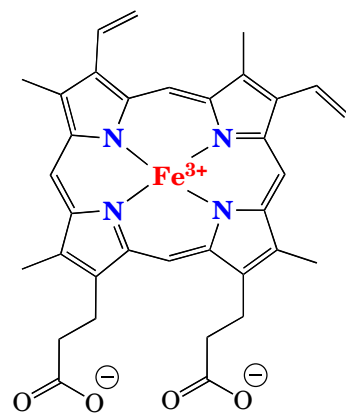


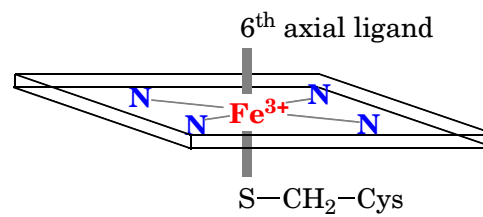
Aromatase

(Turning men into women for millions of
years. . . .)

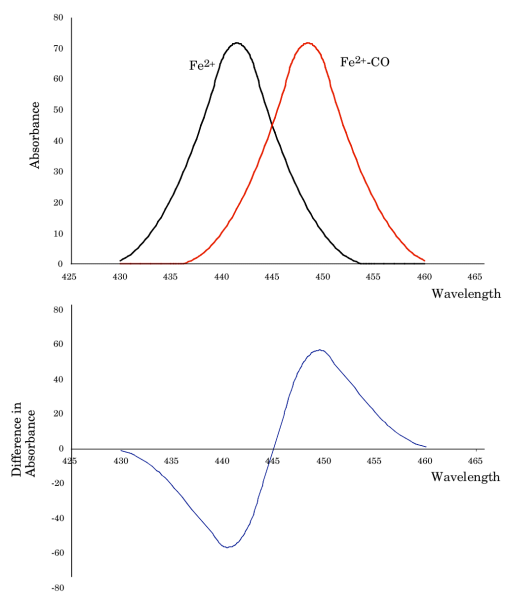
Cytochromes P450



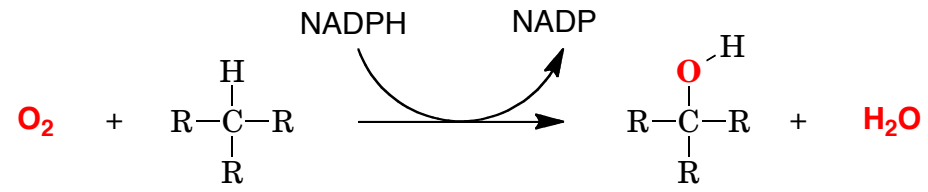
Iron-Protoporphyrin IX



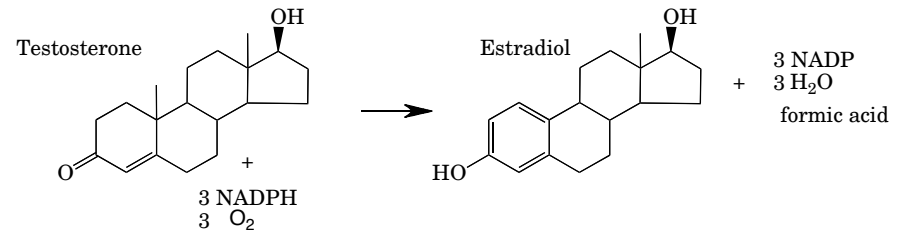
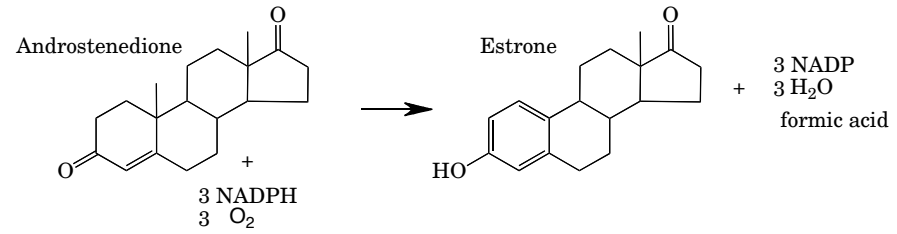
Difference Spectrum



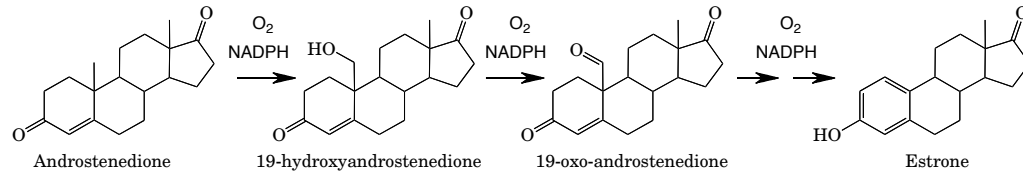
Cytochrome P450



Aromatase



Aromatase



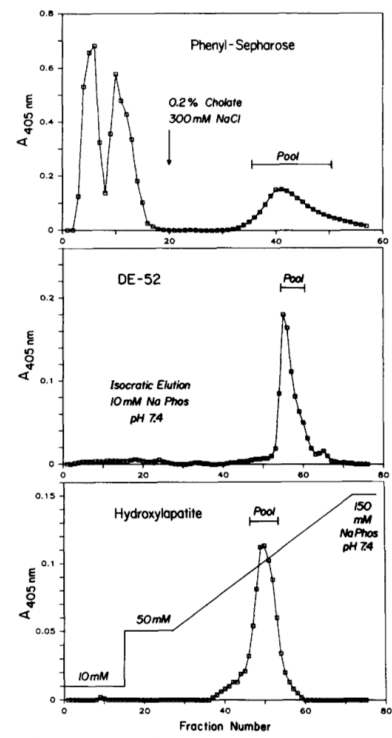


FIG. 2. Elution profiles of the three column chromatographic steps used in the purification of P-450*arom*. Fraction volumes: phenyl-Sepharose, 11 ml; DE52, 7 ml; hydroxylapatite, 4 ml.

TABLE I
Purification of human placental aromatase cytochrome P-450

Purification step	Total protein	Total activity ^a	Specific activity	Total P-450 ^{arom} ^b	Specific content	P-450 ^{arom} yield
	mg	nmol/min	nmol/min/mg protein	nmol	nmol/mg protein	%
Microsomes	4958	915	0.18	255	0.05	100
Cholate extract	3796	626	0.16	251	0.07	98
Ammonium sulfate, 35-55%	934	349	0.37	156	0.17	61
Phenyl-Sepharose	125	304	2.4	107	0.86	42
DE52	7.6	152	20	39	5.1	15
Hydroxylapatite	1.95	111	57	22	11.5	9

^a Samples were reconstituted with 100 nM rabbit liver P-450 reductase and 0.003% Nonidet P-40 as described under "Experimental Procedures."

^b P-450^{arom} concentration was determined by difference spectra induced by (19R)-10-thiiranyl-4-estrene-3,17-dione as described under "Experimental Procedures."

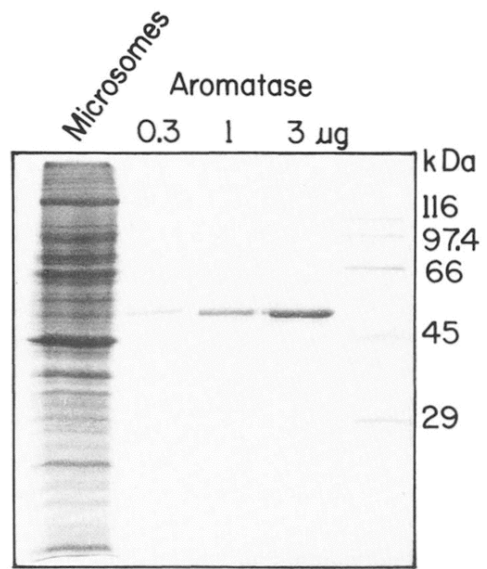


FIG. 3. **SDS-polyacrylamide gel electrophoresis of aromatase.** The lanes show human placental microsomes (200 μg of protein), 0.3, 1, and 3 μg of purified P-450 $arom$ and molecular weight markers.

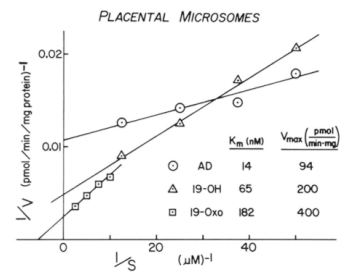
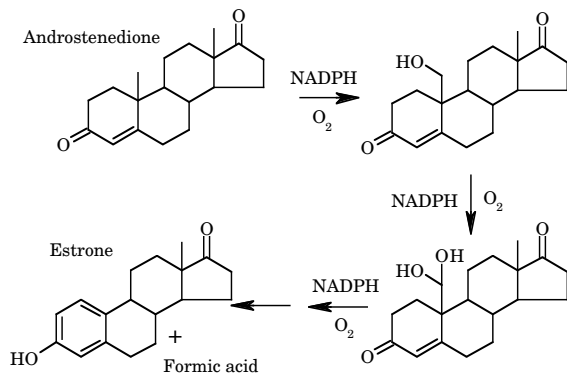


FIG. 8 Kinetic analysis of aromatization using human placental microsomes. Substrates: androstenedione (AD), 19-hydroxyandrostenedione (19-OH), and 19-oxoandrostenedione (19-Oxo). Determination of product formation was performed by radioimmunoassay for estrone.

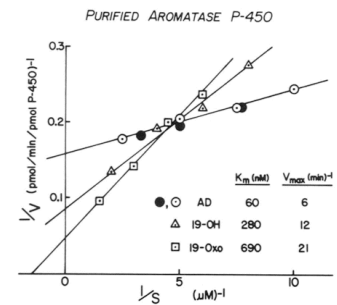


FIG. 9 Kinetic analysis of aromatization using purified, reconstituted P-450arom. Substrates: androstenedione (AD), 19-hydroxyandrostenedione (19-OH), and 19-oxoandrostenedione (19-Oxo). Open symbols indicate that determination of product formation was by radioimmunoassay for estrone. Closed symbols indicate a separate experiment in which AD aromatization was measured by tritium release from [1,2,3-³H]androstenedione.

Aromatase Inhibitors

