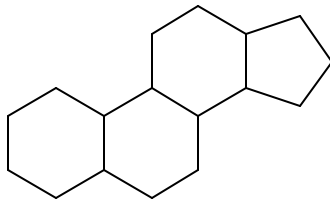


Endocrine disrupting compounds

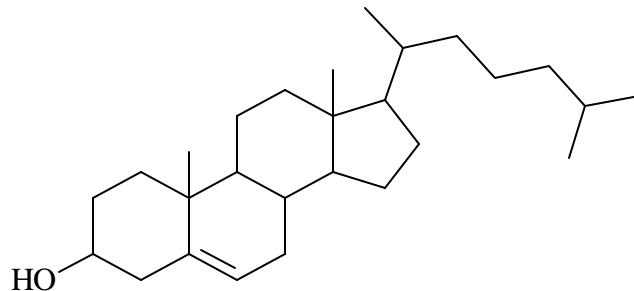
- substances that interfere with hormone action
- estrogen mimics the most widely studied to date:
androgens and thyroid hormone mimics becoming a research focus
- concerns about:
 - sewage works effluents
 - pulp and paper effluents
 - industrial spills
 - natural compounds
- endocrine disrupters interfere with normal hormone action: they can either
 - act as hormone agonists, or
 - antagonize (block) normal hormone action
- a key issue is that hormones act at minuscule concentrations: nM and less. Even weak hormone mimics can show biological activity at μM and less

Steroid hormones

- biosynthesized in vertebrates from cholesterol

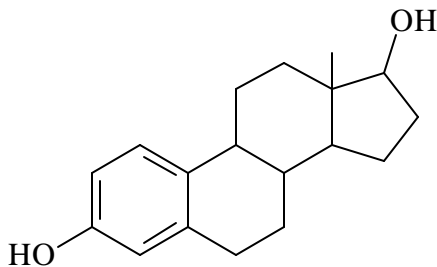


steroid skeleton

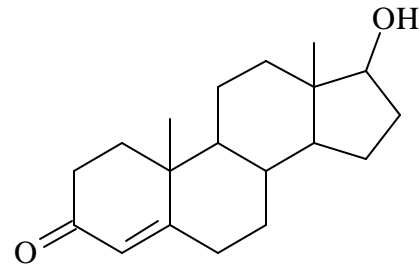


cholesterol

- the most important sex hormones

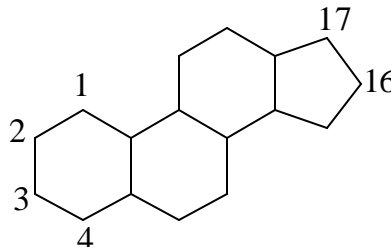


estradiol



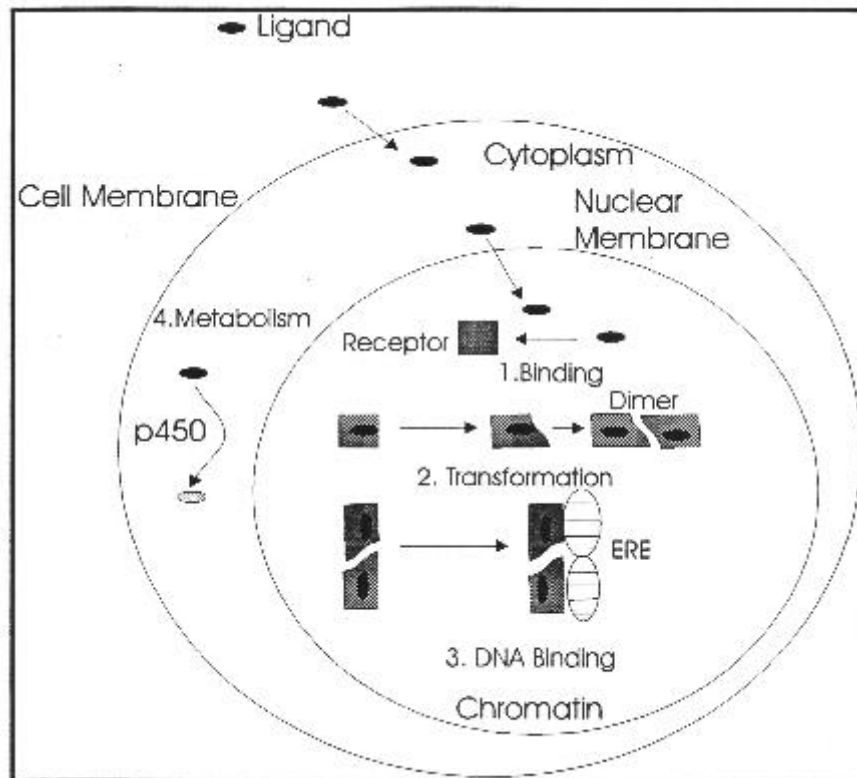
testosterone

- testosterone is an intermediate in the formation of estradiol (aromatase)
- both males and females produce both estrogens and androgens
- these hormones have important roles in development, including fetal development
- “used” steroid hormones are metabolized by cytochrome P-450 enzymes: estradiol \rightarrow 17 ketone (estrone), to 16-hydroxylation (estriol), and 2- and 4-hydroxylation



Mechanism of action of steroid hormones – estradiol

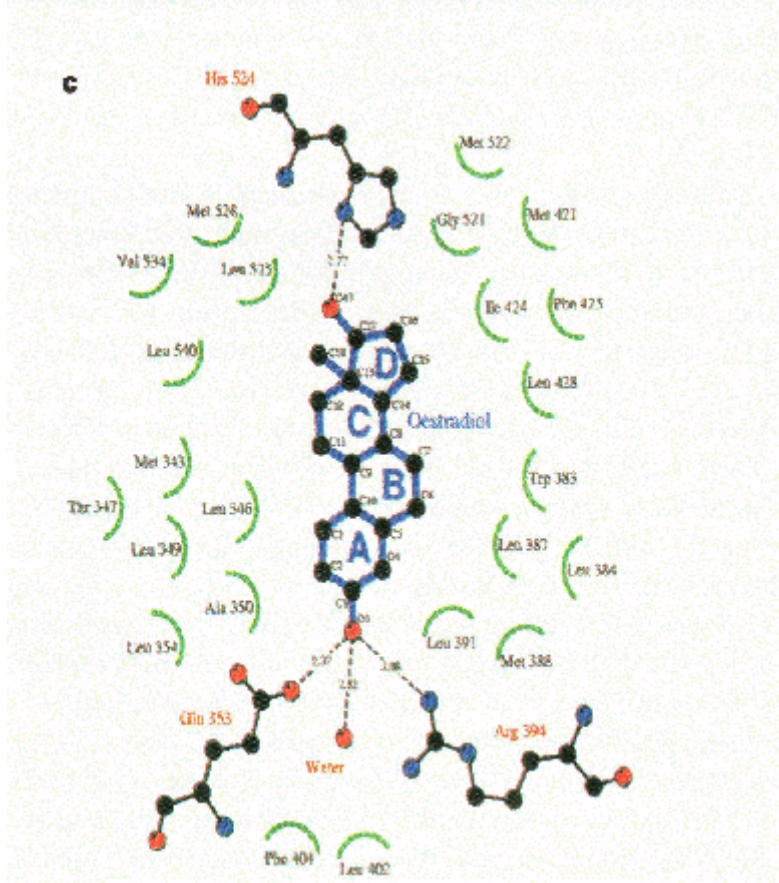
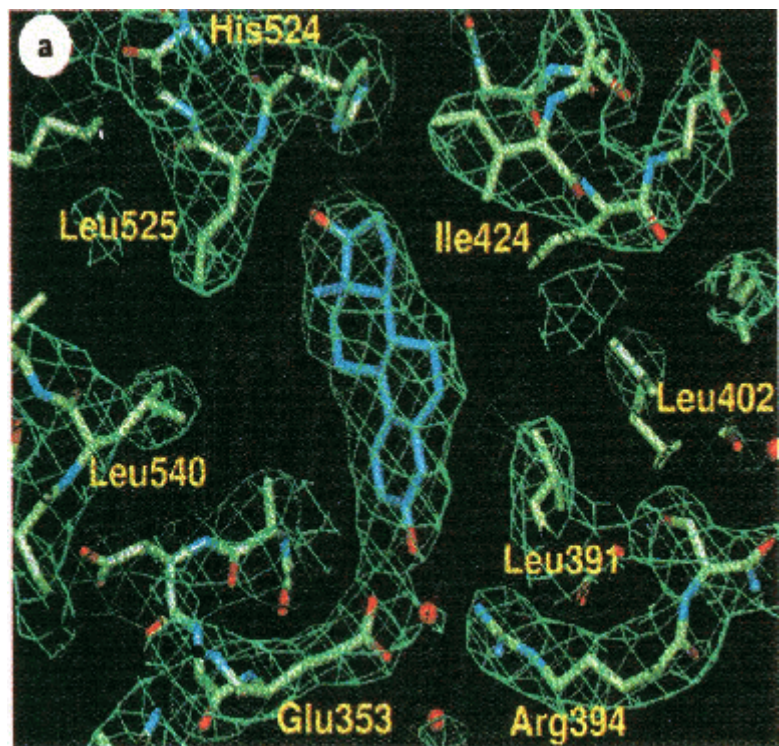
- involves binding of estradiol to a *steroid hormone receptor* – note similarity to mechanism of action of dioxins



B.J. Cox, MSc Thesis

- estrogen receptor found in females especially in liver and ovaries: two forms, ER- α and ER- β .
- estrogen mimics bind to the ER, either leading to estrogenic effects (agonist) or blocking them (antagonist)
- to be an estrogen mimic, the molecule must:
 - be the correct size and shape
 - normally have polar groups (especially –OH) located to hydrogen bond with amino acids in the active site

The ER Binding Domain

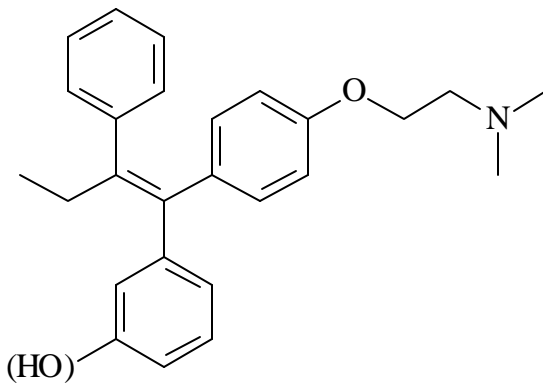


Brzozowski, Nature, 389, 753 (1997)

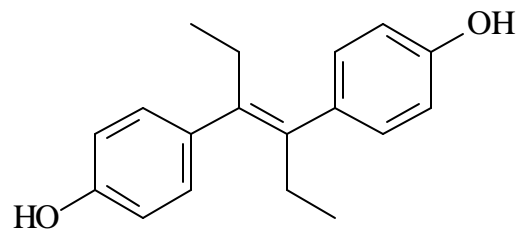
ER Binding

- ER binding site described as “catholic”: other ligands make contacts with different aminoacids in the binding domain
- Presence of at least one –OH group is normally important
- Relative binding affinities, rat liver ER- α vs 17 β -estradiol = 1

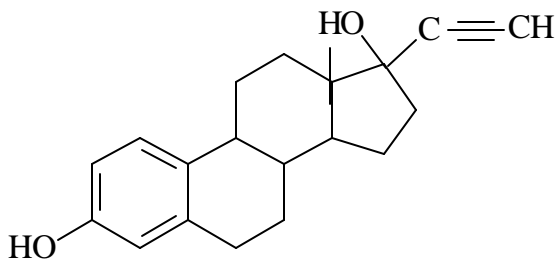
Tamoxifen	3.3	17 α -Ethynelestrodiol	2.0
Diethylstilbestrol	0.9	Estradiol-3-benzoate	0.4
α -Zearolenone	0.11	Estrone	0.08
Estriol	0.05	Tetrachlorobiphenylol	0.004
<i>o,p</i> -DDT	0.0001		



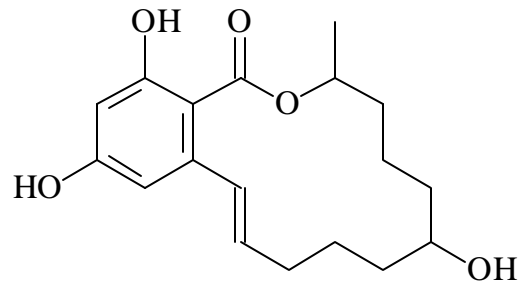
(hydroxy)-tamoxifen



diethylstilbestrol

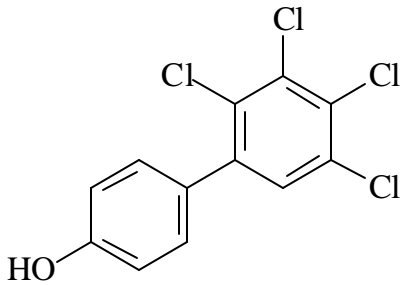


ethynelestrodiol

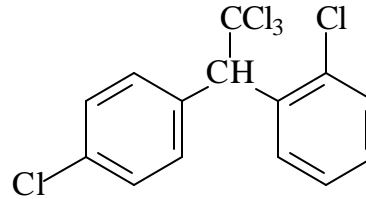


zearolenone

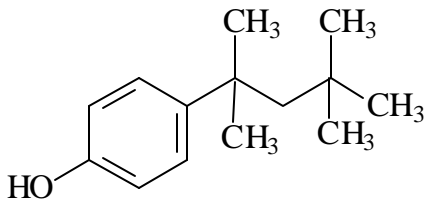
Other estrogenic compounds



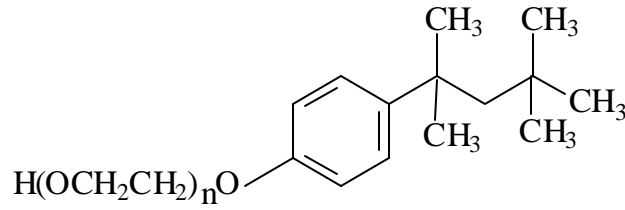
tetrachlorobiphenylol



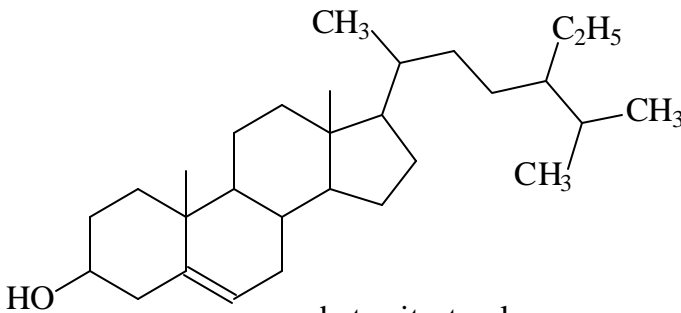
o,p-DDT



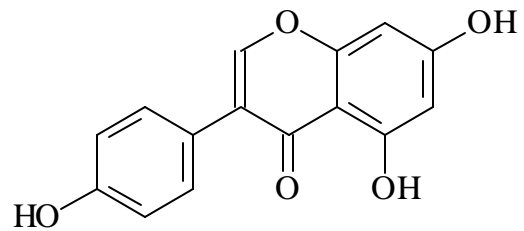
octylphenol



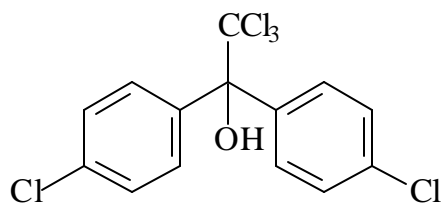
an octylphenoxyethoxylate [detergent]



beta-sitosterol



genistein (in clover and soy)



difocol

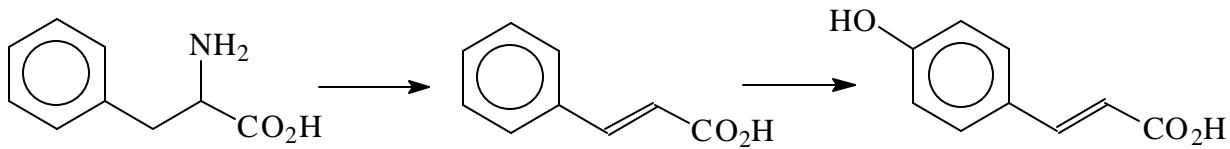
- application of the TEF concept to xenoestrogens

$$\text{TEQ} = \sum(c_i \times \text{TEF}_i) \quad \text{where the TEF of estradiol} = 1$$

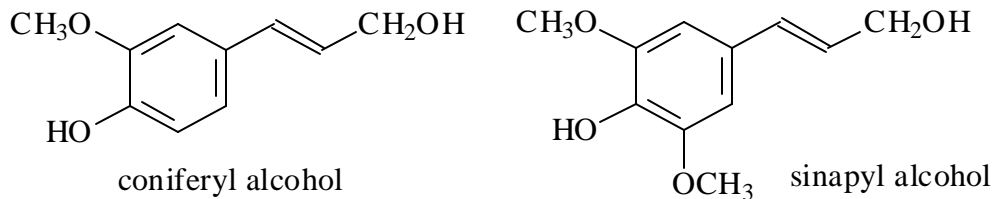
- relative binding affinities (RBAs) sometimes taken as a surrogate for TEF: *why are they not the same?*

The pulp and paper industry

- wood pulp contains both cellulose fibers (—> paper) and lignin (extracted and discarded). Lignin is polymeric – biosynthesized from phenylalanine CYP 73, a cinnamate-4-hydroxylase, catalyzes a key step in the conversion of phenylalanine into lignin.

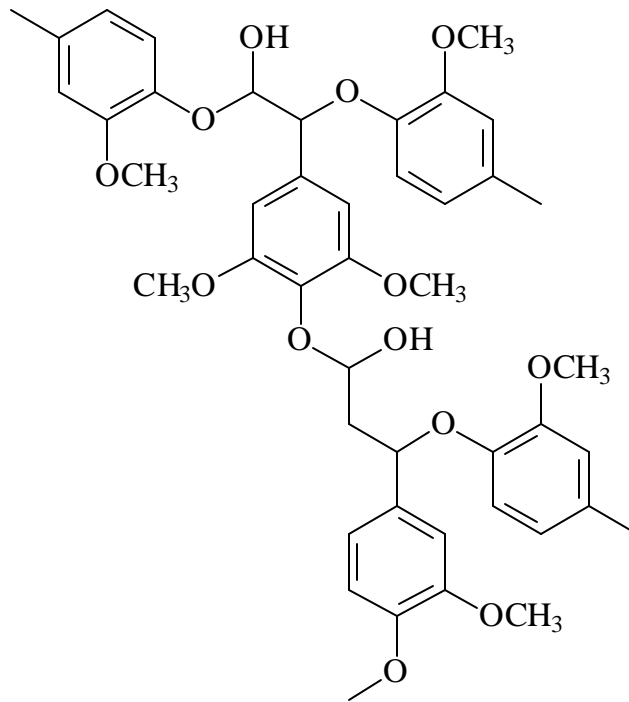


- hardwoods contain ~25% lignin; softwoods ~30% lignin
- lignin building blocks



- lignin formed by polymerization catalyzed by peroxidases and laccases in the plant —> free radical processes
- controversy over whether lignin is a directed structure or a random structure
- paper-making requires removal of the lignin: delignification: Kraft pulping; sulfite pulping; thermomechanical pulping

Partial lignin structure



Separated pulp is **bleached** to make fine paper

- bleaching with chlorine → traces of dioxins (a concern in the late 1980s)
- bleaching with chlorine dioxide
- bleaching with hydrogen peroxide

- endocrine effects down stream of pulp mills are similar for bleached and unbleached pulp → a natural component of the wood. Most likely candidate is β -sitosterol

