

22 December 2011

Chemical	CASRN	STTA ER TA ¹			BG1Luc ER TA ²		Data Source For Classification ⁴			Chemical Class ⁵	Product Class ⁶
		ER TA Activity	PC ₁₀ Value (M)	PC ₅₀ Value ^b (M)	ER TA Activity	EC ₅₀ Value ^{b,3} (M)	Other ER TAs ^c	ER Binding	Uterotrophic		
Methyl testosterone ^a	58-18-4	POS	1.73×10^{-7}	4.11×10^{-6}	POS	2.68×10^{-6}	POS(5/6)	POS	NT	Steroid	Pharmaceutical, Veterinary Agent
Morin	480-16-0	POS	5.43×10^{-7}	4.16×10^{-6}	POS	2.37×10^{-6}	POS(2/2)	POS	NT	Flavonoid, Phenol	Natural product
Norethynodrel ^a	68-23-5	POS	1.11×10^{-11}	1.50×10^{-9}	POS	9.39×10^{-10}	POS(5/5)	POS	NT	Steroid	Pharmaceutical, Veterinary Agent
<i>p,p'</i> -Methoxychlor ^a	72-43-5	POS	1.23×10^{-6}	(no PC ₅₀) ^b	POS	1.92×10^{-6}	POS(24/27)	POS	POS	Hydrocarbon (Halogenated)	Pesticide, Veterinary Agent
Phenobarbital ^a	57-30-7	NEG	-	-	NEG	-	NEG(2/2)	NEG	NT		Pharmaceutical, Analgesic
Reserpine	50-55-5	NEG	-	-	NEG	-	NEG(4/4)	NEG	NT	Heterocyclic Compound, Indole	Pharmaceutical, Veterinary Agent
Spirolactone ^a	52-01-7	NEG	-	-	NEG	-	NEG(4/4)	NEG	NT	Lactone, Steroid	Pharmaceutical
Testosterone	58-22-0	POS	2.82×10^{-8}	9.78×10^{-6}	POS	1.75×10^{-5}	POS(5/10)	POS	NT	Steroid	Natural Hormone

Abbreviations: CASRN = Chemical Abstracts Service Registry Number; M = molar; EC₅₀ = half maximal effective concentration of test chemical; NEG = negative; POS = positive; PC₁₀ (and PC₅₀) = the concentration of a test chemical at which the response is 10% (or 50 % for PC₅₀) of the response induced by the positive control (E2, 1nM) in each plate.

^aCommon chemicals tested in the STTA ER TA and BG1Luc ER TA that were designated as definitive ER Agonists or negatives and used to evaluate accuracy in the BG1 Luc ER TA validation study (ICCVAM BG1Luc ER TA Evaluation Report, Table 4-1 [6].

^bMaximum concentration tested in the absence of limitations due to cytotoxicity or insolubility was 1×10^{-5} M (STTA ER TA) and 1×10^{-3} M (BG1Luc ER TA).

^cNumber in parenthesis represents the test results classified as positive (POS) or negative (NEG) over the total number of referenced studies.

¹Values reported in Draft Report of Pre-validation and Inter-laboratory Validation For Stably Transfected Transcriptional Activation (TA) Assay to Detect Estrogenic Activity - The Human Estrogen Receptor Alpha Mediated Reporter Gene Assay Using hER-HeLa-9903 Cell Line [9].

²ICCVAM Test Method Evaluation Report on the LUMI-CELL[®] ER (BG1Luc ER TA) Test Method: An *In Vitro* Method for Identifying ER Agonists and Antagonists [6].

³Mean EC₅₀ values were calculated with values reported by the laboratories of the BG1Luc ER TA validation study (XDS, ECVAM, and Hiyoshi) [6].

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⁴Classification as an ER agonist or negative was based upon information in the ICCVAM Background Review Documents (BRD) for ER Binding and TA test methods (31) as well as information obtained from publications published and reviewed after the completion of the ICCVAM BRDs [6, 9, 10, 11, 12, 13, 14, 15].

⁵Substances were assigned into one or more chemical classes using the U.S. National Library of Medicine's Medical Subject Headings (MeSH), an internationally recognized standardized classification scheme (available at: <http://www.nlm.nih.gov/mesh>).

⁶Substances were assigned into one or more product classes using the U.S. National Library of Medicine's Hazardous Substances Database (available at: <http://toxnet.nlm.nih.gov/cgi-bin/sis/htmlgen?HSDB>)

Table 2: Chemicals tested in the STTA ER TA Validation Study [9].

Chemical name	CASRN	STTA ER TA Classification ²			
		PC ₁₀ (M)	PC ₅₀ (M)	PC ₁₀	PC ₅₀
17β-Estradiol	50-28-2	<1.00 × 10 ⁻¹¹	<1.00 × 10 ⁻¹¹	P	P
17β-Trenbolone	10161-33-8	1.78E-08	2.73E-07	P	P
17α-Estradiol	57-91-0	7.24E-11	6.44E-10	P	P
17α-Ethinyl estradiol	57-63-6	<1.00 × 10 ⁻¹¹	<1.00 × 10 ⁻¹¹	P	P
2-sec-Butylphenol	89-72-5	-	-	N	N
4-Cumylphenol	599-64-4	1.49E-07	1.60E-06	P	P
4-tert-Octylphenol	140-66-9	1.85E-09	7.37E-08	P	P
Apigenin	520-36-5	1.31E-07	5.71E-07	P	P
Bisphenol A	80-05-7	2.02E-08	2.94E-07	P	P
Bisphenol B	77-40-7	2.36E-08	2.11E-07	P	P
Butylbenzyl phthalate	85-68-7	1.14E-06	4.11E-06	P	P
Coumestrol	479-13-0	1.23E-09	2.00E-08	P	P
Daidzein	486-66-8	1.76E-08	1.51E-07	P	P
Di-n-butyl phthalate	84-74-2	-	-	N	N
Diethylstilbestrol	56-53-1	<1.00 × 10 ⁻¹¹	2.04E-11	P	P
Estrone	53-16-7	3.02E-11	5.88E-10	P	P
Ethyl paraben	120-47-8	5.00E-06	(no PC50)	P	N
Flavone	525-82-6	2.26 × 10 ⁻⁶	-	P	N
Genistein	446-72-0	2.24E-09	2.45E-08	P	P
Kaempferol	520-18-3	1.36E-07	1.21E-06	P	P
Kepone (Chlordecone)	143-50-0	7.11E-07	7.68E-06	P	P
meso-Hexestrol	84-16-2	<1.00 × 10 ⁻¹¹	2.75 × 10 ⁻¹¹	P	P
Methyl testosterone	58-18-4	1.73E-07	4.11E-06	P	P
Morin	480-16-0	5.43E-07	4.16E-06	P	P
Norethynodrel	68-23-5	1.11E-10	1.50E-09	P	P
p-n-Nonylphenol	104-40-5	-	-	N	N
p,p'-Methoxychlor	72-43-5	1.23E-06	(no PC50)	P	N
Phenolphthalin	81-90-3	-	-	N	N
Progesterone	57-83-0	-	-	N	N
Testosterone	58-22-0	2.82E-08	9.78E-06	P	P
2,4,5-Trichloro-phenoxyacetic acid	93-76-5	-	-	N	N
Atrazine	1912-24-9	-	-	N	N
Corticosterone	50-22-6	-	-	N	N
Cyproterone acetate	427-51-0	-	-	N	N
Haloperidol	52-86-8	-	-	N	N
Ketoconazole	65277-42-1	-	-	N	N
L-Thyroxine	51-48-9	1.32E-06	(no PC50)	P	N

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Chemical name	CASRN	STTA ER TA Classification ²			
		PC ₁₀ (M)	PC ₅₀ (M)	PC ₁₀	PC ₅₀
Mifepristone	84371-65-3	-	-	N	N
Phenobarbital (Na salt)	57-30-7	-	-	N	N
Reserpine	50-55-5	-	-	N	N
Spironolactone	52-01-7	-	-	N	N
Vinclozolin	50471-44-8	1.33E-07	7.65E-06	P	P
Zearalenone	17924-92-4	2.44E-11	6.44E-10	P	P
Tamoxifen	10540-29-1	1.49E-07	(no PC50)	P	N
Diethylhexyl phthalate	117-81-7	-	-	N	N
4-Androstenedione	63-05-8	2.56E-07	(no PC50)	P	N
Flutamide	13311-84-7	-	-	N	N
Procymidone	32809-16-8	-	-	N	N
Clomiphene citrate	50-41-9	3.68E-08	(no PC50)	P	N

Abbreviations: Abbreviations: CASRN = Chemical Abstracts Service Registry Number; EC₅₀ – half maximal effective concentration; NEG = negative; PC₁₀ (and PC₅₀) = the concentration of a test chemical at which the response is 10% (or 50% for PC₅₀) of that induced by the positive controls (E2, 1nM); POS = positive.

^jThe classification for this substance is “presumed positive” for ER agonism since the substance was positive in 50% or less of reported studies, or was reported positive in the single study conducted.

¹ICCVAM, 2003. Evaluation of in vitro test methods for detecting potential endocrine disruptors: estrogen receptor and androgen receptor binding and transcriptional activation assays. NIH Publication No. 03-4503. Available at: [<http://iccvam.niehs.nih.gov/methods/endodocs/edfinrpt/edfinrpt.pdf>], Addendum, 2006: [10].

²Takeyoshi, M, Draft Report of Pre-validation and Inter-laboratory validation for stably transfected activation (TA) assay to detect estrogenic activity; The human estrogen receptor alpha mediated reporter gene assay using hER-HeLA-9902 cell line. 2006 [9].

Chemicals Evaluation and Research Institute (CERI): Japan, p. 1-188.

Table 3. Chemicals Tested during the BG1Luc ER TA Validation Study [6].

Substance	CASRN	BG1Luc ER TA Classification ¹
17 β -Estradiol	50-28-2	POS
17 β -Trenbolone	10161-33-8	POS
17 α -Estradiol	57-91-0	POS
17 α -Ethinyl estradiol	57-63-6	POS
19-Nortestosterone	434-22-0	POS
2-sec-Butylphenol	89-72-5	POS
4-Cumylphenol	599-64-4	POS
4-Hydroxy-androstenedione	566-48-3	POS
4-tert-Octylphenol	140-66-9	POS
Apigenin	520-36-5	POS
Bisphenol A	80-05-7	POS
Bisphenol B	77-40-7	POS
Butylbenzyl phthalate	85-68-7	POS
Chrysin	480-40-0	POS
Coumestrol	479-13-0	POS
Daidzein	486-66-8	POS
Di- <i>n</i> -butyl phthalate	84-74-2	POS
Dicofol	115-32-2	POS
Diethylstilbestrol	56-53-1	POS
Estrone	53-16-7	POS
Ethyl paraben	120-47-8	POS
Fenarimol	60168-88-9	POS
Flavone	525-82-6	POS
Fluoranthene	206-44-0	POS
Fluoxymestrone	76-43-7	POS
Genistein	446-72-0	POS
Kaempferol	520-18-3	POS
Kepone	143-50-0	POS
meso-Hexestrol	84-16-2	POS
Methyl testosterone	58-18-4	POS
Morin	480-16-0	POS
Nilutamide	63612-50-0	POS
Norethnodrel	68-23-5	POS
<i>o,p'</i> -DDT	789-02-6	POS
<i>p-n</i> -Nonylphenol	104-40-5	POS
<i>p,p'</i> -Methoxychlor	72-43-5	POS
Phenolphthalin	81-90-3	POS
Progesterone	57-83-0	POS
Testosterone	58-22-0	POS

Substance	CASRN	BG1Luc ER TA Classification ¹
12- <i>O</i> -Tetradecanoylphorbol-13-acetate	16561-29-8	NEG
2,4,5-Trichloro-phenoxyacetic acid	93-76-5	NEG
4-Hydroxytamoxifen	68047-06-3	NEG
Actinomycin D	50-76-0	NEG
Ammonium perchlorate	7790-98-9	NEG
Apomorphine	58-00-4	NEG
Atrazine	1912-24-9	NEG
Bicalutamide	90357-06-5	NEG
Corticosterone	50-22-6	NEG
Cyproterone acetate	427-51-0	NEG
Dibenzo[<i>a,h</i>] Anthracene	53-70-3	NEG
Finasteride	98319-26-7	NEG
Haloperidol	52-86-8	NEG
Hydroxy flutamide	52806-53-8	NEG
Ketoconazole	65277-42-1	NEG
L-Thyroxine	51-48-9	NEG
Linuron	330-55-2	NEG
Medroxyprogesterone acetate	71-58-9	NEG
Mifepristone	84371-65-3	NEG
Phenobarbital	50-06-6	NEG
Pimozide	2062-78-4	NEG
Propylthiouracil	51-52-5	NEG
Raloxifene HCl	82640-04-8	NEG
Reserpine	50-55-5	NEG
Sodium azide	26628-22-8	NEG
Spirolactone	52-01-7	NEG
Vinclozolin	50471-44-8	NEG

Table is sorted by classification and then alphabetically by substance name.

¹ICCVAM Test Method Evaluation Report on the LUMI-CELL[®] ER (BG1Luc ER TA) Test Method An In Vitro Method for Identifying ER Agonists and Antagonists [6].

Table 4. Summary of the Reliability and Accuracy Values Obtained for the STTA and BG1Luc TAs: Validation Studies.

Place holder: Data to be added for Intra- and Intra-laboratory reproducibility, Sensitivity, Specificity, Positive and Negative predictivity, and Overall Accuracy values from respective validation studies.

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