Pharmacokinetics of tilmicosin in serum and milk of goats

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SUMMARY

Tilmicosin was administered to goats intravenously and subcutaneously to determine its concentration in blood and milk and its kinetic behaviour. After a slow intravenous injection, the serum concentration-time curve indicated a two compartment open model with a mean (SEM) elimination half-life ($t_{1/2\alpha}$) of 4.36 (0.04) hours. After a subcutaneous injection the drug was eliminated more slowly from serum and milk, with $t_{1/2\beta}$ of 29.3 and 41.4 hours, respectively. The apparent volume of distribution of tilmicosin was more than 1 litre kg$^{-1}$. The peak serum tilmicosin concentration was 1.56 µg ml$^{-1}$ 6.39 hours after a subcutaneous injection of 10 mg kg$^{-1}$. Tilmicosin was extensively secreted into milk, reaching a maximum concentration of 11.6 µg ml$^{-1}$ and having a large $AUC_{milk}/AUC_{serum}$ ratio of approximately 12:1. Tilmicosin was detectable in milk for 11 days after a single subcutaneous dose.

The poor response of Staphylococcus aureus udder infections to intramammary antibiotic therapy is a major problem for veterinarians and dairy farmers and is of considerable interest to mastitis researchers. It is attributed to the restricted penetration of antibiotics into areas of scarring and inflammation, the partial inactivation of antibiotics by milk, intracellular or metabolically active organisms, resistance to antibiotics, and improper treatment procedures (Shem-Tov et al. 1994). The macrolide antibiotics and the fluoroquinolones appear to be suitable for systemic dry cow therapy because they have long half-lives and good penetration, and are transferred effectively from blood into milk. Tilmicosin is a novel macrolide antibiotic, designed to have a high level of activity against Pasteurella species while retaining good efficacy against Mycoplasma species and Gram-positive organisms (Walters et al. 1994). It has been approved for the treatment of respiratory infections in cattle and for dry cow therapy (Shem-Tov et al. 1994). The pharmacokinetics and persistence of tilmicosin in milk have been investigated in sheep by Parker et al. (1994a,b), but similar studies do not appear to have been made in goats. The objectives of the present study were to investigate the disposition, distribution pattern and residual content of tilmicosin in the milk of goats after subcutaneous and intravenous dosing.

MATERIALS AND METHODS

Tilmicosin

The drug (Micotil injection; Elanco Animal Health; England) contains 300 mg tilmicosin ml$^{-1}$.

Animals

Five clinically healthy, non-pregnant milking female goats (60 days post partum), weighing 18 to 20 kg and aged two-and-a-half to three years, were used. During acclimatisation for three weeks, and the subsequent treatment periods, they were fed alfalfa with drinking water freely available. The animals were kept in individual metabolic cages in an enclosed room. They were shorn over a jugular vein to facilitate the collection of blood samples.

Drug administration

Tilmicosin solution (300 mg ml$^{-1}$) was diluted in saline to 2.5 mg ml$^{-1}$ and the diluted solution was administered to each of the five goats by slow intravenous infusion into the left jugular vein for 13.33 minutes at the rate of 0.75 mg kg$^{-1}$ min$^{-1}$, a total dose of 10 mg kg$^{-1}$. Three weeks later, each of the five goats was given a single dose of 10 mg kg$^{-1}$ of tilmicosin subcutaneously in the left dorsolateral chest wall. All five goats received the doses of drug on the same day.

Blood samples

Blood samples were obtained from the catheterised right jugular vein. To maintain the catheters patent a sterilised metal stylet was used. Blood samples were collected immediately before and at 0.25, 0.5, one, two, four, eight, 12, 24, 48, 72, 96 and 120 hours after the intravenous infusion began. After the subcutaneous injections blood samples were collected at the same intervals. The blood was centrifuged at 3000 rpm for 15 minutes and the serum was used for the estimation of tilmicosin concentration. The serum samples were stored at −80°C before analysis, and the assay was performed within seven days.

Milk samples

Milk was collected from the five animals at two, four, eight, 23, 30, 47 and 72 hours after the subcutaneous dose of the drug, and the daily milk was pooled and sampled on the following 12 days. The milk samples were stored at −80°C before being analysed for tilmicosin within seven days.

Analytical methods

The free tilmicosin concentrations in serum and milk were measured by a microbiological assay technique (Bennett et al. 1966) using Bacillus subtilis (ATCC 663) as the test organism. The limits of detection of tilmicosin in serum and milk were 5 and 10 ng ml$^{-1}$, respectively.
Pharmacokinetic analysis

The concentrations of tilmicosin in serum and milk were subjected to kinetic analysis, and the pharmacokinetic parameters were calculated for each animal by the method described by Baggot (1978). Five points were used to determine the terminal slope. The mean half-lives were calculated as harmonic means. The results are given as mean (SEM).

RESULTS

The slow intravenous infusion of a dilute solution of tilmicosin resulted in clinical signs suggesting acute cardiac toxicity. The goats were clinically normal within 25 minutes of the end of the infusion, however, and no side effects were observed after the subcutaneous injection.

Values for the kinetic parameters describing the disposition of the drug are given in Table 1. After the intravenous injection of the drug at a dose of 10 mg kg⁻¹, its concentration decreased in a biexponential manner that could be described by a two-compartment open model. The drug was rapidly distributed and slowly eliminated with mean half-lives of 0.27 and 4.36 hours for the distribution and elimination phases. The volume of distribution was more than 1 litre kg⁻¹, indicating good penetration into tissues. The total body clearance of the drug was 0.79 (0.02) litre kg⁻¹ h⁻¹.

The mean peak concentration of tilmicosin in serum (Cmax = 1.56 (0.04) µg ml⁻¹) was reached 6.39 hours after the subcutaneous injection. Table 2 and Fig 1, show that the drug was absorbed slowly from the injection site, as indicated by its long absorption half-life (t1/2αb = 0.16 (0.02) hours). The mean elimination half-life (t1/2β) for tilmicosin after the subcutaneous injection was 29.3 (1.53) hours, indicating that the drug was eliminated slowly.

TABLE 1: Mean (SEM) pharmacokinetic parameters of tilmicosin in five goats after a slow intravenous injection of 10 mg kg⁻¹ bodyweight

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Mean (SEM)</th>
</tr>
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<tbody>
<tr>
<td>Cmax (µg ml⁻¹)</td>
<td>10.5 (0.12)</td>
</tr>
<tr>
<td>A (µg ml⁻¹)</td>
<td>9.04 (0.09)</td>
</tr>
<tr>
<td>a (h⁻¹)</td>
<td>2.55 (0.01)</td>
</tr>
<tr>
<td>t1/2αb (h)</td>
<td>0.27 (0.001)</td>
</tr>
<tr>
<td>B (µg ml⁻¹)</td>
<td>1.46 (0.04)</td>
</tr>
<tr>
<td>β (h⁻¹)</td>
<td>0.16 (0.002)</td>
</tr>
<tr>
<td>t1/2β (h)</td>
<td>4.36 (0.04)</td>
</tr>
<tr>
<td>Vdss (litre kg⁻¹)</td>
<td>2.27 (0.02)</td>
</tr>
<tr>
<td>Vd(area) (htre kg⁻¹)</td>
<td>4.95 (1.00)</td>
</tr>
<tr>
<td>CI(B) (litre kg⁻¹ h⁻¹)</td>
<td>0.79 (0.02)</td>
</tr>
</tbody>
</table>

The mean concentrations of tilmicosin in milk after a subcutaneous injection of 10 mg kg⁻¹ are shown in Fig 1. The maximum concentration (11.6 ± 0.13 µg ml⁻¹) occurred 5-66 hours after dosing. The drug was slowly eliminated from the milk with an apparent half-life of 41.4 hours (Table 2).

DISCUSSION

The results of this study show that the serum concentration of tilmicosin in goats remained above the minimum inhibitory concentrations (MICs) for the most sensitive bacteria (A pyogenes and S aureus) isolated from cattle, which range from 0.04 to 0.78 µg ml⁻¹ (Shem-Tov et al, 1994), for two days after a slow intravenous or single subcutaneous injection. The subcutaneous administration of tilmicosin at 10 mg kg⁻¹ resulted in free drug concentrations in the milk which were above the MIC of the drug for S aureus and A pyogenes for three and eight days, respectively. Some preliminary field efficacy data (Soback et al 1990) suggest that this duration is associated with high cure rates. Similar results have been reported by Shem-Tov et al (1994) who found that free tilmicosin concentrations in the udder of cows remained above the MIC of the drug for S aureus for seven-and-a-half days after a subcutaneous injection.

After intravenous dosing the plasma concentration data were best fitted to a two-compartment pharmacokinetic model. The slow intravenous infusion of a dilute solution of tilmicosin resulted in clinical signs suggesting acute cardiac toxicity but these side-effects were not observed after the subcutaneous injection. These clinical signs were also reported in cattle (Shem-Tov et al 1994). As a result the intravenous route of administration is clearly unsuitable for goats, but the subcutaneous route was found to be safe.

The apparent volume of distribution at steady-state of a drug (Vdss) is an indication of its diffusion into body tissues (Gilman et al 1980). The mean Vdss value of tilmicosin in goats was 2.27 litre kg⁻¹, twice that of 1.11 litre kg⁻¹, reported in cows (Shem-Tov et al 1994). This difference may be attributable to the difference in the extent of protein binding or the different methods of calculation used. The higher values of Vdss, however, indicate that tilmicosin was widely distributed in the extravascular tissues. In sheep, Parker et al (1994b) found that the half-life of tilmicosin in the lungs (26.9 hours) was similar to the half-life in serum.
The subcutaneous injection of tilmicosin appeared likely to be more useful than an intravenous injection because the serum concentrations of the drug were sustained for a longer period. After the subcutaneous injection, the peak concentrations of tilmicosin in serum and milk (1.56 and 11.6 μg ml⁻¹, respectively) were reached after 6.39 and 5.66 hours, respectively). Similar results were reported by Shem-Tov et al (1994) who found that after a subcutaneous injection of tilmicosin in cows, the maximum concentration in milk was 8.21 μg ml⁻¹. After a subcutaneous injection, the mean apparent elimination half-lives of tilmicosin in serum and milk were 29.3 and 41.4 hours, respectively. A similar half-life (42.4 hours) was reported in the milk of cows by Shem-Tov et al (1994) and in the serum of sheep (29.6 hours) by Parker et al (1994b). Tilmicosin reached the milk rapidly after subcutaneous dosing, maintained a concentrations above 1 μg ml⁻¹ for three days, reached a high maximum concentration, had a large AUCₘᵢ𝑙ₖ/AUCₜₛᵉʳᵘₜ ratio and was eliminated from the udder slowly. These results were consistent with those previously reported by Shem-Tov et al (1994) in cows. In goats, the subcutaneous injection of tilmicosin prolonged the effective concentrations of the drug in the udder, and this may be helpful in the treatment of mastitis.

REFERENCES

BAGGOT, J. D. (1978) Some aspects of clinical pharmacokinetics in veterinary medicine i. Journal of Veterinary Pharmacology and Therapeutics 1, 5-18


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