



COMMITTEE FOR VETERINARY MEDICINAL PRODUCTS

TILMICOSIN (extension to chicken)

SUMMARY REPORT (1)

1. Tilmicosin is a macrolide antibiotic synthesized from tylosin. It has an antibacterial spectrum similar to tylosin with enhanced activity against *Pasteurella multocida* and *Pasteurella haemolytica*.
2. Tilmicosin has been evaluated by CVMP for its use in cattle (calves only), pigs and sheep. A microbiological ADI of 0.004 mg/kg bw was established (240 µg/day for a 60 kg person), based on a NOEL of 0.4 mg/kg bw in an *in vivo* study with HFA rats, and a safety factor of 100.

Tilmicosin is entered into Annex I of Council Regulation (EEC) No 2377/90 for cattle, pigs and sheep in accordance with the following table:

Pharmacologically active substance(s)	Marker residue	Animal species	MRLs	Target tissues	Other provisions
Tilmicosin	Tilmicosin	Bovine, ovine, porcine	1000 µg/kg 50 µg/kg	Liver, kidney Muscle, fat	
		Ovine	50 µg/l	Milk	

3. An application has now been submitted for extension of the annex entry to include chicken (broilers only). For the use of tilmicosin in poultry, tilmicosin is indicated for the treatment and control of respiratory diseases associated with mycoplasma and other organisms susceptible to tilmicosin in broiler chickens at a dose of 75 mg/l of drinking water for 3 days (equivalent with approximately 15 mg tilmicosin/kg bw/day). Tilmicosin is not indicated for laying hens.
4. After treatment of broilers with tilmicosin at the recommended dosage of 75 mg/l drinking water for 3 days, a peak plasma level of 10 µg/ml was reached after 84 hours, while peak concentrations in lung and airsac (2.3 and 3.29 µg/ml, respectively) were reached earlier at 48 hours. The latter indicates that tilmicosin is accumulated well in these tissues.
5. After oral treatment of broiler chickens with radiolabelled tilmicosin at doses of 25-450 mg/l in drinking water for 3-5 days (equal to 4.4-75.1 mg/kg bw/day) radioactivity was mainly distributed to liver and kidney, and to a lesser extent to muscle and fat. High levels of radioactivity were also found in bile, indicating a substantial biliary excretion. This finding is consistent with findings in cattle, pigs and sheep.
6. Metabolite identification revealed that the parent compound was the main residue in tissues, excreta and bile, followed by metabolites T-1, T-3 and T-4. Hence, tilmicosin is partly desmethylated (yielding T-1), hydroxylated (yielding T-3) or reduced and sulfated (yielding T-4). This metabolic pathway is also observed in rats, cattle, pigs and sheep. Another metabolic pathway in chickens is reductive glycosidic cleavage of the mycinose sugar moiety, yielding metabolites T-9 and T-10.

7. After administration of tilmicosin to broiler chickens at the recommended dosage, the highest total radioactive residues were observed in liver (6.6 mg/kg at day 3, declining to 0.4 mg/kg at day 14), and to a somewhat lesser extent in kidneys (1.6 mg/kg at day 3, declining to 0.4 mg/kg at day 14). Relatively low total residues were found in muscle and fat (less than 0.2 mg/kg at day 3, declining to below 0.02 mg/kg at day 14). Although the quantitative residue composition varied with time, in all tissues at all time points the parent compound was the main component, followed by T-1, and (in kidney only) T-9 and T-10.
8. Comparable amounts of parent compound were found in a cold residue study with broiler chickens treated with tilmicosin at the recommended dosage. Highest residues were found in liver: 2.6 mg/kg at day 3, declining to 0.13 mg/kg at day 17. Residue levels in kidney averaged 0.65 mg/kg at day 3 and declined via 0.08 mg/kg on day 10 to non-detectable levels (less than 0.06 mg/kg) thereafter. Residues in muscle, fat and skin were approximately 0.10 mg/kg at day 3 and non-detectable (less than 0.014 mg/kg) from day 14 on.
9. Information on the microbiological activity of the metabolites indicates the following activity ratios: T-1 equals 25% of parent tilmicosin, T-3 is non-active, T-9 equals 400% of parent tilmicosin, and T-10 equals 100% of parent tilmicosin.
10. Based on the information on the microbiological activity ratios of the metabolites of tilmicosin compared to parent tilmicosin and the residue distribution at approximately 10 days after cessation of treatment it is possible to express the activity of the metabolites as tilmicosin equivalents. This leads to the following mean ratios of parent tilmicosin versus total microbiological active residues: liver 0.9; kidney 0.15. The radioactivity at this point was no longer quantifiable in muscle and skin + fat, hence the ratio marker to total microbiologically active residues is no longer relevant for these tissues.
11. Tilmicosin is considered to be the most suitable marker residue. For the calculation of the intake of tilmicosin residues of animal origin, all uses of the compound should be taken into account, i.e. the uses in chicken, cattle, sheep and pigs. As tilmicosin is also used in lactating ewes, part of the ADI (75 µg/day; MRL for sheep milk (50 µg/l) x milk consumption (1.5 l/day)) is already occupied by intake of residues via milk from sheep. This leaves 165 µg per day for residues in liver, kidney, muscle and skin + fat from treated animals.
12. Using the data from the cold residue study, the intake of residues in organs and tissues from chickens is below the remainder of the ADI of 165 µg at approximately 10 days after cessation of treatment. The residue distribution of tilmicosin at this time point is 12 : 3 : 1 : 1 for liver : kidney : muscle : skin + fat.
13. A new HPLC-method for the determination of tilmicosin residues in edible tissues of chickens, pigs and cattle was provided. The method is described in an acceptable format. However, the method is not fully validated with respect to specificity: the possible interference of tilmicosin related substances such as metabolites and other veterinary drugs is not investigated.

Conclusions and recommendation

Having considered that:

- the microbiological ADI is 4 µg/kg bw (i.e. 240 µg per 60 kg person),
- tilmicosin is the marker residue,
- the tissue distribution indicates highest and most persistent microbiologically active residues in liver and kidney, and to a much lesser extent muscle and skin + fat,
- at 10 days withdrawal, the ratio marker residue to total microbiologically active residues is 90% in liver and 15% in kidney and that in muscle and skin + fat residues were not longer detectable at this time point,
- although tilmicosin is not the major residue in chicken kidney and the ratio tilmicosin to total microbiologically active residues is only 15% at later time points, this tissue constitutes only a small portion (10 g) of the daily food package,
- the proposed routine analytical method for the determination of tilmicosin in chicken tissues is well described but not yet fully validated,

the Committee recommends the inclusion of tilmicosin for chicken in Annex III of Council Regulation (EEC) No. 2377/90 in accordance with the following table:

Pharmacologically active substance(s)	Marker residue	Animal species	MRLs	Target tissues	Other provisions
Tilmicosin	Tilmicosin	Chicken	75 µg/kg 75 µg/kg 1000 µg/kg 250 µg/kg	Muscle Skin + fat Liver Kidney	Provisional MRLs expire on 1.1.2000 Not for use in animals from which eggs are produced for human consumption

LIST OF QUESTIONS

1. The applicant should provide additional data concerning the proposed routine analytical method for the determination of tilmicosin residues in edible tissues of chickens. The method should be fully validated with respect to the possible interference of tilmicosin related substances, such as metabolites, and other veterinary drugs.