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Rosoxacin Distribution in Kidney and Prostate: Experimental Studies in Dogs*

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Summary. Concentrations of rosoxacin, a new quinolone derivative, in the renal hilar lymph, renal interstitial fluid, prostatic interstitial fluid, and prostatic secretion were investigated in dogs during constant intravenous infusion. The lymph was obtained by direct cannulation of the lymphatics and the interstitial fluid from small plastic tissue chambers implanted six weeks before the experiments. Renal clearance of rosoxacin was compared to the glomerular filtration rate, as measured by ^{125}I-iothalamate clearance, and showed a net reabsorption of rosoxacin. The renal lymph and interstitial fluid concentrations in both kidney and prostate were found to be lower than the simultaneous plasma concentrations, corresponding well with earlier findings for several other antimicrobial agents. However the concentrations in lymph and interstitial fluids were higher than the minimum inhibitory concentrations for most enterobacteriaceae normally encountered in urinary tract infections.

Key words: Rosoxacin - Renal lymph - Interstitial fluid - Kidney - Prostate.

INTRODUCTION

Rosoxacin is a new quinolone derivative structurally related to nalidixic acid and oxolinic acid. It is effective in vitro against a wide variety of gram-negative organisms with minimum inhibitory concentrations (MICs) five-to-ten times lower than those of nalidixic acid (6). The antibacterial spectrum of rosoxacin includes Pseudomonas aeruginosa, many strains of which are sensitive to concentrations easily achieved in both blood and urine of humans (6). Rosoxacin is a weak acid with a pKa of 8.5 and good stability at physiological pH ranges. The drug is moderately lipid soluble and preliminary studies have shown rosoxacin to be 75-80% protein bound in human serum (6). Since this new drug is of great potential interest in the treatment of urinary tract infections, we investigated the distribution of rosoxacin in the kidney and the prostate, two organs often involved in these infections. In order to ascertain the drug concentrations at the actual site of infection, i.e. the interstitial tissue, we employed two different techniques: cannulation of hilar lymphatics in the kidneys, and implantation of multiperforated tissue chambers in the prostate and the kidneys from which interstitial fluid can be obtained.

MATERIALS AND METHODS

Renal Study. Three adult mongrel dogs (26-28 kg) were anaesthetised with sodium thiopental, and through a midline abdominal incision, one 16 x 6 mm multiperforated, polyethylene tissue chamber with connective tubings was implanted in the upper and one in the lower pole of the left kidney. Approximately four weeks later, assuming the tissue chambers were healed in completely, the dogs were again anaesthetised as above. Through a flank incision, the left kidney was exposed and the ureter cannulated in order to obtain urine. By careful dissection, a hilar lymph vessel was exposed and cannulated with a 26-gauge needle connected to a silastic tube (0.3 mm i.d., 0.6 mm o.d., Dow Corning). Every 30 minutes, 0.3 to 1.0 ml of lymph could be collected in micro test tubes (Eppendorf); one drop

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(1/40 ml) of 5000 U/ml heparin was then added. From the connected tubes, 0.02 to 0.1 ml of renal interstitial fluid could be collected every 30 minutes. One drop of heparin, as above, was added to each sample. In order to maintain a constant drug concentration in the blood, a bolus injection of 10 mg/kg rosoxacin was given, followed by a constant intravenous infusion of 3 mg/kg per hour for four hours. ^{125}I-iothalamate was administered simultaneously (0.5 μCi as priming dose, and 0.3 μCi/min in saline as sustaining infusion dose) in order to estimate the glomerular filtration rate (GFR).

Samples of lymph, chamber fluid, and urine were obtained every 30 minutes, while plasma samples were drawn in the middle of each collecting period from the cannulated femoral artery exposed by a femoral cutdown. At the end of each experiment the kidney was removed and tissue concentrations in cortex and medulla were estimated.

Prostate Study. Four male dogs (30-33 kg) were anaesthetised with sodium thiopental intravenously. Through a low midline abdominal incision, one plastic chamber as described above was implanted in each lateral lobe of the prostate. One month later, the studies were carried out.

The dogs were again anaesthetised and prostatic secretion was obtained via the catheterized urethra. To prevent urine contamination, a low midline abdominal incision was made and the urethra was ligated at the bladder neck. Urine was obtained by a cystostomy. The tubings connected to the implanted tissue chambers were exposed to collect the interstitial fluid. Rosoxacin was administered by a constant infusion technique as described in the renal study. Samples of urine, prostatic secretion (PS), and prostatic interstitial fluid (PIF) from the tissue chambers were obtained before and at 30, 60, 120, 180, and 240 minutes following the bolus injection. Again, plasma samples were obtained from the exposed femoral artery (femoral cutdown) in the middle of each collecting period. Pilocarpine (0.1 mg/kg) was given intravenously before administration of the antibiotic in order to stimulate prostatic secretion. At the end of the experiment, three of the dogs were sacrificed and rosoxacin concentrations were determined in various tissue samples.

Assay Methods. ^{125}I-iothalamate samples were counted in an automatic gamma scintillation counter (Autogamma Scintillation Spectrometer, Packard 5220) at the proper window setting. Radioactivity in lymph and interstitial fluid samples was determined by saturating a paper disc (Sterile Blanks®, 1/4", Difco Laboratories, Detroit, MI) with the fluid in question and counting directly on the disc. In previous studies we have estimated the volume of the discs to be 0.021 ± 0.001 ml (± SE) (3). Rosoxacin concentrations were determined using a disc diffusion method on seed agar with Bacillus subtilis ATCC 6633 as the test organism. Phosphate buffered saline was used as the reference fluid in the preparation of standard curves for the bioassay of rosoxacin in urine, lymph, and interstitial fluid, while standard curves for plasma were generated from pooled dog plasma. Statistical methods employed were paired and independent t tests.

RESULTS

Kidneys. Rosoxacin concentrations in renal hilar lymph (L) attained a constant level after only 30 minutes, but a constant level in renal interstitial fluid (RIF) took more than 60 minutes. Assuming that the concentrations hereafter were time independent during constant infusion, the mean values for RIF and L in all three dogs were 8.07 ± 0.74 μg/ml and 2.21 ± 1.93 μg/ml (± 1 SE), respectively. These values are shown in figure 1 as the L/plasma (PL) and RIF/PL ratios. The concentration of rosoxacin in PL averaged 11.89 ± 0.52 μg/ml. The L/PL ratios (Mean .75 ± .04) were significantly higher than the interstitial fluid/PL ratios (0.20 ± 0.05) ($p < 0.001$). The clearance of rosoxacin was calculated from the formula: $\frac{U \times V}{P}$ (U = urine concentration, V = urine volume, and P = plasma concentration). The clearance/GFR ratios (GFR = iothalamate clearance) are shown in Figure 1. The average clearance of rosoxacin elimination in dog kidneys is characterised by tubular reabsorption. The ratios between L/PL for rosoxacin and L/Pl for iothalamate, and the ratios between RIF/PL for rosoxacin and RIF/PL for iothalamate were not significantly different from unity ($p = 0.86$ and $p = 0.99$, respectively) (Fig. 1).

Prostate. Of the eight implanted chambers, seven were functioning, giving a satisfactory amount of 0.03 to 0.1 ml every 30 minutes. The pH in prostatic secretion was determined using pH paper (Fisher Scientific Co., New Jersey) and averaged 6.5. Figure 2 shows the rosoxacin concentrations in PL, PS/PL ratio, and PIF/PL ratios, which (in all four dogs) averaged 0.09 ± 0.01 and 0.30 ± 0.05, respectively. The mean concentrations for PS and PIF in all four dogs were 1.09 ± 0.17 μg/ml and 2.62 ± 0.31 μg/ml, respectively. The PIF concentrations of rosoxacin were as an average about twice the concentrations in PS, but never exceeded the corresponding plasma concentrations. Table 1 outlines the tissue concentrations of rosoxacin in the prostate, testes, epi-

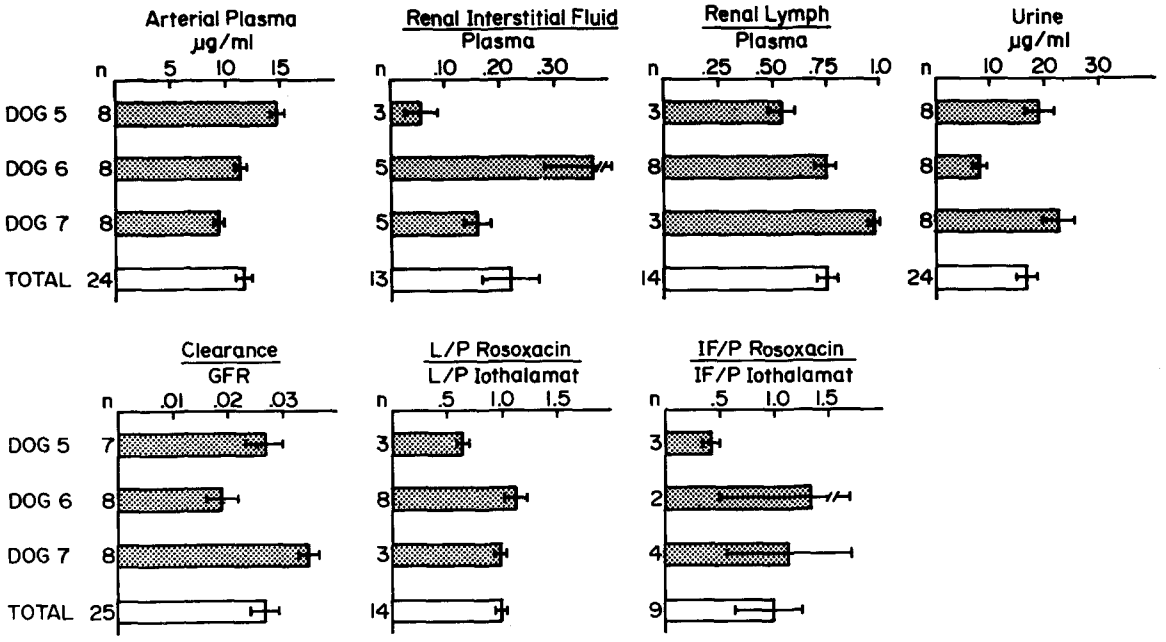

Fig. 1: Urine and arterial plasma concentrations together with renal interstitial fluid to arterial plasma concentration ratios and renal lymph to arterial plasma concentration ratios of rosoxacin. Lymph/plasma ratio of rosoxacin to lymph/plasma ratio of iothalamate and renal interstitial fluid/plasma ratio of rosoxacin to renal interstitial fluid/plasma ratio of iothalamate as compared to clearance of rosoxacin/clearance of iothalamate ratio. n = number of collecting periods and numbers of total experiments, respectively

Table 1. Tissue concentrations (µg/g, mean ± 1 SE) of rosoxacin following constant infusion for four hours

	Rosoxacin		
	Concentration	Ration Tissue/plasma	n
Prostate	3.90 ± 0.70	0.38 ± 0.13	3
Testis	4.97 ± 2.57	0.42 ± 0.25	3
Epididymis	7.30 ± 4.35	0.70 ± 0.42	3
Kidney (Cortex)	33.0 ±10.00	2.74 ± 1.39	2
Kidney (Medulla)	68.5 ±41.5	6.09 ± 4.50	2

didymis, and kidney. The tissue plasma ratios were calculated from the average of the last three plasma samples obtained before sacrificing the animals. The prostatic tissue/plasma ratio corresponded with the PIF/ plasma ratio, but the tissue concentrations in kidney cortex and medulla were much higher than the concentrations found in plasma, lymph or renal interstitial fluid.

DISCUSSION

The plasma concentrations in the dogs in both the prostate and the kidney study correlate with the peak plasma concentrations obtained in humans after an oral dose of 500 mg of rosoxacin (6). The concentrations in prostatic interstitial fluid, renal lymph and interstitial fluid were all higher than the minimum inhibitory concentrations (MICs) of most enterobacteriaceae normally encountered in urinary tract infections. The concentrations of rosoxacin in the prostatic secretion were lower or only in the range of the MICs of these bacteria. In addition, plasma and urine concentrations of rosoxacin were higher than the MICs for many strains of Pseudomonas aeruginosa.

Our results in the renal lymph, RIF, and PIF correspond with earlier findings (3-5). Cockett et al. (1), after single-dose administration to dogs, found constantly lower concentrations in renal hilar lymph of nalidixic acid than in corresponding plasma samples. According to Naber et al. (4), the renal hilar lymph concentrations of antibiotics do not correlate with protein binding but with the mode of excretion of the drug. Theoretically this corresponded with our resutls. In spite of the fairly high protein binding, rosoxacin produced the same concentrations in lymph as iothalamate (Fig. 2). As previously shown for trimethoprim, rosoxacin concentration in renal interstitial fluid was significantly lower than the corresponding concentrations in renal lymph (3). This discrepancy

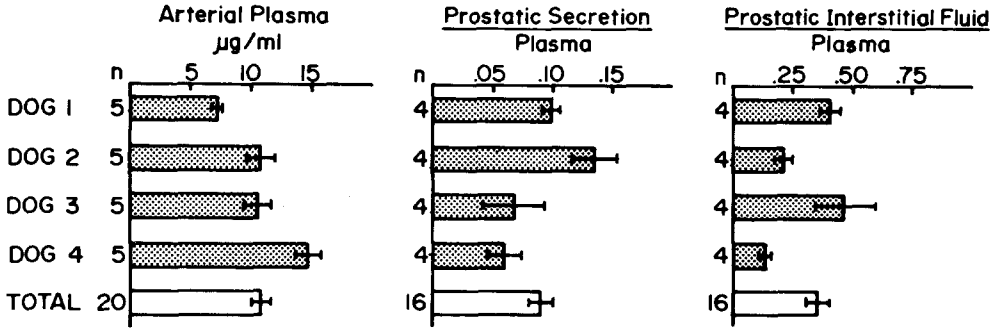

Fig. 2: Prostatic secretion to arterial plasma concentration and prostatic interstitial fluid to arterial plasma concentration ratios of rosoxacin. n = numbers of collecting periods and number of total experiments, respectively

may be explained by the difference in production site of the two fluids. The greater part of the tissue chambers was positioned in the cortex of the kidneys, while the hilar lymph constitutes interstitial fluid produced both in the cortex and medulla. Since tubular reabsorption may influence the hilar lymph concentrations, this could explain the higher concentrations in lymph than in the renal interstitial fluid.

The high concentration of rosoxacin in the kidney may indicate that tissue binding occurs since these high concentrations are not reflected in the lymph or in the interstitial fluid (Table 1).

The concentration of rosoxacin in prostatic secretion and in PIF were as expected from a weak acid. While bases diffuse readily into the prostatic secretion resulting in higher concentrations here and in the interstitial fluid than the correspondong plasma concentrations, acids penetrate poorly into the prostate (2). However, the concentrations in the PIF were high enough to predict a beneficial effect in treatment of lower urinary tract infection, where the prostatic gland may be involved.

In conclusion, it appears that rosoxacin produces effective concentrations both in the renal and prostatic interstitium as well as in the plasma and urine. Because of its good activity against common urinary pathogens, including Pseudomonas aeruginosa, the drug deserves further attention, especially with regard to treatment of urinary tract infections.

REFERENCES

1. Cockett, A.T.K., Moore, R.S., Roberts, A. P.: Distribution of some newer antibiotics within the kidney interstitium: a therapeutic consideration in pyelonephritis. Investigative Urology 5, 250 (1967)
2. Frimodt-Möller, N., Maigaard, S., Madsen, P.O., Naber, K.G.: Co-trimazine distribution in the canine prostate. Infection 7, (Suppl. 4) 345 (1979)
3. Maigaard, S., Frimodt-Möller, N., Naber, K.G., Madsen, P.O.: Renal lymph- and interstitial fluid concentration of co-trimazine: an experimental study in dogs. Infection 7, (Suppl. 4) 349 (1979)
4. Naber, K.G., Madsen, P.O.: Renal lymph concentrations of cephalosporins: the importance of protein binding and renal excretion. Infection 4, (Suppl. 2) 131 (1976)
5. Naber, K.G., Madsen, P.O., Maroske, D., Bichler, K.-H.: Antibiotics and chemotherapeutics in renal lymph. An experimental study in dogs. Investigative Urology 14, 23 (1976)
6. Sterling-Winthrop Research Institute. A Summary of Laboratory Data on WIN 35213. Rensselaer, New York: Sterling-Winthrop Research Institute 1972

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