

Progesterone and Hydroxyflutamide: Dose-response for Activation of the Androgen Receptor in HepG2 Cells

Rory B. Conolly
 CIIT Centers for Health Research
 6 Davis Drive
 RTP, NC 27709, USA
 rconolly@ciit.org

Kevin W. Gaido
 CIIT Centers for Health Research
 6 Davis Drive
 RTP, NC 27709, USA
 Gaido@ciit.org

ABSTRACT

In vitro characterization of the interactions of xenobiotics with steroid hormone receptors is part of the overall evaluation of potential for endocrine-mediated toxicity *in vivo*. Maness et al. (1998) used human hepatoma HepG2 cells transiently infected with the human androgen receptor and an androgen-sensitive luciferase reporter to characterize the dose-response behaviors of a series of compounds with the androgen receptor. The potential for antagonism was evaluated by conducting the dose-response assays in the presence of 10^{-7} M dihydrotestosterone, which causes maximal expression of the reporter system. Among the chemicals tested by Maness et al. were progesterone and hydroxyflutamide, both of which showed complex, U-shaped dose-response behaviors for androgen receptor activation in the presence of dihydrotestosterone (Table 1).

A simulation model of the steroid hormone receptor system was coded in MATLAB[®] to investigate potential mechanisms for the dose-response behaviors found by Maness et al. (1998). The model describes reversible, saturable and competitive interactions of two ligands (i.e., dihydrotestosterone and either progesterone or hydroxyflutamide) with a receptor. Ligand-receptor complexes in turn form homo- or heterodimers. The dimers compete for reversible binding to a response element. The rate of transcription of the reporter gene is a saturable (Michaelis-Menten) function of the occupancy of the response element by dimers. The model was able to produce qualitatively

accurate simulations of the data. Interestingly, adjustment of only one parameter, the dissociation constant for the binding of the xenobiotic homodimer with the response element, was sufficient to obtain a qualitatively accurate simulation of the data shown in Table 1. The significance of this result should not be over interpreted, however, since no effort was made at this point in the study to identify biologically realistic parameter values for the model. The point of this exercise was to evaluate the capability of the model to produce the dose-response shapes seen for hydroxyflutamide and progesterone and to thereby provide insight into the potential biological bases of the data.

Discussion and Conclusions

The data described by Maness et al. (1998) for progesterone and hydroxyflutamide are interesting in the context of human health risk assessment because they raise the possibility that the dose-response for endocrine mediated adverse health effects is not linear with dose. Since risk assessment policy assumes low dose linearity by default for receptor-mediated toxicants, demonstration of something other than low dose linearity for receptor-mediated gene transcription and association of the gene product with frank toxicity would challenge the validity of the default assumption. The present analysis suggests that a targeted program of experimental work combined with simulation modeling to properly evaluate this possibility would be worthwhile

REFERENCES

- [1] Maness, S.C., McDonnell, D.P., and Gaido, K.W. Inhibition of androgen receptor-dependent transcriptional activity by DDT isomers and methoxychlor in HepG2 human hepatoma cells. *Toxicol. Appl. Pharmacol.* 151, 135-142, 1998.

Table 1. Reporter gene activation by hydroxyflutamide and progesterone¹

Log dose (M)	% maximum response (hydroxyflutamide)	% maximum response (progesterone)
-8	85	85
-7.5	ND ²	85
-7	55	75
-6.5	38	ND
-6	20	65
-5.5	18	70
-5	30	90
-4.5	35	ND

¹Maness et al. (1998)

²No data