Diffusion of cloxacillin into synovial tissue

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After a 30 min i.v. infusion of 1 g cloxacillin, the concentrations of this antibiotic were measured in plasma and synovial tissue samples from 11 patients undergoing total hip replacement. Assuming passive distribution between plasma and tissue the rate constants of distribution were estimated. The mean half-life of distribution was 22 min. The concentration of free drug in synovial tissue was estimated to be 77% of the total tissue concentration. The maximum tissue drug concentration after an i.v. bolus dose is predicted to occur at about 37 min.

Keywords cloxacillin synovial tissue antibiotics

Introduction

Cloxacillin is a narrow-spectrum antistaphylococcal antibiotic used for perioperative prophylaxis of wound infections, particularly following joint replacement procedures. The antibiotic is administered some time before the operation to ensure sufficient concentrations in the tissues at the time of anticipated infection (Bergamini & Polk, 1989). Since little is known about the rate of diffusion of the antibiotic between plasma and tissues it is difficult to estimate the optimal timing of antibiotic administration. The purpose of the present study was to calculate the rates of diffusion of cloxacillin between plasma and synovial tissue to devise a rational dosing schedule.

Cloxacillin (1 g) (Beecham Farma B.V., Amstelveen, The Netherlands) was administered by continuous intravenous infusion over 30 min. The duration of the infusion was exactly 30 min in five patients; in the other six patients it was slightly longer (up to 35 min; five patients) or shorter (29 min; one patient). Blood samples were collected in heparinized tubes at 0, 10, 30 and 60 min after discontinuation of the infusion and then at hourly intervals until the end of the operation. Synovial tissue specimens were taken as soon as the joint capsule was opened, between 31 min and 2 h 19 min after the start of the infusion; all samples were taken after the end of the infusion period.

Antibiotic assay

Blood samples were centrifuged for 10 min at 1500 g. An equal volume of acetonitrile was added to 250 µl of the supernatant to precipitate plasma protein. The solution was centrifuged for 5 min at 1500 g, and 3 ml dichloromethane were added to the supernatant. After centrifugation for 5 min at 1500 g, the cloxacillin concentration was measured in 20 µl of the supernatant by reversed phase high-performance liquid chromatography, using a Spectroflow 400 constant-flow pump (Kratos Analytical Instruments, Ramsey, NJ, USA) and a Kratos Spectroflow 773 variable-wavelength u.v.-detector at a wavelength of 220 nm (Kullberg *et al.*, 1991). The lowest concentration which yielded measurable peaks was about 0.2 mg l⁻¹; the coefficient of variation of the assay at this concentration was 8.2% ($n = 10$). Tissue samples were rinsed with sterile saline, blotted dry, weighed and homogenized in a tissue grinder (type X-1020, Ystral GmbH, Dottingen,

Methods

Patients

Eleven patients (two male, nine female, ranging in age from 55 to 80 years) undergoing an elective hip replacement procedure because of osteoarthritis entered the study. According to the guidelines of the Hospital these patients received 24 h antibiotic prophylaxis using cloxacillin and kanamycin. The aim of the study and the procedure were explained to the patients before informed consent was given. The Hospital Review Committee approved the study.

Experimental procedure

All patients received the usual i.v. dose of kanamycin (7 mg kg⁻¹) preoperatively, together with cloxacillin.

FRG) with 2 ml phosphate buffer. The tissue homogenate was centrifuged for 10 min at 1500 g and the supernatant was processed as described above.

All samples were assayed within 2 h of collection. In a subgroup of eight patients plasma protein binding was determined by equilibrium dialysis using a Dianorm dialysis apparatus (Diachema A.G., Zürich, Switzerland), consisting of two 0.22 ml dialysis chambers separated by a 2 cm² cellulose membrane (MWC 5000). One chamber contained 0.20 ml plasma and the other a solution of cloxacillin in saline at various concentrations. The chambers were placed in a rotator (Scientific Ind. Inc., Springfield, Mass., USA) and dialysis was carried out at 12 rev min⁻¹ at 37° C until equilibrium was reached (about 4 h). Cloxacillin concentrations in the two chambers were measured as described above.

Pharmacokinetic analysis

It was assumed that distribution of drug from plasma to tissue and disappearance of the drug from tissue were proportional to its concentration in plasma and tissue, respectively (Kunst & Mattie, 1978; Mattie *et al.*, 1987a). Thus

$$dC_T/dt = k_p C - k_T C_T \quad (1)$$

where dC_T/dt is the rate of change of the tissue drug concentration, C_T and C are the tissue and plasma drug concentrations, respectively, and k_p and k_T are first order rate constants describing the distribution from plasma to tissue and disappearance from tissue, respectively. If it is assumed that the rate constant for the distribution of unbound drug from plasma to tissue (ku) is equal to the rate constant for the distribution of extracellular unbound drug from tissue to plasma, equation 1 can be written as follows:

$$d(fu_T \cdot C_T)/dt = ku \cdot fu \cdot C - ku \cdot fu_T \cdot C_T \quad (2)$$

where fu and fu_T are the unbound fractions in plasma and tissue, respectively. Division of both sides of equation (2) by fu_T and combining equation (1) gives:

$$ku = k_T \quad (3)$$

$$fu_T = fu \cdot k_T / k_p \quad (4)$$

The disposition of drug was described by a mono-exponential function. Accordingly the plasma drug concentration after the infusion period is given by equation (5):

$$C = C(\tau) \cdot e^{-k \cdot t} \quad (5)$$

where $C(\tau)$ is the plasma drug concentration at the end of the infusion, k the elimination rate constant and t the time since the end of the infusion.

Substituting equation 5 into equation 1 and integrating gives:

$$C_T = \frac{k_p C(\tau)_p}{k_T - k} (e^{-k \cdot t} - e^{-k_T t}) + C_T(\tau) e^{-k_T t} \quad (6)$$

where $C_T(\tau)$ is the tissue drug concentration at the end of the infusion.

The plasma drug concentration at the termination of the infusion is given by:

$$\frac{R_o}{k \cdot V} (1 - e^{-k\tau}) \quad (7)$$

where τ is the duration of the infusion. The infusion rate, R_o , is the dose divided by τ .

Substitution of equation 7 into equation 1 and integrating gives:

$$C_T(\tau) = \frac{k_p R_o}{k_T k V} \left(\frac{1 - k_T e^{-k\tau} - k e^{-k_T \tau}}{k_T - k} \right) \quad (8)$$

Values of the pharmacokinetic constants in equations 5 and 7 were calculated for the individual data sets using the curve fitting procedure NLIN of the SAS computer program package (SAS Institute Inc., SAS/STAT® Users Guide, version 6, fourth Edition, vol. 2, Cary, NC: SAS Institute Inc; 1989). Substitution of these values into equation 6, in which equations 7 and 8 have been substituted for $C(\tau)$ and $C_T(\tau)$, respectively, yields an equation for tissue drug content with two unknowns (k_p and k_T). Using the NLIN procedure average estimates of k_p and k_T for all patients, and their respective standard errors, could be calculated such that the sum of the squares of the differences between C_T according to equation 6 and the observed values of C_T was minimized (Mattie *et al.*, 1987b).

Results

Figure 1 illustrates the wide range of drug concentrations in both plasma and tissue. The calculated estimates of the rate constants k_p and k_T in equation 6 were 0.34 h⁻¹ (s.e. mean 0.11; 95% CI: 0.90 and 0.59) and 1.88 h⁻¹ (s.e. mean 0.86; 95% CI: 0.07 and 3.83), respectively. The value of k_T , representing the rate constant for the distribution of free drug between plasma and tissue and *vice versa*, corresponds to a distribution half life of 22 min. The best estimate of the free extracellular concentration, expressed as a fraction of the total tissue concentration (fu_T in equation 4), was 0.77.

By substituting the mean calculated values of V (13.08 l) and k (1.33 h⁻¹) into equation 7, in which τ is replaced by the time after the start of the infusion, and in equation 5, the average concentration-time profile for cloxacillin in plasma during and after the infusion could be calculated. By substituting the same mean values, together with the mean values of the distribution constants, k_p and k_T , into equations 6 and 8 the average concentration-time profile for cloxacillin in tissue could be determined. The results of these calculations are shown in Figure 2.

The mean extent of plasma protein binding of cloxacillin was 85.8% (s.d. 1.9%). Over the drug concentration range of 10 to 100 mg l⁻¹ the free fraction was constant.

Figure 1 Concentrations of cloxacillin in plasma (a) and synovial tissue (b) from 11 patients during and after a 30 min infusion of 1 g cloxacillin. Time 0 is the start of the infusion.

Figure 2 Predicted average cloxacillin concentrations in plasma and synovial tissue, based on the analysis of data shown in Figure 1.

Discussion

For our group of eleven patients the global value of the distribution rate constant of free cloxacillin (k_T) was 1.88 h^{-1} . The value of 0.77 for fu_T is much lower than that for various other beta-lactam antibiotics in murine muscular tissue (Kunst & Mattie, 1978; Mattie *et al.*, 1987a) indicating that, unlike those agents, cloxacillin is not only present in extracellular water but also binds to tissue components.

The calculated parameters of plasma and tissue pharmacokinetics can be used to predict concentrations after other modes of administration. For instance, it can be predicted that after an i.v. bolus injection of 1 g cloxacillin the maximum synovial tissue concentration will be reached at 37 min after administration. At this time the estimated free drug concentrations in plasma and tissue should be 0.9 mg l^{-1} . Since both plasma and synovial tissue drug concentrations subsequently decline, it is advisable to administer the cloxacillin within 30 min of the start of the surgical procedure.

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