

Answer 1:

Bibliographic Information

In vivo bioluminescence imaging to evaluate estrogenic activities of endocrine disrupters. Pillon, Arnaud; Servant, Nadege; Vignon, Françoise; Balaguer, Patrick; Nicolas, Jean-Claude. *Endocrinologie Moléculaire et Cellulaire des Cancers*, INSERM Unite 540, UM I, Montpellier, Fr. *Analytical Biochemistry* (2005), 340(2), 295-302. Publisher: Elsevier, CODEN: ANBCA2 ISSN: 0003-2697. Journal written in English. CAN 142:477200 AN 2005:331970 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

Abstract

Reporter gene technol. is widely used to measure activity of hormone analogs, and bioluminescent in vitro assays have allowed rapid screening of numerous chems. either to identify new agonists or antagonists of hormones or to detect the presence of endocrine disrupters in the environment. Stable bioluminescent cell lines have been established and they provide reproducible dose-response curves and accurate detn. of in vitro efficiencies of various chems. In vivo, however, these mols. can be metabolized, bound by proteins, or stored in fats and thus could display efficiencies different from those obsd. in vitro. In vivo assays, such as the uterotrophic bioassay, require numerous sacrificed animals, and responses not only are dependent on an estrogenic action but also imply other factors. For a faster assay and to avoid the use of numerous animals, we developed an in vivo biosensor constituted of stable bioluminescent cells implanted in nude mice. MCF-7 bioluminescent cell lines were chosen since their proliferation is low in the absence of estrogen and the xenograft size can thus be stable for several weeks. Luciferase gene expression was monitored noninvasively with a cooled charge-coupled device camera. Quant. anal. allowed us to compare in vitro and in vivo actions of different estrogenic compds. (estradiol, estrone) and endocrine disruptors (ethynylestradiol, genistein, octylphenol, and 2,4'-dichlorodiphenyldichloroethylene) in the same cell lines and to follow hormone action on a living animal as a function of time. Different administration protocols have been used and good correlation was obsd. for most products. However, we found that ethynylestradiol was the most efficient chem. when orally administered.

Answer 2:

Bibliographic Information

Bioluminescent reporter cell lines to study the estrogenic activity of xenoestrogens in vitro and in vivo. Pillon, A.; Gomez, E.; Gauthier, P.; Escande, A.; Boussioux, A. M.; Duchesne, M. J.; Pelegrin, A.; Casellas, C.; Nicolas, J. C.; Balaguer, P. *Inserm U540 Endocrinologie moléculaire et cellulaire des cancers*, Montpellier, Fr. *Techniques, Sciences, Methodes: Genie Urbain--Genie Rural* (2004), (4), 93-101. Publisher: Association Scientifique et Technique pour l'Eau et l'Environnement, CODEN: TSMREA ISSN: 0299-7258. Journal written in French. CAN 141:167982 AN 2004:570846 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

Abstract

In order to characterize the estrogenic activity of chems. and biol. samples, we established bioluminescent cell lines starting from human breast cancer cells (MCF-7) and human uterus cervix cells (HeLa). MCF-7 cells, which naturally express estrogen receptor α (ER α) were transfected with estrogen regulated luciferase reporter gene (MELN cell line). HeLa cells, which do not express endogenous ER, were transfected with the same luciferase reporter gene (HELN cell line) and with a plasmid coding for human estrogen receptor (α or β) or mouse estrogen receptor (mER) or *Onchorhynchus mykiss* rainbow trout estrogen receptor (rtER) (resp. HELN hER α , HELN hER β , HELN mER or HELN rtER cell lines), in order to study ligands specificities of all these estrogen receptors. We used these cellular models as a tool to measure the activity of several potential xenoestrogens suspected of adversely affecting wildlife and human health. These cellular models also allowed us to detect estrogenic activity in water of urban sewage treatment plants. The estrogenic activity detected in water of these sites is about 0.5 nM estradiol equiv. and could be mainly due to human estrogens. Finally, these cells enabled us to develop a bioluminescent in vivo test for estrogens. We established xenografts by implanting MELN cells in male nude mice to take into account accumulation and metab. of xenoestrogens. These cells, which express luciferase after estrogenic stimulation, form tumors which are stable for at least two months. This model will enable us to study the in vivo effect of various estrogenic compds.