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Special Article

Pharmacokinetically Guided Dose Escalation in Phase I Clinical Trials. Commentary and Proposed Guidelines

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INTRODUCTION

IN BOTH its preclinical development and its clinical application, cancer chemotherapy remains an essentially empirical science. The early clinical strategy currently employed focusses the ethical problems, inherent in any empirical study in humans, upon a small group of patients and the medical and nursing staff who care for them. In the first instance a new compound undergoes phase I evaluation to characterize the toxic effects and determine the maximum tolerated dose (MTD). This is followed by phase II studies in which evidence of activity against particular disease types is sought. Although there will be a move towards phase II studies based on human cell line and xenograft data, until such time as routine individual patient chemosensitivity testing becomes a practical proposition it is likely that phase II trials will remain with us in their current form. However, given that the phase I evaluation has been correctly performed, all patients in phase II studies should receive a potentially therapeutic yet safe dose. The same statement cannot be made for phase I studies.

At the start of a phase I study patients are deliberately treated at what is predicted to be a non-toxic and often non-therapeutic dose. This is usually 1/10th of the dose lethal to 10% of mice so treated (the LD_{10}), with the drug given by the same schedule and the dose expressed in units of mass/surface area (e.g. mg/m^2) [1, 2]. Provided toxicity is not observed at the starting dose, doses are then escalated until side-effects are encountered which are deemed to be dose-limiting. A dose is then selected which is considered safe for phase II trials. However, this phase I strategy can require over 10 escalation steps, with the majority of patients treated at non-toxic and probably non-therapeutic doses. Although the human MTD is often similar to the mouse LD_{10} sufficient variation exists so as to preclude the use of larger escalation steps. In recognition of this problem, Collins *et al.* in the Blood Level Working Group at the U.S. National Cancer Institute have proposed modifications to the current system [3].

The central concept of the new proposals is that pharmacokinetic variables may largely account for interspecies differences in toxic doses and that the area under the plasma drug concentration vs. time curve (AUC or $C \times T$) may, therefore, be a more representative measure of drug exposure than dose. Thus the ratio of the absolute AUC values (in units of concentration × time, e.g. mM× h) at the human MTD and mouse LD_{10} should be closer to unity than the ratio of the doses. In a retrospective analysis, data presented for eight drugs indicated that this was on the whole the case [3]. Doxorubicin was a particularly marked example where the ratio of the human MTD to the mouse LD_{10} was 5 whilst the ratio of the AUC values at the human MTD and mouse LD_{10} was 0.8. Reasonable agreement between AUC values (ratios 0.5–1.5) was observed for four other agents although the

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ratios of the human MTD to mouse LD_{10} values were also close to unity. Notable exceptions to the apparent rule were three antimetabolites, for which there was either a similarly poor correlation for both dose and AUC (PALA and F-araAMP) or a better correlation for dose than AUC (dihydroazacytidine).

On the basis of these data Collins *et al.* have proposed that doses in phase I trials should be escalated in a manner so as to achieve a dose which gives the same AUC as that seen in mice at the LD_{10} within 3–4 escalations from the starting dose. Such a reduction in the number of dose levels would decrease the demand on clinical resources and improve the individual patient's chances of receiving a therapeutic dose. It should be emphasized that in none of the eight drugs evaluated retrospectively would patients have been put at higher risk compared to current dose-escalation procedures.

In view of these interesting observations and proposals, the Pharmacokinetics and Metabolism (PAM) group of the EORTC recently surveyed the pharmacokinetic and toxicity data available to it in an attempt to find information supporting or contradicting the results and conclusions of Collins *et al.* The group met to discuss the findings during the EORTC Joint Groups meeting in Edinburgh, June 1986. A number of potential problems and pitfalls were highlighted and with these in mind the current proposals have been drawn up for discussion by preclinical and clinical pharmacologists and by clinicians involved in phase I studies.

POTENTIAL PITFALLS FOR PHARMACOKINETICALLY GUIDED DOSE ESCALATION

Technical factors

The importance of matching the conditions used to derive the preclinical and clinical pharmacokinetic and toxicity data cannot be over-emphasized. The route and schedule of administration and the drug vehicle can markedly influence both plasma concentrations and toxicity. Preclinical studies should employ intravenous administration in the proposed clinical vehicle, preferably with the clinical formulation, whereas the oral route should be used when this is the intended means of clinical adminstration. Intraperitoneal or subcutaneous administration should not be used. For those drugs requiring multiple administration, pharmacokinetic studies should be performed on at least the first and last treatment days to ensure that no induction or inhibition of metabolism or other elimination pathways has taken place, as well as to check for accumulation.

As well as sex and strain differences, variations between groups of animals and circadian rhythms can also be important. Ideally the toxicity and pharmacokinetic studies would be performed on the same randomized group of mice. Otherwise care should be taken to ensure that the animals and conditions are as identical as possible.

The drug assay used should be specific for the parent drug and any known active metabolites. It should be of sufficient sensitivity to allow accurate analysis of concentrations following LD_{10} doses *and* 1/10th LD_{10} doses. These latter concentrations should be similar to those encountered at the initiation of the clinical trial, and hence those used to determine the dose escalation required to achieve the desired final AUC. The data points should be distributed appropriately over time so as to allow rapid identification of all disposition phases. Particular care should be taken to quantify absorption and distribution phases as well as prolonged terminal elimination phases, as these may contribute significantly to the total AUC. Where drugs are administered by intravenous infusion and the infusion time is ignored, the AUC should be corrected when appreciable error would be otherwise introduced. Quality control should be rigorous and where possible the same laboratory should analyse preclinical and clinical samples. Otherwise samples should be exchanged and good agreement between collaborating centres demonstrated.

Pharmacokinetic factors

Although species differences in pharmacokinetics can result in interspecies differences in toxic doses, the pharmacokinetically guided dose escalation scheme will compensate for those which alter the plasma AUC. However, certain problems may still be encountered when toxicity is not solely a function of total parent drug plasma levels.

(i) *Differences in plasma protein binding.* The majority of drug assays quantitate the total amount of drug in the plasma whilst toxicity may be a function of the free drug concentration for agents with low tissue extraction ratios. Thus, for those compounds which undergo significant protein binding, the comparative extent and affinity of binding to human and murine plasma proteins should be determined. If these are not comparable, yet the binding is linear over the concentration range of interest, the appropriate correction factor could be included and escalation made on the basis of the free drug AUC.

(ii) *Differences in metabolism.* The majority of antitumour agents undergo host metabolism which can affect their activity in a number of ways. Given

that the extent and/or route of metabolism may vary between species it is important to define the metabolic pathways of a new compound and to describe the activity and toxicity of its metabolites. In general, three classes of anticancer drug can be defined with regard to metabolism at sites other than the target organ or tissue.

(a) Active (toxic) drugs which are metabolized to inactive (non-toxic) species: in this case estimation of the parent drug alone is all that is required. That is, species differences in metabolism will be reflected in differences in the parent drug AUC value and hence allowed for in the new procedures.

(b) Active (toxic) drugs which are metabolized to active (toxic) species: as metabolite concentrations will not necessarily be predicted by those of the parent drug, both should be quantitated. This is particularly important where the metabolites represent a significant proportion of total drug-related material.

(c) Inactive (non-toxic) drugs which are metabolized to active (toxic) species: as in (b) parent drug concentrations may be very misleading, and estimation of the AUC for active metabolites is essential.

(iii) *Non-linear pharmacokinetics.* Non-linear pharmacokinetics can be encountered when a saturable step is involved in drug absorption, distribution or elimination, e.g. as in carrier mediated transport or metabolic clearance. Thus although the comparative AUC at the human MTD and mouse LD_{10} may not be affected, dose escalation is greatly complicated when a linear relationship between dose and AUC does not exist. As a result of non-linearity a small increase in dose can result in a large increase in AUC. For such drugs pharmacokinetic studies are in any case essential for safe dose escalation. However, dose increments will depend on the extent of non-linearity found for a given drug and the concentration at which it is encountered in each species.

(iv) *Patient heterogeneity.* Many factors contribute to differences in AUC in individual patients. These include age, sex, hepatic and renal function and drug interactions. Important information can be gained by identifying such variables. An adequate number of patients should be studied so that the error in the AUC estimate is minimized.

It is important to remember that the preclinical pharmacokinetics and LD_{10} will be determined in previously untreated, healthy animals while the pharmacokinetics and MTD in man may be established in previously treated individuals with extensive disease.

(v) *Tissue drug concentrations.* Drug concentrations at the site of toxicity may not be equal to those in the plasma. So long as these are directly proportional to each other by the same factor in both species this will not cause a problem. The accrual of data on tissue concentrations is limited by ethical, logistic and analytical considerations. Nevertheless efforts should be made to obtain tissue data, and this should be facilitated by new non-invasive techniques such as magnetic resonance spectroscopy.

(vi) *Chronopharmacology.* Preclinical and clinical data indicate that time of drug administration may be an important variable for pharmacokinetics and toxicity, through the effects of circadian rhythms.

Pharmacodynamic factors

The AUC required to produce toxicity may differ between species as a result of variations in the intrinsic sensitivity of the target organ or cell. There may for example be differences in the activities of activating or detoxifying enzymes or their cofactors in susceptible cells. The structure of the molecular target may be different or one species may lack the ability to repair particular lesions whilst the other may be competent. Furthermore, as a result of such differences the dose-limiting organ or tissue may not be constant between species. The proposed pharmacologically guided dose escalation scheme does not in itself allow compensation for pharmacodynamic variables. In cases where pharmacodynamic differences between species (or individuals) are suspected, rational use of the new procedure will require the input of data on the comparative *in vitro* sensitivity of dose-limiting tissues or cells. Such assays may not always be available.

The current data [3] suggest that antimetabolite toxicity is not well predicted either by dose or AUC data. As a class, the cytotoxicity of nucleic acid antimetabolites is highly dependent upon the target cell levels of activating and inactivating enzymes, as well as the levels of extracellular nucleic acid precursors. Hence it is no surprise that antimetabolites may not be good candidates for pharmacokinetically guided dose escalation. Related problems may also be encountered with the biological response modifiers. Indeed it is not at all clear whether dose escalation to the MTD is appropriate with such agents. Nevertheless it will be important to monitor circulating drug concentrations and to attempt to relate exposure to biological or biochemical indicators of drug effect.

SUMMARY OF DATA AVAILABLE TO THE EORTC PAM GROUP ON PRECLINICAL AND CLINICAL PHARMACOKINETICS AND TOXICITY

Given the large number of potential pitfalls indicated above, it is clear that retrospective analyses of murine and human pharmacokinetic and toxicity data must be interpreted with extreme caution.

Particular problems encountered were the widespread use of non-parenteral routes of administration in murine studies, the lack of an accurate LD_{10} in mice and the use of doses other than the LD_{10} for pharmacokinetic studies. In addition, for certain established agents there is no clear agreement on the single agent MTD in man. Despite this, certain overall conclusions can be reached on the use of mouse AUC values to predict for MTD in man.

For the anthracyclines, doxorubicin, daunorubicin, 4′-deoxydoxorubicin and 4′-epidoxorubicin, AUC was as good or better a predictor of toxicity than dose. This was not apparently so for 4′-demethoxydaunorubicin.

For platinum complexes a reasonable correlation between AUC and toxicity could also be obtained. This is the subject of a prospective study to be published shortly. Data for the chloroethylnitrosoureas BCNU, CCNU and TCNU, as with the mechanistically related mitozolomide, suggested that AUC was on the whole a poorer predictor of toxicity than dose. A very thorough series of experiments have shown that for the alkylating agent diacetyl dianhydrogalactitol both dose and AUC predict poorly for clinical toxicity, humans being substantially less sensitive than mice. Interestingly, human bone marrow (the target tissue) is also less sensitive *in vitro* and hence this drug may be susceptible to pharmacodynamic variables. The data base for the more established alkylating agents was insufficient to allow sensible conclusions to be made.

The *N*-methyl drugs hexamethylmelamine, pentamethylmelamine, dacarbazine and *N*-methylformamide are all examples of compounds which probably require metabolism in order to display activity. Hence it was not surprising that in each case the parent compound AUC was a poor predictor of toxicity. In contrast, the toxicity of the new agent trimelamol, an analogue of hexamethylmelamine selected specifically because it does not require metabolic activation, was more accurately predicted by AUC than by dose.

The only data available with antimetabolites indicated that for the novel thymidylate synthase inhibitor N^{10}-propargyl-5,8-dideazafolic acid AUC and dose were similar in predicting nephrotoxicity. Preliminary data with the new antifolate MZPES suggest that peak plasma concentration, rather than dose or AUC is the best predictor of toxicity. This is probably related to the acute neurotoxic nature of the dose-limiting side-effect in both mice and humans. Peak plasma concentrations are likely to be indicative of peak concentration in the central nervous system.

In addition to the European experience, the U.S. National Cancer Institute database has been updated to a total of 13 compounds. This now demonstrates the advantage of AUC over dose in mouse/human correlations for 5-azacytidine, doxorubicin, teroxirone, trimelamol (European data), and thiotepa, and similar results for AUC and dose with diaziquone, indicine-*N*-oxide, amsacrine, deoxycoformycin, tiazafurin, PALA and F-ara-AMP, with only dihydroazacytidine giving a poorer result for AUC.

It is important to emphasize that the worldwide experience to date reveals no example in which the use of pharmacokinetically guided dose escalation would introduce a greater risk to patients than the procedures currently employed. In many cases the new procedures would lead to a more efficient dose escalation.

CONCLUSIONS AND RECOMMENDATIONS

Although care should be exercised in drawing firm conclusions, particularly in view of the generally retrospective nature of the analyses, the data provide clear encouragement for the further evaluation of pharmacokinetically guided dose escalation schemes in phase I studies. On the basis of the review outlined above, the proposals in Table 1 are advanced for discussion. It may not be possible or practical to cover all the steps for a given compound. The *minimum* requirements are considered to be: (i) determination of the drug AUC at the mouse LD_{10} and below, checking for non-linearity and protein binding; (ii) determination of the drug AUC at the phase I starting dose; (iii) dose escalation to MTD as appropriate, with AUC monitoring at all steps.

It is our view that the rational development of new anticancer agents should always include pharmacokinetic analyses during preclinical and phase I clinical evaluation. This is now already the case for most new drugs. The proposals outlined in Table 1 would not therefore require a major increase in resources. Perhaps the greatest logistic problem is the integration of preclinical pharmcokinetic and toxicity studies, so as to eliminate the need to duplicate the determination of the LD_{10}.

In view of the apparent safety of the proposed procedures and the benefit in terms of more rapid definition of the MTD for many compounds, we propose they be evaluated prospectively. Each com-

Table 1. *Proposed scheme for the conduct of pharmacokinetically guided phase I dose escalation studies*

Preclinical

1. Determine the presence of metabolites and the effect of host metabolism on drug activity and toxicity.*
2. Develop a drug assay for the parent drug and any active or toxic metabolites with adequate sensitivity for 1/10th LD_{10} dose.
3. Randomize mice, and, using the proposed clinical route, vehicle and schedule, determine:
   a. LD_{10}
   b. AUC at the LD_{10} for the parent drug and active/toxic metabolites.
   c. AUC at $0.5 \times LD_{10}$ and $0.1 \times LD_{10}$.†
   d. If possible, quantitate the dose-limiting toxicity at the LD_{10}, $0.5 \times LD_{10}$ and $0.1\ LD_{10}$ and correlate with AUC.
4. Determine protein binding in mouse and human plasma at concentrations achieved with the above doses.

Clinical

1. Initiate the clinical study at 1/10th mouse LD_{10} and treat 3–5 patients so as to determine the AUC with acceptable accuracy.
2. Using an appropriate escalation scheme (e.g. see ref. [3]) arrive at the projected MTD AUC with monitoring of drug and relevant metabolite concentrations at every dose level and modification as required for non-linearity.§

*The study may be extended by the use of additional animal species, enzyme inducers/inhibitors, or *in vitro* studies with microsomal preparations or hepatocytes.
†Check for non-linear kinetics.
‡For example, the square root (geometric mean) or extended factors of two method with modification as required, e.g. for linearity.
§Considering also metabolic, mechanism of action and pharmacodynamic (e.g. normal tissue) data.

pound should be considered individually on its own merit, and the pharmacokinetic data considered against a background of information on metabolism and mechanism of action, and in conjunction with pharmacodynamic data and *in vitro* normal tissue toxicity testing where possible. Potential escalation schemes [3] include: (i) the square root or geometric mean method, in which the first dose escalation factor is equal to the square root of the ratio of the AUC at the mouse LD_{10} (target AUC) to the AUC at the phase I entry dose; (ii) the extended factors of two method, in which doses are doubled until the range of the target AUC is reached. In both cases the escalation can be completed with a modified Fibonacci procedure. Other modifications are possible, based on the available data for individual drugs. Particular care will be required where non-linear pharmacokinetics are known or suspected. These recommendations are broadly in agreement with those currently advocated by the Blood Level Working Group of the U.S. National Cancer Institute.

It is hoped that by adopting such an approach the number of patients participating in phase I trials can be reduced and their chances of receiving a therapeutic yet safe dose significantly improved.

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