In the treated groups, incidences of 0%, 12% and 58% were seen in the +/+ x +/+, +/- x +/+ and +/- x -/- groups respectively. No cleft palates were seen in +/+ treated foetuses the incidence in -/- and +/- treated foetuses was 30 out of 31 and 29 out of 70.

11. Abamectin is not used in human medicine. Ivermectin has been administered to humans for treatment of parasitic diseases. Around fifty million doses, generally about 150 μg/kg bw have been given worldwide, primarily for treatment of onchocerciasis. Adverse reactions are reported to be generally mild and transient involving fever, pruritus, arthralgia, myalgia, asthenia, postural hypotension, tachycardia, lymphadenopathy, gastro-intestinal effects, sore throat cough and headache. Ocular inflammation, somnolence, eosinophilia and raised hepatic enzymes may occur. These effects are consistent with a Mazzotti reaction arising from the effect of ivermectin on the microfilariae.

12. The revised ADI established by JMPR in 1997 was based on the NOEL of 0.12 mg/kg bw from the rat reproduction study, with a reduced uncertainty factor of 50 to account for neonatal hypersusceptibility in this species, and the NOEL from the one year dog study of 0.24 mg/kg bw with an uncertainty factor of 100. The ADI value was rounded down to 2 μg/kg bw (150 μg/person, for a 60 kg adult).

13. Information has recently become available that polymorphisms of the *MDRI* gene exist within the human population; these appear to include functional variations that result in over-expression of P-glycoprotein as well as under-expression. The data that are currently available suggest that the 10-fold safety factor for inter-individual variability within the overall 100-fold standard uncertainty factor used to establish ADIs is sufficient to account for any increased sensitivity to abamectin as a result of these polymorphisms. However should other variations resulting in greater effects of P-glycoprotein expression be identified in future, it may be necessary to revise this value accordingly.

Conclusions and recommendation

Having considered that:

- Sensitivity to avermectin toxicity is linked to the expression of P-glycoprotein that plays a critical role in the development of the blood-brain barrier.
- CF-1 mice show mutations that result in deficient/absent P-glycoprotein in the endothelial cells of brain capillaries and sensitivity to avermectin toxicity shows a direct link to P-glycoprotein genotype. Similar mutations have not been reported in CD-1 mice, rats or non-human primates.
- The data available on P-glycoprotein expression polymorphisms in humans indicates that a 10-fold factor for inter-individual variability is sufficient to account for any resultant increased sensitivity to abamectin.
- In humans, and primates development of the blood brain barrier occurs relatively early during the prenatal period, whereas in rodents development occurs postnatally. Cross-fostering studies in rats indicate that neonatal toxicity from avermectins appears to be linked to exposure during this critical postnatal period, rather than prenatal exposure.
- In CF-1 mice the P-glycoprotein genotype not only determines sensitivity to avermectin toxicity, but also appears to indicate susceptibility to teratogenic effects such as cleft palate.
- The NOEL of 0.05 mg/kg bw for the developmental toxicity study in CF-1 mice is not directly relevant for human risk assessment due to the genetic predisposition of this strain to avermectin toxicity. The NOEL for of 0.12 from a two-generation reproduction study in the rat is also considered an unsuitable basis for setting an ADI, as there is evidence that neonatal rats are hypersusceptible to avermectin toxicity.

the Committee for Veterinary Medicinal Products recommends that a revised ADI for abamectin should be established based on the NOEL of 0.25 mg/kg bw from the one-year repeated dose study in dogs. Using an uncertainty factor of 100, this would give a value of 2.5 μg/kg bw (150 μg/person, for a 60 kg adult).

Based on the existing MRLs, it was calculated that the consumer intake of total residues from the consumption of bovine and ovine tissues would account for approximately 7 and 10% of the ADI, respectively. These values would allow adequate provision to accommodate intakes from pesticidal uses that are not expected to exceed around 5 μg/person. Consequently, total maximum theoretical intakes from both veterinary and pesticidal uses should not exceed around 13% of the revised ADI.