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Evaluation of non-sink dialysis methods for the measurement of drug release from colloids: effects of drug partition

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Summary

The technique of non-sink dialysis across a membrane has been used in the past for determining the release profiles of drugs from colloidal formulations. It is demonstrated that in most cases the results obtained are in fact independent of the release rate, and that the observed release rate is solely dependent on the partition coefficient of the drug between the colloid and its continuous phase. It is suggested that this may form a useful method for measuring partition in disperse systems.

Introduction

In the design of colloidal drug–carrier formulations, it is of central importance to study the rate at which the drug is released from the carrier. This can be used as quality control data, to predict in vivo behaviour, or to study the structure and release mechanism of the system. Many methods have been used to measure release rate, including continuous flow methods (e.g. Burgess et al., 1987; Koosha et al., 1988), sampling methods (Henry-Michelland et al., 1987) and in situ methods (Illum et al., 1986). In all of these it is appreciated that, in order to measure the true release profile, it is necessary that the carrier has the maximum opportunity to release its drug load, i.e. that there should be an insignificant amount of released drug in the continuous phase at all times. This is usually achieved by either having a large volume of continuous phase, or by replacing it in a flow experiment. An experiment carried out under these conditions is referred to as a perfect sink experiment; although a perfect sink is not attainable in practice, it can often be approached sufficiently closely to obtain useful results.

A further method of measuring drug release is that in which the undiluted colloidal carrier and the sink phase are separated by a dialysis membrane (Sasaki et al., 1984; Hashida et al., 1980; Miyazaki et al., 1986; Benita et al., 1986). This method appears to have evolved from experiments designed to study release from systems which are applied topically and not diluted, for which it is a valid performance indicator. The carrier suspension is never diluted, and so the experiment is not performed under sink conditions. As a consequence, it does not measure the true release rate, as will be demonstrated, and so it has widely fallen out of favour, although it still reappears

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occasionally. We demonstrate that the results obtained in most cases reflect only the partition of drug between carrier and continuous phase, which is less widely appreciated. It is possible that this may form a useful method for measuring partition in disperse systems which are difficult to separate without perturbing the equilibrium.

Theoretical

Consider the kinetic situation shown in Fig. 1. The undiluted colloidal carrier system consists of a volume V_a of disperse phase in a volume V_b of continuous phase. This is separated from an acceptor phase of volume V_c by a dialysis membrane. We denote the drug concentration in the 3 compartments as [A], [B], and [C]. Drug in compartments A and B is in equilibrium due to partition. We denote the partition coefficient as K_p and note that, in the systems we are considering, k_{-1} will be fast, so that K_p will be large and [B] will be small. There would be little point in constructing a drug delivery system in which this was not true. Finally we allow [B] to transfer through the membrane to [C] with a first-order rate constant k_2. We assume that V_c is large so that the reverse transfer, with rate constant k_{-2}, is effectively zero.

Although this kinetic system can be solved analytically, it is simpler to use an approximation such as the steady-state hypothesis, which assumes

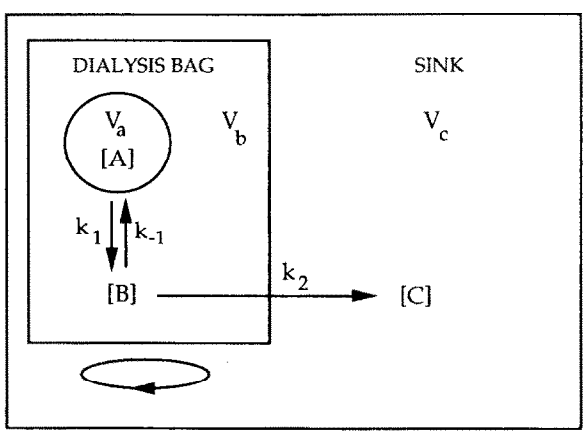

Fig. 1. Kinetic scheme for non-sink dialysis.

that, after a short equilibration time, the concentration of the intermediate [B] is small (which is true if K_p is large) and so $d[B]/dt = 0$. Thus we can say that the rate at which drug leaves compartment A is equal to the rate at which it enters compartment C:

$$\frac{d[A]}{dt} = -\frac{d[C]}{dt} \cdot \frac{V_c}{V_a} \tag{1}$$

If the partition equilibrium is fast (we will examine this further below), then [B] will be given by partition:

$$[B] = \frac{[A]}{K_p} \tag{2}$$

But by mass action:

$$\frac{d[C]}{dt} = k_2[B]\frac{V_b}{V_c} = k_2\frac{[A]}{K_p} \cdot \frac{V_b}{V_c} \tag{3}$$

From Eqn. 1

$$\frac{d[A]}{dt} = -k_2\frac{[A]}{K_p} \cdot \frac{V_b}{V_a} \tag{4}$$

Thus

$$[A]_t = [A]_0 \exp\left(-k_2\frac{[A]}{K_p} \cdot \frac{V_b}{V_a} \cdot t\right) \tag{5}$$

So the rate of loss of A is exponential (since we have assumed first-order processes), but with a rate constant that is determined by the system volumes, the membrane transfer rate, and the partition coefficient. Note that V_c does not appear, largely since we have assumed that it was sufficiently large to cause the back-diffusion across the membrane to be zero. The most important point is that all the information concerning k_1, the release rate, has disappeared. Additionally the rate at which drug appears in compartment C is dependent on experimental parameters which may be difficult to keep constant during the experiment; for example, k_2 may alter with membrane hydration or age, and V_b may increase or decrease due to osmosis.

Discussion

It has been shown that the 'release rate' determined in this experiment depends largely on the drug partition between the colloid and its continuous phase (modified by the appropriate volumes of the experiment). It is interesting to ask how this method could have been accepted in the past as measuring the true release rate. This probably stems in part from the result obtained when a bolus of drug is added to one side of the dialysis cell and its appearance on the other side measured. This control measurement is essential in all drug release experiments to ensure that the apparatus has a sufficiently short time resolution to resolve release from the carrier. We have demonstrated the distorting effects that such 'instrument response functions' can have on drug release data (Washington and Koosha, 1989).

If a drug bolus is added in this way, its appearance on the acceptor side of the membrane is rapid. This is due to the fact that diffusion across the membrane is driven by the full concentration of drug in the cell, since all the drug is in the continuous phase. This makes the experiment appear to have a time resolution which is relatively short (of the order of an hour for many dialysis membranes). Consequently an experimenter is misled into believing that the experiment can measure the release rate of a drug which is being released more slowly than this from the carrier. This is in fact not the case; when the carrier is present, the drug concentration driving diffusion is reduced by a factor of the drug/carrier partition coefficient. Drug release cannot increase this driving concentration since it is partition controlled (in the model under consideration).

The partition coefficient dependence of the observed 'release rate' is well illustrated by the data of Sasaki et al. (1984). These authors produced a range of mitomycin prodrugs which were incorporated into liposomes or emulsions. The 'release rates' were measured by dialysis techniques similar to those discussed here. The observed release rates correlate well with the octanol–water partition coefficients of the prodrugs quoted by the authors. The most rapid release was observed from the prodrug with the lowest partition coefficient, while the slowest was observed from the most hydrophobic prodrug. Between these limits the rank correlation of 'drug release' and hydrophobicity appears to be extremely good.

It is interesting to consider cases in which this experiment may be capable of obtaining results which reflect the true release rate. In two cases useful results may be obtained:

(1) The previous analysis assumes that transport across the membrane is sufficiently slow that A and B are in equilibrium. This will not be true if the release rate is extremely slow. In this case k_1 becomes rate limiting; this will be true when B is removed by k_2 much faster than k_1 can produce it:

$$k_2[\mathrm{B}] \gg k_1[\mathrm{A}] \cdot \frac{V_b}{V_a} \tag{6}$$

Introducing the partition coefficient:

$$k_1 \ll \frac{k_2}{K_p} \cdot \frac{V_a}{V_b} \tag{7}$$

In a typical experiment k_2 may be 2 h^{-1}, K_p approximately 100 and V_a/V_b approximately 0.01. Thus k_1 must be less than 2×10^{-4} h^{-1} to influence the results; this corresponds to a release half-life of around 150 days. Sustained release of this degree is unlikely to be achieved from a colloidal delivery system.

(2) The analysis applies only to the case in which drug release is driven by diffusion out of a carrier which retains its composition and structural integrity during the experiment. In some cases part or all of the drug may be released by carrier attrition or dissolution, or the drug may be dissolved by leaching from its carrier. In these cases accumulation of the drug in the surrounding phase is not controlled by partition and the experiment will provide a useful result if the membrane diffusion rate is much faster than the release rate after the volume changes have been taken into account:

$$k_1 \ll k_2 \cdot \frac{V_a}{V_b} \tag{8}$$

This is essentially (7) with $K_p = 1$.

In practice it would be unwise to perform an experiment in this case because the relatively large amount of drug accumulating in the continuous phase on the sample side of the membrane may reassociate with the carrier by adsorption, or the solubility of the drug in this phase may be exceeded.

It may be suggested that measurement of the infinite sink release rate is not necessary if the colloidal carrier is not going to be diluted on injection, i.e. is not to be injected intravenously but will be administered intramuscularly or intraperitoneally. In these cases the degree of dilution is small or ill-defined, and the release characteristics are dependent on local diffusion and transport in the local tissues. Consequently the technique described here may not provide a good model of the in vivo situation, if that is what was desired. However, if a correlation was observed between the dialysis 'release rate' and the behaviour of the system in vivo, it is likely to be a consequence of the central importance of partition on both experiments.

It is possible that the sensitivity of the method to partition coefficient may be a useful technique for the measurement of that parameter in colloidal systems. Partition is of central importance in the design of a drug-carrier system; there is little value in designing a sophisticated colloidal delivery system if, after an unpredictable shelf-life, half the drug is present in the aqueous phase. It can be difficult to measure this in practice, as the separation of a colloid from its continuous phase can be difficult, and the separation may perturb the partition equilibrium and lead to an erroneous value. However, the sensitivity of the dialysis method to variations in the experimental volumes and membrane characteristics would require these parameters to be carefully controlled in such an experiment.

Conclusions

It has been demonstrated that non-sink dialysis can only provide useful kinetic drug release data in a limited number of cases. Despite this the method is still used by some workers. The model discussed here is particularly relevant for emulsion delivery systems in which partition is of major importance, but the method suffers from major disadvantages with many other types of colloidal delivery systems unless their release rate is extremely slow. However, it is possible that it may form a useful way to measure partition in colloidal systems.

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