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Changing the field of carcinogenicity testing of human pharmaceuticals by emphasizing mode of action

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Abstract

Lifetime testing for carcinogenicity of pharmaceuticals in rodents has been a controversial issue since the start of the International Conference on Harmonisation in 1990. Since 2010 the debate reached a new level following the proposal that a negative outcome of carcinogenicity studies can be predicted based upon the findings of 6 months studies. In addition, the value of pharmacological mode of action (MoA) data for positive prediction has become apparent.

Such predictions rest heavily on prior data and first-in-class compounds are difficult to evaluate in this way. Virtual waivers are rarely given to such compounds. We discuss here the utility of in vitro -omics approaches to identify involvement of signalling pathways in the mode of action of human pharmaceuticals that might bear relevance for prediction of carcinogenic properties. It is of particular significance that this approach to mode of action analysis would comprise human relevance, and would not relate solely to the prediction of carcinogenicity outcome in rats.

Our ultimate aim is to establish in vitro fluorescent reporters in human cells where individual key events that are functionally relevant in the signalling programs that drive cell proliferation are integrated. This would allow the qualitative and quantitative evaluation of key event activation as a predictive tool for the determination of the intrinsic carcinogenic potential of compounds. In the first instance, this involves the nuclear hormone receptor-mediated tumor promotor activity (e.g. estrogen receptor signalling or peroxisome proliferator PPAR-gamma signalling), which are both current topics of debate.

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Pharmaceuticals, Carcinogenicity testing, Mode of action, Genomic approaches, Nuclear Receptors.

1. Introduction

The most important long-term safety issue for human pharmaceuticals is the potential for cancer causing properties. Since the middle of the 20th century this is addressed by the performance of life-time carcinogenicity studies in rats and mice.

However, this approach has been criticized from all quarters. In particular the use of mice was found to be redundant [19], and the use of the Maximum Tolerated Dose (MTD) approach to dose selection has been the target of intense debate.

The testing of human pharmaceuticals for carcinogenicity has been under discussion since the early 1990's under the auspices of the International Conference on Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (shortly ICH). Various aspects have been introduced into guidance on this topic, such as the selection of the maximum dose for a life-time study (ICH S1C) [9] and the introduction of short-term animal models (i.e. transgenic or knock-out mice) with greater sensitivity for the induction of tumours of human-relevance (ICHS1B) [8].

The fact that more than 50% of chronic administration human pharmaceuticals are carcinogenic in rodents despite being authorised for therapeutic use [25,5] casts some doubt to the usefulness of the rodent studies in assessing human relevance.

It has to be said that carcinogenicity studies are usually conducted during the final stages of drug development, not the least to avoid performing studies with drug candidates that prove to be clinically inefficacious. The costs of rodent lifetime carcinogenicity studies are high, and Sponsor companies will naturally prefer to avoid the conduct of unnecessary or pointless studies.

2. Reduction of carcinogenicity studies by predicting the outcome

2.1. Histopathology and pharmacology as predictors

Since 2010 there is renewed debate on the need for rodent lifetime carcinogenicity studies following the analysis of Ref. [16], later extended by Ref. [21], showing that the negative outcome of a carcinogenicity study could be predicted by the findings of the preceding 6 months general toxicity study in the same species. Absence of hormonal stimulation and genotoxic effects were also indicated as essential for outcome prediction. This prediction of negative outcome entirely based on histopathology was criticised as too limited in approach. Hyperplasia, rather than hypertrophy, is generally accepted as a putative pre-neoplastic tissue change. In a recent review of 289 human pharmaceuticals, however, in many cases no hyperplasia was observed in the 6 month studies, while tumours were observed in lifetime studies with the same compounds at a similar dose-range. Moreover, in 6 cases showing liver hyperplasia in the 6 months study, no liver tumours were subsequently seen [27]. These observations suggest that the use of hyperplasia alone as a predictive factor is less reliable than expected. This was also shown for calciferol analogues, which all induce adrenal pheochromocytomas but showed no evidence of hyperplasia in the 6 months studies [26]. However, with a specific detection method (i.e. BrdU-labelling or specific staining such as Ki-67) enhanced proliferation can be detected at 6 months [23,29]. These findings with biomarkers such as BrdU and Ki-67 suggest that greater insight into the mode-of-action is needed, especially in relation to potential proliferative properties.

In order to gain a more specific prediction, we have introduced data on the pharmacological class of the compound into the analysis. Direct and indirect pharmacological data was added retrospectively to predict the outcome of carcinogenicity testing, and this enhanced the precision of both negative and positive carcinogenicity prediction to >95% [27].

The pharmacological properties relating to possible direct or indirect cell proliferation are more important than just the evidence for hormonal effects, proposed by Ref. [21]. Yet, a difficulty may arise for the first-in-class compounds. How can we predict the pharmacological properties when almost nothing is known about the target tissue and its relation with proliferative aspects? Since prior class experience is, by definition, absent for first-in-class compounds, it is agreed that a higher standard of relevant evidence would be essential to support a proper prediction of carcinogenicity [11,12]. In our retrospective analysis a similar problem was encountered for single-in-class compounds. In some cases there is evidence from literature that, for example, stimulation of a certain receptor will lead to cell proliferation, in other cases the available data for a single-in-class agent is the first evidence for the presence or absence of such an association. To make a step really possible we should preferably predict such a relation without first conducting a rodent lifetime carcinogenicity study for the first-in-class compound. From a regulatory viewpoint it will be difficult to know which compound can really be designated as first-in-class, as it depends also on the efficiency of a sponsor to plan and conduct its development, in relation to the competing industry sponsors.

2.2. ICH-initiative

The industrial and regulatory parties in ICH observed possibilities to substantially reduce the number of carcinogenicity studies by at least 40%, if a carcinogenicity prediction approach is introduced. In a rather unique regulatory experiment Drug Regulatory Authorities are currently testing the hypothesis that the outcome of rodent carcinogenicity studies can be readily predicted taking into consideration the findings of 6 months repeated dose toxicity data, genotoxicity data and direct and indirect pharmacological data. From 2013 onwards, companies are invited to provide a Carcinogenicity Assessment Document (CAD). The CADs are expected to be based on a weight-of-evidence approach taking into account the possible hyperplastic and hypertrophic findings observed in the chronic rat toxicity study. An important condition is that this should be done before the start of or early enough after (<14 months) the start of the carcinogenicity study, before any evidence of obvious hyperplasia or tumours is detectable.

However, as explained above, an important factor is the mode-of-action. As this weight-of-evidence approach will include all the data present at the stage of deciding on further development of a pharmaceutical compound at the end of Phase II, available knowledge on the mode-of action can be integrated with results from repeated dose toxicity studies, e.g. as evidence for hormonal effects. The assessment should result in classification of the compound in one of the categories explained in Table 1 [10].

Compounds classified into category 2 will warrant further carcinogenicity studies because of uncertainty about the outcome. Both for category 1 and 3 a (virtual) waiver for carcinogenicity studies will be given. Category 1 is most important from a human risk assessment perspective. If a compound is likely to be tumorigenic in humans based on the mode of action then the addition of rodent carcinogenicity studies will not add value to the weight of evidence analysis (Table 1). This is especially the case for non-genotoxic compounds which are categorized as IARC Group 1 [proven human carcinogens] [7]. Cyclosporine as an immunosuppressive drug serves as a good example.