

## Tissue concentrations: do we ever learn?

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Over the last decades, numerous papers have appeared—and still are appearing—that describe concentrations in tissues in an effort to predict the efficacy of an antimicrobial agent based on these concentrations and MICs for microorganisms. A common method is to use measurements of concentrations in tissue homogenates, comparing these with values derived from the corresponding blood samples and on that basis draw conclusions with respect to the potential clinical use of the drug. This approach is not justifiable for a number of reasons that includes both pharmacokinetic as well as pharmacodynamic causes. This way of presenting data with the derived conclusions is often misleading and may ultimately be harmful in patient care.

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#### Introduction

Over the last decades, studies have been performed that describe whole tissue concentrations of antimicrobials derived from homogenates. In a number of these the authors subsequently use the results to draw conclusions towards the activity and efficacy of the drug. <sup>1-6</sup> Measuring concentrations in tissue homogenates, comparing them with the corresponding blood samples and recommending the drug for clinical use based on such pharmacokinetic data, is not justifiable as has been shown more then 25 years ago. <sup>7</sup> We consider this way of presenting data with its derived conclusions misleading and, because erroneous conclusions may be drawn with respect to the activity of the drug, also potentially harmful in patient care.

In the following, we briefly provide the scientific basis for our arguments and show a number of examples to underscore our point of view based on current knowledge on concentrations at the site of infection.

### What do tissue concentrations represent?

Whole tissue concentrations are usually obtained by grinding up (homogenizing) or lysing tissue and subsequently determining the concentration of the antibiotic in the tissue homogenate. However, this does not take into account the fact that tissues are

made of distinct compartments (interstitial fluid, cells, and within cells the various subcellular organelles) in which the drug is not necessarily distributed in a homogeneous fashion. Moreover, measurements in tissue homogenates do not give any information on whether the drug is actually available for activity. Thus, when whole tissue concentrations are determined by measuring the overall concentration in the tissue homogenate, the concentrations found are not informative with respect to the (active) concentration of the antimicrobial at the site of infection.<sup>7–9</sup> Since most bacterial infections are located in the extracellular compartment, those concentrations are of primary interest. If a compound is distributed mainly extracellularly, such as β-lactams and aminoglycosides, grinding up the tissue means dilution of the drug by mixing intracellular and extracellular fluids, resulting in underestimation of its concentrations at the site of infection. Conversely, if drugs are accumulated by cells (such as fluoroquinolones or macrolides), assay of total tissue levels will lead to gross overestimation of the extracellular concentration. The opposite is true for intracellular infections.

# Pharmacokinetic relationships between serum and tissue concentrations

After administration, an antibiotic (as any drug) becomes distributed into various compartments in the body. The pharmacokinetics

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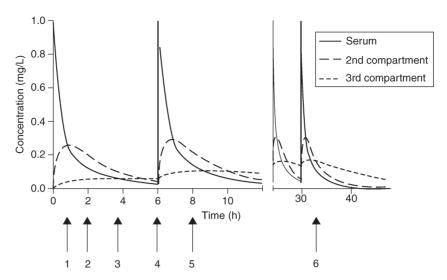


Figure 1. Schematic figure showing the concentration—time course in three locations: serum, extracellular and intracellular. The six arrows indicate sample time-points in each of the three locations; the concentrations were used to determine the ratios in Table 1.

of most antibiotics can be described by either a two or a three compartment model, and the amount of drug transferred between compartments is described by intercompartmental clearance. The equilibrium between the compartments does not occur instantaneously and the shape of the concentration-time curve in either compartment can therefore be markedly different. 10 The reasons for the differences are delayed steady-state, time-lags to and from other compartments as documented in numerous studies, including those where concentrations have been determined in specific compartments.8,11-15 Depending on the time a whole tissue sample is taken (in most situations only once), any value of a ratio of tissue concentration versus serum concentration can be obtained. Figure 1 and Table 1 illustrate this by showing the concentrationtime curves in serum and two tissue compartments and the ratio of serum to tissue concentration in these two compartments. To circumvent the problem, multiple samples would have to be taken in a steady-state situation using an appropriate technique in a single patient, which is often unpractical and even ethically questionable. Alternatively, samples could be taken at multiple time-points from different individuals, in both cases directly from the specified compartment, but this would introduce a large possibility for errors due to interindividual variabilities.

**Table 1.** Extracellular and intracellular concentration/serum concentration ratios as a function of sample drawing time

	Extracellular concentration/ serum concentration ratio	
0.85	1.00	0.15
2	1.58	0.45
3.8	1.60	1.00
6	1.40	1.87
8	1.59	0.78
32	1.57	1.21

#### Pharmacokinetic/pharmacodynamic relationships

Pharmacokinetic/pharmacodynamic relationships have been described for a number of drugs, using serum concentrations as a surrogate for the concentrations at the actual site of infection. These relationships show a marked consistency, and the pharmacodynamic index values (such as AUC/MIC) that result in a certain effect have been determined for most classes of antibiotics. The pharmacokinetic/pharmacodynamic index values are derived from concentrations measured in serum over time and it is inappropriate to infer similar effects from tissue concentrations or local concentrations. Statements such as 'concentrations in tissue *x* h after dosing are much higher than the MICs for common pathogens<sup>2,4</sup> that cause disease' are meaningless.

The concentrations may have been lower or higher in relation to the serum concentrations, and the values of the denominator are therefore incorrect, while the drug exposure over time is not included either. If inferences are drawn from local concentrations, the concentration–effect relationships at the actual infection site should be used and these are usually unknown.

#### When are tissue concentrations of value?

It is very difficult to determine the concentration of a drug at the site of the infection and/or at the receptor site. This is one of the reasons that the surrogate values in serum are being used. Although the results of using these surrogates are remarkably consistent, aberrations do occur, for instance unexpected failure or success of treatment. In those cases, concentrations at the site of infection, using a validated method, could certainly be of value. Measurements in specific compartments (such as epithelial lining fluid and CSF) or determining free concentrations in interstitial fluid by microdialysis contribute to our understanding of concentrations at the site of infection. It should be borne in mind, however, that all these methods may suffer from methodological problems that are not always immediately apparent. Finally, whole tissue concentrations can be of value in

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initial studies during drug development to determine the overall distribution of a drug.

#### **Conclusions**

We conclude that, comparable to the expressions used by both the European Medicines Agency<sup>19</sup> as well as the ASM,<sup>20</sup> the use and determination of tissue concentrations derived from homogenates to draw direct conclusions on drug activity are unwarranted and/or unreliable. 21,22 Statements that infer activity of the drug based on concentrations measured in relation to the susceptibility (MIC) of the agent should be avoided. Alternatively, measurements of concentrations in specified compartments may yield information that is much needed to understand the efficacy of an antimicrobial but should be supplemented with effective measures of activity and/or clinical evidence. In that respect, the use of unbound serum concentrations has proved to be useful as a surrogate correlate.<sup>23</sup> Using concentrations in tissue-homogenates alone to determine exposure-response relationships and drawing conclusions with respect to clinical use are unjustified and may ultimately be harmful to patients.

#### **Transparency declarations**

None to declare.

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