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Short Communication

Dissolution – Bioequivalence Non-Correlations

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The World Health Organization (WHO) rules (1996) recommend in chapter 3 "Technical data for regulatory assessment" that information for marketing authorization should contain among others in-documentation, equivalence data (comparative bioavailability, pharmacodynamic or clinical studies), and comparative *in vitro* dissolution tests. European rules concerning evaluation of bioavailability and bioequivalence (CPMP/EWP/QWP/1401/98) specify in chapter 5.2: "Dissolution studies are always necessary and consequently required. *In vitro* dissolution testing forms a part of the assessment of a bioequivalence waiver". The consequence is that over the last years all over Europe dissolution studies were connected to bioequivalence studies and the natural tendency was to correlate the results obtained in the pair studies in order to obtain models allowing dissolution results as predictor for *in vivo* results.

Let us consider only as a mnemotechnique, the living body as a "mathematical operator" which transforms parameters characterizing dissolution curves in pharmacokinetic parameters associated to plasma levels curves.

$$\psi \; (dissolution) = pharmacokinetics$$

If we consider the two spaces as ordered metric spaces, a natural question is that the operator preserves the distances and order. For example, if we compare a tested drug T with a reference one R, we would be interested to know if

$$d_{in\ vitro} \; (R, T) = d_{in\ vivo} \; (R, T) \quad \text{and} \quad R \leq T \Rightarrow \psi(R) \leq \psi(T)$$

Less formally speaking we are interested to know if *in vitro* similarity implies bioequivalence and if a better dissolution implies a better bioavailablity. It is clear that the response depends on the active substance and physiopatological parameters but also on the particular metrics choiced for characterizing *in vitro* and *in vivo* curves as well as distances between its. The response is not so simple, first of all since

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due to the complexity of the phenomena studied, both metrics and order *in vitro* and *in vivo* are not well defined. The great number of dissolution and bioequivalence metrics (Enachescu *et al*. 2003) show that the problem is yet to be solved.

In vitro dissolution tests. Dissolution tests were performed using the method indicated by USP or according to the specifications provided by the producers. As metrics of dissolution were considered the factors f_1 and f_2.

Clinical trials. Each study was performed on healthy volunteers. Experiments were of the standard type: cross-over, with two periods and two sequences.

Analytical methods. Plasma levels of the drugs were evaluated using validated liquid chromatographic methods, with UV or mass spectometry detection.

In judging the results it should be kept in mind that that in so-called *in vitro*/*in vivo* correlations, we practically jump over one step – *in vivo* dissolution, which is by far more variable and more complex than the *in vitro* dissolution. Since *in vitro* dissolution conditions are often far from the *in vivo* conditions, we have non-correlation between the two dissolutions.
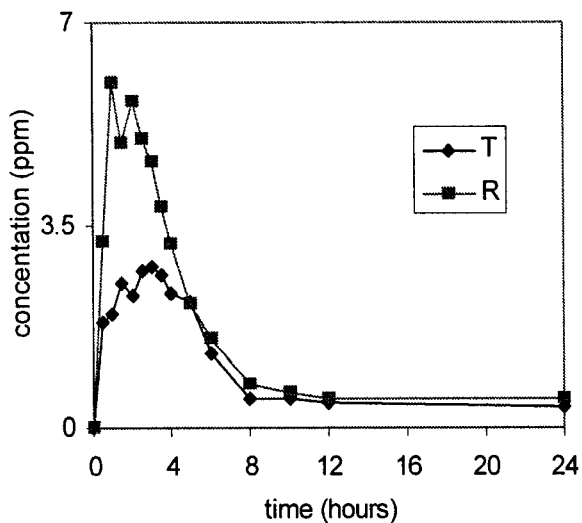
Similar dissolution and non-bioequivalence. This is the case for many acidic or basic drugs. Most representative is mefenamic acid (fig. 1). If we calculate factors f_1 or f_2, dissolution curves are similar. The curves are practically much more similar that these factors indicate, negative and positive areas between the curves being approximately equal. Consequently, if we chose as norm of dissolution curves area under experimental data (AUC), the distance between the two curves $d(ref, test) = | AUC_{ref} - AUC_{test} |$ is approximately zero. In spite of this high similarity in dissolution, plasma leves are quite different when it concerns c_{max} and AUC.

Non-correlation issues from the fact that *in vitro* release medium had a pH=8, which is far from physiological conditions. Since mefenamic has a very low solubility in acidic and neutral media following its lipophilic, week acid character, the alkaline medium was chosen by the producer in order to obtain a "good dissolution" *in vitro*, without considering to a correlation with *in vivo* release conditions.
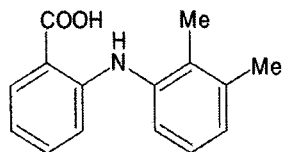
Mean plasma levels of mefenamic acid

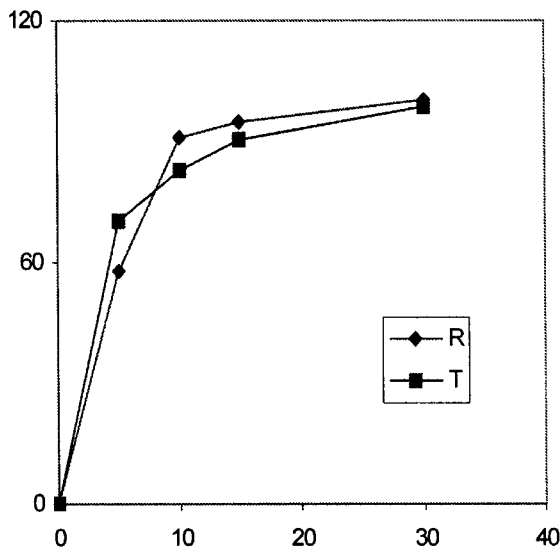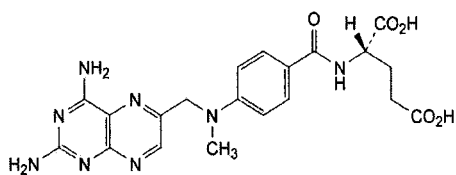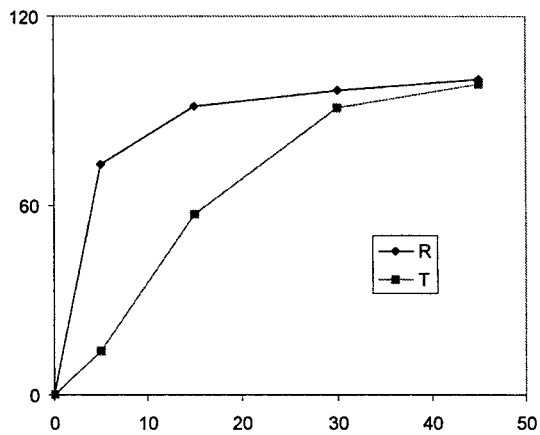Mean dissolution profiles for mefenamic acid

Fig. 1.

Mean dissolution profiles for methotrexat

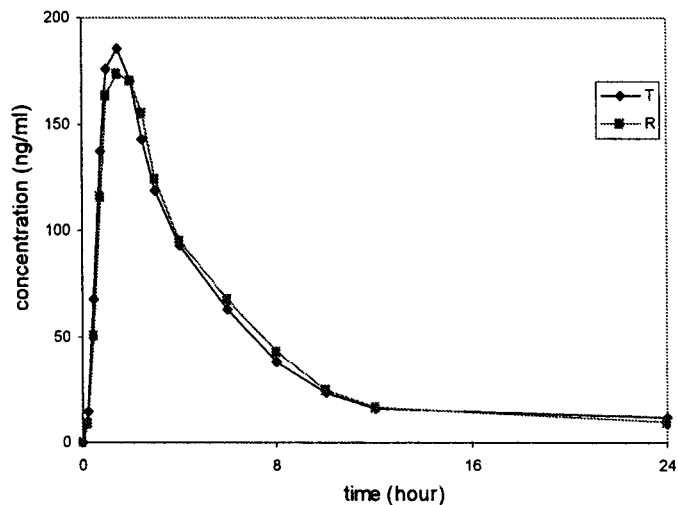Mean plasma levels of methotrexate

Fig. 2.

Non-similar dissolution but bioequivalence. This is quite a frequent case for many molecules of different structures. One explanation could arise from the fact that *in vitro* non-similarity is connected with a somewhat 10% difference (for all pairs of points of dissolution curves there is 10% difference, i.e. $f_2 = 50$) and *in vivo* non-bioequivalence is about a 20% difference. This is a very roughly characterization for the acceptance limits of dissolution and bioequivalence, but the idea that dissolution metrics are more refined that bioequivalence metrics deserves much more attention.

In some cases, the mechanism may be more specific, for example in the case of methotrexate (fig. 2).

The reference and the product tested attained approximately superposable mean plasma levels curves, though *in vitro* dissolution curves were dissimilar whatever the dissolution metrics used to measure their distance. Since methotrexate has a great molecular weight, a free diffusion mechanism of absorption is less probable. An active transport was not described. An absorption via embedding in micelles of physiological surface active agents (mainly bile acids) remains a more reliable hypothesis. If formation and transfer of micelles across intestinal barrier is the rate-limiting step of the entire process (*in vivo* release and absorption), quenching of differences in dissolution could appear.

Simvastatin was also in this category of non-correlation, but in this case there is clearly a problem of metrics of dissolution. As discussed above, since the areas under dissolution curves were approximately equal, it is more reasonable to think that the products have similar dissolution though all usual metrics argue against this idea.

Conclusion. Though helpful, the use of a comparative dissolution in prediction of *in vivo* bioequivalence offers some problems and the risk of false predictions should be kept in mind. The use of acidic dissolution medium for basic drugs or basic medium for acidic drugs lead to more optimistic estimations than the actual situation. In upcoming rules for prediction of *in vivo* behaviour from dissolution results we need more knowledge about adequate tests for use and comparison of tests made under different conditions.

References

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