Endocrine Disruption

Androgen Receptor Binding Assay using Rat Prostate Cytosol

Background Information

- The Androgen Receptor Binding assay identifies chemicals that have the potential to interact with the androgen receptor (AR) \textit{in vitro} using AR isolated from rat ventral prostates.
- There is a high degree of DNA sequence conservation in the AR across mammalian phylogenetic lines, therefore, substances that bind the AR from rats are presumed to be capable of binding the AR in humans.
- The Androgen Receptor Binding assay measures the receptor-binding affinity of chemicals by evaluating their ability to displace a bound reference androgen, typically radiolabelled R1881, a synthetic androgen.
- Such interference with normal androgen binding has the potential to interfere (i.e., compete) with normal androgen activity \textit{in vivo} by acting as either an agonist which can produce androgen-like effects, or as an antagonist, which prevents or blocks the normal actions of androgens. Although the assay identifies compounds that compete for AR binding in vitro, it cannot distinguish between agonist and antagonist activity.
- Cyprotex follow the EPA OPPTS 890.1150 guideline\textsuperscript{1} for the androgen receptor binding assay using rat prostate cytosol and the assay can be performed under GLP or non-GLP conditions.

Guidance Protocol
EPA OPPTS 890.1150

Test System
Rat prostate cytosol

Test Article Exposure Concentrations
8 concentrations ranging from \(10^{-10}\) to \(10^{-3}\) M or up to limit of solubility

Exposure Time
24 hours

Number of Replicates
3 replicates per exposure concentration

Typical Vehicle Controls
< 10% DMSO, ethanol, water

Reference Controls
Strong binding: R1881 (metribolone)
Weak binding: dexamethasone

Test Article Requirements
Solid, water and liquids
Alternative formulations available on request

Analysis Method
Solubility by visual assessment
Displacement of \(^{3}H\) R1881

Report
Final Report
Completed Data Entry Spreadsheet (DEST)
Completed Data Evaluation Report (DER) upon request
OSRI assistance upon request

Proficiency Data Report
Available upon request

\textsuperscript{1}Luccio-Camelo DC and Prins GS (2011) J Steroid Biochem Mol Biol 127(1-2), 74–82

Animal models and epidemiological evidence link exposure to androgen disrupting chemicals with reduced sperm counts, increased infertility, testicular dysgenesis syndrome, and testicular and prostate cancers.
‘Epidemiological data show increases in incidence and prevalence of diseases associated with endocrine-disrupting chemicals, such as breast, prostate, and testis cancer, diabetes, obesity, and decreased fertility over the last 50 years.’

**Figure 1**
Representative graph showing potent positive reference control, R1881 (3 replicates/exposure as dark blue circles) and weak positive reference control, dexamethasone (3 replicates/exposure as light blue circles) in the Androgen Receptor Binding Assay using Rat Prostate Cytosol.

Cyprotex and its partners are able to offer the full range of EDSP series 890 protocols. A number of general endocrine disruption screening services are also available.

### Related Services

**EDSP Series 890**
- OPPTS 890.1200: Aromatase (human recombinant)
- OPPTS 890.1250: Estrogen receptor binding using rat uterine cytosol
- OPPTS 890.1300: Estrogen receptor transcriptional activation (human cell line HeLa-9903)
- OPPTS 890.1550: Steroidogenesis using human cell line H295R

**Screening Services**
- Estrogen Receptor Transactivation (ER-TA)
- Androgen Receptor Transactivation (AR-TA)
- Estrogen Receptor alpha (ER-α) binding
- Estrogen Receptor beta (ER-β) binding
- Androgen Receptor (AR) Binding
- Steroidogenesis

### References
1. OPPTS 890.1150 Androgen Receptor Binding (Rat Prostate Cytosol)